Gain-of-Function Deliberative Process
Written Public Comments
Oct. 19, 2014 - Jun. 8, 2016

The following are written comments submitted to the National Science Advisory Board for Biosecurity (NSABB) and/or U.S. Government for the period October 19, 2014 – June 8, 2016.

Interested persons may file written comments with the Board at any time via an email sent to nsabb@od.nih.gov. Written statements should include the name, contact information, and when applicable, the professional affiliation of the interested person.

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October 19, 2014

MEMORANDUM TO: NSABB

FROM: RADM Kenneth W. Bernard, MD, USPHS (ret.)
Former Special Assistant to the President
for Biodefense (2002-2005)

SUBJECT: NSABB: A bit of historical context

In 2003, after reading the Institute of Medicine (IOM) Fink Committee report on dual use research of concern (DURC), I wrote the first NSABB Charter (with significant input from Bob Kadlec, Rajeev Venkayya, Curt Mann, Larry Kerr and John LaMontagne, among others). The White House Homeland Security Council created the NSABB in 2004 to ensure that the Fink Report's recommendations were addressed by USG policy to encourage and improve security without hobbling good science. In fact, we used the successful 1974-75 NIH Recombinant DNA Advisory Committee (RAC) as a model. The White House wanted to avoid having the Fink Report recommendations used, *de facto*, as hard "red-lines" that restricted whole categories of research out of fear, without adequate debate or discussion.

Our original intent for the NSABB was to set up a process for *institutional* level discussion of proposed projects that might be considered DURC (such as "gain of function") before starting the research, not after it was completed. The NSABB was also to be a national resource for DURC related issues, and, as such, act in an
expert advisory role not only to the USG, but also to IBC-like committees at research institutions that would be reviewing specific research proposals for potential DURC (now called "institutional review entities" (IREs)). The NSABB was to have been asked for expert advice as needed — not provide primary research approval.

Of interest, the following is (a partial) list of the NSABB functional activities approved on January 21, 2004 by the White House Homeland Security Council in the original Charter.

"The NSABB will perform the following activities (*inter alia*):

- Advise on national policies governing local review and approval processes for dual-use biological research, including the development of guidelines for the case-by-case review and approval by Institutional Biosafety Committees (IBCs).
- Advise on criteria and processes for referral of classes of research or specific experiments by IBCs to the NSABB for guidance.
- Review and provide guidance on specific experiments insofar as they exemplify a significant or particularly complex permutation of an existing category of dual-use research, or represent a novel category of dual-use research that requires additional guidance from the NSABB.
- Respond to requests submitted by research institutions for the interpretation and application of the guidelines to specific research proposals in instances where a proposal has been denied by an IBC and the institution seeks additional advice."

Over time, many of these explicit activities were "delayed," and the current 2014 charter only includes a very vague and general list of NSABB duties, and no specific reference local institutional oversight.

Two years after its first meetings in 2005, the NSABB did publish a plan for institutional review for DURC, the excellent 2007 "Proposed Framework for the Oversight of Dual Use Life Sciences Research." Unfortunately, the framework remained "under review" for 5 more years.

Initial guidelines for government DURC oversight were finally adopted in 2012 – but only after the well-known influenza publication fiasco. Just last month —7 years after they were proposed, "*The United States Government Policy for Institutional Oversight of Life* Sciences *Dual Use Research of Concern*" was finally announced, but won't be implemented for yet another year.

Every stakeholder has a different reason for this unconscionable delay, mostly related to research community fears and questions, institutional and agency intransigence, and government process fouls. International science and security communities were ill served as a result.

Unfortunately, no one prioritized the institutional-level DURC oversight originally intended for NSABB until research on increased transmissibility of H5N1 influenza was done, and papers were accepted for publication at two prestigious journals. Then – far too late -- the NSABB was asked to opine. It was saddled with the impossible job of trying to adjudicate publication, rather than doing its intended job of advising IREs on whether, or how the research should be done in the first place. Predictably, and for obvious reasons, the NSABB failed in its newly assigned mission. Finally acknowledging a need for early institutional review research, the USG Institutional Oversight Policy for DURC was put on a fast track to approval.

I applaud your engaging in a discussion of "gain of function" research pressured, I presume, by the ongoing work on increased transmissibility of pandemic flu viruses. But while this discussion is a necessary component of the NSABB's work, it is not an end in itself. The NSABB's conclusions on this thorny subject can and should be framed so they can be applied by local IREs (and the Director of NIH and Secretary of HHS) *before* proposals are approved, funded, and undertaken — not after results are available and ready to publish.

The NSABB would best meet the DURC research and security needs of the United States (and internationally) if it adopted a "concept of operations" (CONOPS) that reasserts its original IRE advisory activities. The new Program on Biosecurity and Biosafety in the office of the Director at NIH should, therefore, implement and quickly operationalize a CONOPS for the new 2014 Institutional Oversight Policy as its single highest priority in the coming months.

CENTER FOR ARMS CONTROL AND NON-PROLIFERATION
BIOLOGICAL AND CHEMICAL WEAPONS CONTROL PROGRAM

Written comments in advance of the NSABB meeting on October 22, 2014

To: Contact Person: Carolyn Mosby, The National Science Advisory Board for Biosecurity Program Assistant (e-mail: carolyn.mosby@nih.gov)

From: The Scientists Working Group on Biological and Chemical Weapons,
Center for Arms Control and Non-proliferation, 322 4th St., NE | Washington, D.C. 20002 | 202.546.0795

Date: October 20, 2014

Dear NSABB Committee,

I am writing on behalf of The Scientists Working Group on Biological and Chemical Weapons at the Center for Arms Control and Non-proliferation. We fully support the *Cambridge Working Group Consensus Statement on the Creation of Potential Pandemic Pathogens (PPPs)* [http://www.cambridgeworkinggroup.org/].

Our grave concern is the risk of a pandemic from release (escape) of a potential pandemic pathogen (PPP) from a laboratory. Risk has two components, the likelihood of release and the consequences of release. While the likelihood of release from a single laboratory in a single year is small, likelihood of release from one of several laboratories over several years is intolerably high. A potential consequence of a release is a pandemic with millions of deaths. As it stands, there is no proactive oversight nor regulations for this PPP research, so any and all of the world's nations can carry out this dangerous work without regard to consequences. But consequences would be shared by all of us.

The Center's Scientists Working Group has published or issued several papers on the risk of PPP research and documented escapes of deadly pathogens in the past, which have led to considerable fatalities. As an integral part of this comment, the titles of our key papers are listed below along with web addresses to access the papers.

The Human Fatality and Economic Burden of a Man-made Influenza Pandemic: A Risk Assessment [http://armscontrolcenter.org/The_Human_Fatality_Burden_of_Gain_of_Function_Flu_Research_v1-5-14.pdf]

Laboratory Escapes and "Self-fulfilling prophecy" Epidemics [http://armscontrolcenter.org/Escaped_Viruses-final_2-17-14.pdf]

The consequences of a lab escape of a potential pandemic pathogen [http://journal.frontiersin.org/Journal/10.3389/fpubh.2014.00116/full]

Biological threats: A matter of balance [http://thebulletin.org/biological-threats-matter-balance]

The unacceptable risks of a man-made pandemic [http://www.thebulletin.org/unacceptable-risks-man-made-pandemic]

CENTER FOR ARMS CONTROL AND NON-PROLIFERATION
BIOLOGICAL AND CHEMICAL WEAPONS CONTROL PROGRAM

Threatened pandemics and laboratory escapes: Self-fulfilling prophecies
[http://thebulletin.org/threatened-pandemics-and-laboratory-escapes-self-fulfilling-prophecies7016]

Biological threats: A matter of balance
BY SCIENTISTS WORKING GROUP ON BIOLOGICAL AND CHEMICAL WEAPONS | 2 FEBRUARY 2010

Sincerely yours,

Lynn C, Klotz, PhD
Senior Science Fellow
Center for Arms Control and Non-proliferation, and
Member of the Scientists Working Group on Biological and Chemical Weapons

5 Duley Street
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Elizabeth Hart
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For the attention of:
Samuel L Stanley
Chair, National Science Advisory Board for Biosecurity (NSABB)

22 October 2014

An open letter to the NSABB challenging potentially dangerous 'gain of function' research

Dr Stanley

Re the upcoming NSABB meeting to discuss controversial 'gain of function' research, to be held on 22 October 2014.[1]

As a global citizen, I wish to register my opposition to lab-engineering of potentially lethal pathogens being sponsored by the United States Government and other parties.

There appears to be seriously inadequate ethical oversight of this dangerous research, which may be taking place around the world in an unknown number of facilities in universities, research laboratories and pharmaceutical companies.

It is most concerning that there is an arrogant attitude about this type of research in the scientific community, perhaps exemplified most tellingly in the comments of Vincent Racaniello, Professor of Microbiology and Immunology in the College of Physicians and Surgeons of Columbia University.

In an interview re controversial influenza H7N9 gain of function experiments[2], broadcast on Dispatch Radio in August 2013[3], Professor Racaniello stated:

> *So a 'gain of function'[4] simply means that you take a virus and you change it in some way* ***so it does something new, so it does something that it didn't do before****. That's all that means. It's quite simple.* ***So you could for example take this H7N9 virus and make it resistant to an anti-viral drug, that would be a gain of function...***
>
> *So, to really understand how this virus works, and really any other virus,* ***we do gain of function studies all the time. We don't make a big deal of it, we don't write letters telling the world that we're going to do them because that's not the way science works. Science works by just doing your experiments****. We do this because we would like to see what kinds of changes would lead to a gain of function, and what would be the consequences.*
>
> *So, in the case of this virus,* ***these investigators want to make the virus drug resistant****. As you know, there are a couple of anti-virals that you can use if you get influenza – Tamiflu, Relenza -* ***and these investigators want to make the virus resistant****.* ***And the reason they want to do that is to see if a drug resistant mutant would have any properties that would make it scarier in people.***
>
> *So there is really a goal to these experiments. They want to know if you change the virus what might be the consequences for people.* ***And as I said this is done all the time but these virologists decided to tell the world about it.***
>
> (My emphasis.) (Full transcript of interview attached to view comments in context.)

Professor Racaniello says ***"we do gain of function studies all the time. We don't make a big deal of it, we don't write letters telling the world that we're going to do them because that's not the way science works. Science works by just doing your experiments."*** Professor Racaniello seems to infer that it is acceptable for scientists to manipulate viruses, e.g. make a ***"virus drug resistant...to see if a drug resistant mutant would have any properties that would make it scarier in people"*** without telling ***"the world about it"***. (I challenged Professor Racaniello about his comments on his Virology blog post "Virologists plan influenza H7N9 gain of function experiments"[5], but he did not respond.)

I suggest Professor Racaniello's attitude is arrogant and irresponsible.

How many other scientists are undertaking this type of research *"without telling the world about it"*, and with scant regard for potentially disastrous consequences? For example, are scientists manipulating the ebola virus to ***"make it scarier in people"***?

As well as scientists manipulating potentially deadly pathogens with little or no effective ethical oversight, careless practices are also a serious problem in some laboratories. A CIDRAP report on lab biosafety refers to ***"recent incidents in which lab workers at the Centers for Disease Control and Prevention (CDC) inadvertently sent potentially viable Bacillus anthracis samples to a low-containment lab and shipped nonpathogenic avian flu virus samples contaminated with the deadly H5N1 virus to a US Department of Agriculture lab. Those mishaps were followed by the discovery of smallpox virus samples in a Food and Drug Administration facility"***.[6]

Following these significant lapses in biosafety and biosecurity at US Federal research facilities, The White House Office of Science and Technology Policy advises the US Government ***"has taken a number of steps to promote and enhance the Nation's biosafety and biosecurity, including immediate and longer term measures to review activities specifically related to the storage and handling of infectious agents"***.[7]

It has also been announced that there will be a pause in US Government funding of any new studies involving gain of function experiments with influenza, SARS, and MERS, and a 'deliberative process' is being launched to assess the potential risks and benefits associated with gain of function research.[8] The NSABB and the National Research Council (NRC) of the National Academies will be involved in this deliberative process, commencing with a NSABB meeting on 22 October 2014.[9]

Francis S Collins, Director of the National Institutes of Health, notes that ***"public involvement in this deliberative process is key, and the process is thus designed to be transparent, accessible, and open to input from all sources"***. Dr Collins encourages us to ***"follow these deliberations closely"***.[10]

I hope that Dr Collins is paying more than lip service to the notion that ***"public involvement in this deliberative process is key"***. **Sponsoring of potentially dangerous gain of function research, and mishandling of pathogens, are matters of serious concern for global citizens. Discussion on these matters should not be restricted to scientists and bureaucrats with possible conflicts of interest.**

In regards to 'public involvement' in this matter, I have sought to make submissions previously, ie:

- In January 2012 I forwarded an open letter to the NSABB re the political and ethical implications of lethal virus development to Dr Paul Keim, then Acting Chair of the NSABB, and Dr Michael Osterholm, then a member of the NSABB. Beyond acknowledgement of receipt of my letter, I received no further response to the important matters raised, e.g. **my suggestion that by sponsoring development of a potentially lethal flu virus, the United States could be in breach of the Biological Weapons Convention**. Please see attached my letter to Dr Keim for further background.

- In December 2012 I made a submission to the Centers for Disease Control and Prevention (CDC), Department of Health and Human Services (HHS) re my opposition to lab-engineering of potentially lethal pathogens, also attached.

I request that the recently revised NSABB membership consider this submission, and my previous submissions as detailed above and attached, in the deliberative process for gain of function research. I also question what processes are being put in place to allow interested parties such as myself to be kept abreast of developments on this matter, e.g. email updates?

Sincerely
Elizabeth Hart

Please note this letter will be forwarded to other parties for information, including the cc list below

cc:
- Francis S Collins, US National Institutes of Health
- Anthony Fauci, US National Institute of Allergy and Infectious Diseases
- Ralph J. Cicerone, US National Academy of Sciences

- Paul Keim, past Chair of the NSABB
- Michael Osterholm, past member of the NSABB
- Vincent Racaniello, Columbia University
- Ron Fouchier, Erasmus MC
- Ab Osterhaus, Erasmus MC
- Yoshihiro Kawaoka, University of Wisconsin-Madison
- Marc Lipsitch, Harvard School of Public Health
- Peter Palese, Mount Sinai School of Medicine
- Tom Jefferson, Cochrane Vaccines Field
- Peter Gøtzsche, The Nordic Cochrane Centre
- Lord Robert May, Oxford University
- Philip Campbell and Declan Butler, *Nature*
- Caroline Ash and Martin Enserink, *Science*
- Fiona Godlee and Deborah Cohen, *British Medical Journal*
- Brian Martin, University of Wollongong
- Bea Mies, Independent Vaccine Investigator
- Monika Peichl, Independent Vaccine Investigator
- Carolyn Mosby, National Institutes of Health

References: (Links and hyperlinks active as at 22 October 2014.)

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[1] Doing Diligence to Assess the Risks and Benefits of Life Sciences Gain-of-Function Research. Issued by The White House Office of Science and Technology Policy. 17 October 2014: http://www.whitehouse.gov/blog/2014/10/17/doing-diligence-assess-risks-and-benefits-life-sciences-gain-function-research

[2] The interview was about the proposed gain of function experiments on H7N9, which were discussed in a letter by Ron A.M. Fouchier, Yoshihiro Kawaoka and 20 co-authors, published in Nature (*Nature* 500, 150-151, 8 August 2013) and Science (*Science* 9 August 2013: Vol. 341 no. 6146 pp. 612-613).

[3] Vincent Racaniello spoke with Robert Herriman, executive director of The Global Dispatch, about the proposed avian influenza H7N9 virus gain of function experiments on Dispatch Radio, August 2013: http://www.virology.ws/2013/08/13/influenza-h7n9-gain-of-function-experiments-on-dispatch-radio/ I have also prepared a transcript of this interview which can be accessed via this link: http://users.on.net/~peter.hart/Racaniello_GOF_transcript_10_August_2013.pdf

[4] Screen shot of Vincent Racaniello's description of 'gain of function', i.e. it "...simply means you take a virus and you change it in some way so it does something new... something that it didn't do before." http://users.on.net/~peter.hart/Gain%20of%20function%20Racaniello.PNG as depicted in Professor Racaniello's interview on Dispatch Radio: http://www.virology.ws/2013/08/13/influenza-h7n9-gain-of-function-experiments-on-dispatch-radio/

[5] Virologists plan influenza H7N9 gain of function experiments. Published on virology blog, 7 August 2013: http://www.virology.ws/2013/08/07/virologists-plan-influenza-h7n9-gain-of-function-experiments/

[6] More voices call for action on lab biosafety. (Robert Roos). CIDRAP. 31 July 2014: http://www.cidrap.umn.edu/news-perspective/2014/07/more-voices-call-action-lab-biosafety

[7] Doing Diligence to Assess the Risks and Benefits of Life Sciences Gain-of-Function Research. Issued by The White House Office of Science and Technology Policy. 17 October 2014: http://www.whitehouse.gov/blog/2014/10/17/doing-diligence-assess-risks-and-benefits-life-sciences-gain-function-research

[8] *Ibid.*

[9] More information about the NSABB meeting to be held on 22 October, including a hyperlink to the draft agenda, is accessible via this link: http://osp.od.nih.gov/office-biotechnology-activities/event/2014-10-22-121500-2014-10-22-200000/nsabb-meeting

[10] Statement on Funding Pause on Certain Types of Gain-of-Function Research. Issued by Francis S Collins, Director, National Institutes of Health. 17 October 2014: http://www.nih.gov/about/director/10172014_statement_gof.htm

Elizabeth Hart
Adelaide, South Australia
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31 January 2012

An open letter to the NSABB re the political and ethical implications of lethal virus development

For the attention of:

Paul Keim, Acting Chair, National Science Advisory Board for Biosecurity

Please note this letter and your response will be forwarded to other parties for information.

cc: Michael Osterholm, NSABB
Ron Fouchier, Erasmus MC
Ab Osterhaus, Erasmus MC
Yoshihiro Kawaoka, University of Wisconsin-Madison
Peter Palese, Mount Sinai School of Medicine
Tom Jefferson, Cochrane Vaccines Field
Philip Campbell, *Nature*
Caroline Ash, *Science*
Deborah Cohen, *British Medical Journal*

Dr Keim

Further to our previous correspondence on the controversial topic of 'lethal flu virus' development.

Please accept this open letter as a layperson's perspective on this topic.

Ron Fouchier and Ab Osterhaus have questioned "whether it is appropriate to have one country, i.e. the United States, dominate a discussion that has an impact on scientists and public health officials worldwide".[1]

I am astonished at the naiveté of these scientists. Surely it must be obvious by now that by sponsoring development of a potentially lethal flu virus the United States could be in breach of the Biological Weapons Convention[2], which entered into force in March 1975, i.e.

Article 1 of the Convention states:

> Each State Party to this Convention undertakes never in any circumstances to develop, produce, stockpile or otherwise acquire or retain:
>
> (1) Microbial or other biological agents, or toxins whatever their origin or method of production, of types and in quantities that have no justification for prophylactic, protective or other peaceful purposes;

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[1] Preventing pandemics: The fight over flu. Nature (2012). Published online 15 January 2012: http://www.nature.com/nature/journal/vaop/ncurrent/full/481257a.html

[2] Biological Weapons Convention (BWC). Convention on the Prohibition of the Development, Production and Stockpiling of Bacteriological (Biological) and Toxin Weapons and On Their Destruction. Entered into force March 26, 1975: http://www.armscontrol.org/treaties/bwc

Can it be argued that development of a lethal flu virus is justified for "prophylactic, protective or other peaceful purposes"? **I suggest this is highly questionable**. For instance Thomas V. Inglesby, Anita Cicero, and D.A. Henderson say: "We are not opposed to research in high-containment labs using dangerous pathogens, including H5N1. Over the past decade, the Center for Biosecurity has publicly argued for the importance of such research to develop diagnostics, medicines, and vaccines for the most threatening infectious diseases. **But research and development for those purposes does not require engineering lethal viruses to make them more transmissible between humans.**"[3]

In *Foreign Policy*[4], Laurie Garrett describes U.S. Secretary of State Hillary Clinton's surprise visit to the review summit on biological weapons in Geneva in December last year, saying that Secretary Clinton warned of al Qaeda's call to arms for "brothers with degrees in microbiology or chemistry to develop a weapon of mass destruction".

It is interesting that Secretary Clinton deflected concern about the development of biological weapons onto al Qaeda, **while it is actually the U.S. that is actively involved in sponsoring the development of lethal flu viruses**. I wonder if this irony was lost on other signatories[5] to the Biological Weapons Convention, such as Iran and Pakistan?

What position would the U.S. take if countries such as Iran and Pakistan sponsored lethal virus development?

Dr Fouchier has announced his team "mutated the hell out of H5N1", and warned that "this is a very dangerous virus".[6] His claims must be subjected to scrutiny. There are doubts as to whether the flu virus developed in the Erasmus MC laboratory is indeed as deadly to humans as we've been led to believe. (See for example a paper by Peter Palese and Taia Wang: *H5N1 influenza viruses: Facts, not fear*[7]).

Regardless of whether the Erasmus MC developed virus is lethal to humans or not, **the question remains whether it is legitimate for any party to sponsor the development of lethal viruses?**

Perhaps if the controversial flu virus research studies conducted by Erasmus MC and the University of Wisconsin-Madison had undergone an effective ethics approval process we would not be in the unfortunate position we are in now?

In his discussion of *Governance of dual-use research: an ethical dilemma*[8], bioethicist Michael Selgelid describes the conflict between the voluntary self-governance of the scientific community, and the security concerns of bureaucrats and security experts. Selgelid notes that "most of the debates about the dual-use dilemma have primarily involved science and security experts rather than ethicists". Selgelid argues that "biological weapons development may turn out to be one of the most serious consequences of the genetics revolution in biology". He concludes: "It is thus crucially important that there is more ethical input into debates about the governance of dual-use research." Selgelid's paper was published in 2009. Given the current controversy, it appears his warning fell upon deaf ears.

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[3] Editorial by Thomas V. Inglesby, Anita Cicero, and D.A. Henderson. The Risk of Engineering a Highly Transmissible H5N1 Virus. Center for Biosecurity of UPMC. 15/12/2011: http://www.upmc-biosecurity.org/website/resources/publications/2011/2011-12-15-editorial-engineering-H5N1

[4] Laurie Garrett. Flu Season. Foreign Policy. January 5, 2012: http://www.foreignpolicy.com/articles/2012/01/05/flu_season

[5] Biological Weapons Convention Signatories and States-Parties: http://www.armscontrol.org/factsheets/bwcsig

[6] Katherine Harmon. What Really Happened in Malta This September When Contagious Bird Flu Was First Announced. Scientific American. December 30 2011: http://blogs.scientificamerican.com/observations/2011/12/30/what-really-happened-in-malta this-september-when-contagious-bird-flu-was-first-announced/

[7] Peter Palese and Taia T. Wang. H5N1 influenza viruses: Facts, not fear. Proceedings of the National Academy of Sciences of the United States of America. Published online before print January 25, 2012: http://www.pnas.org/content/early/2012/01/24/1121297109.full.pdf+html

[8] Michael J Selgelid. Governance of dual-use research: an ethical dilemma. Bull World Health Organ 2009;87:720-723: http://www.who.int/bulletin/volumes/87/9/08-051383.pdf

A recent article in the *Canadian Press*[9] reports that a "small – in relative terms – group of technical experts will be invited to Geneva in mid-February to begin the difficult task of trying to break an impasse arising from the proposed publication of controversial bird flu research… Participants will include representatives of the Dutch and American research teams that conducted the studies, experts from WHO's network of influenza laboratories and people with first-hand involvement in the dispute."

The *Canadian Press* article quotes Dr Keiji Fukuda, the WHO's assistant director-general for health security and environment, who says: "We are not setting this up as a political meeting. We are setting this up as a meeting of extremely knowledgeable technical people."

Government sponsored lethal virus development ***is*** an important political and ethical issue for the world's citizens. I for one am not comforted by this proposed meeting being confined to "extremely knowledgeable technical people" with possible conflicts of interest. In this regard, I refer you to the following articles *(please see full reference details in the footnotes)*:

- The handling of the H1N1 pandemic: more transparency needed[10]
- WHO and the pandemic flu "conspiracies"[11]
- WHO and the pandemic flu "conspiracies" – Recent Rapid Responses[12]
- Flu experts rebut conflict claims[13]
- WHO failing in duty of transparency[14]
- The Swine Flu Panic of 2009[15]
- 'A Whole Industry is Waiting For A Pandemic'[16]
- In Holland, the Public Face of Flu Takes a Hit[17]
- Mexican flu: a bad and expensive joke[18]

I also suggest a press release published by the industry-funded European Scientific Working Group on Influenza (ESWI), titled ***Doubting the benefits of influenza vaccines is dangerous from both a scientific and ethical point of view***[19] (26 October 2011), should be subjected to critical analysis. Ab Osterhaus, the ESWI chairman, is a contact on this press release.

This brazen attempt to stifle any questioning of flu vaccination is shocking, and must be considered along with other material which does question the benefits of influenza vaccines, such as the Cochrane Reviews *Vaccines for preventing influenza in healthy adults*[20]; and

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Vaccines for preventing influenza in the elderly[21]; and the systematic review and meta-analysis published in the Lancet, i.e. Efficacy and effectiveness of influenza vaccines[22]. A paper on which Dr Osterhaus and Dr Fouchier are included as authors should also be included in the mix, i.e. *Annual vaccination against influenza hampers development of virus-specific CD8+ T cell immunity in children*[23].

The WHO has indicated we are experiencing a vaccine boom.[24] In 2009, Linda Johnson, Associated Press, advised: "Vaccines now are viewed as a crucial path to growth, as drug companies look for ways to offset a slowing of prescription-medicine sales amid intensifying generic competition and government pressure to restrain prices under the federal health-care overhaul".[25] In a report in New Scientist in September 2011, Debora MacKenzie says: "While the rest of the pharmaceutical sector struggles to keep afloat as expiring patents send profits plummeting, the vaccine industry has become remarkably buoyant."[26]

A recent press release on PR Newswire (January 2012) titled *Influenza Vaccine Market Opportunities and Challenges: Worldwide Forecast* notes: the "last few years have seen renewed interest in the vaccines market, overcoming the prevailing view that vaccines are a low-margin business with high barriers to entry. The flu vaccines market has been at the forefront of this trend, partially fuelled by the fear of an impending pandemic. As a result global influenza vaccine market has experienced phenomenal growth in recent years at a compound annual growth rate of more than 65% between 2008 and 2010. This growth was mainly driven by the global spread of H1N1 influenza. But in the year 2011 H1N1 pandemic flu vaccine market declined due to waning threat of swine flu disease. However seasonal influenza vaccine market is predicted to grow year on year and cross US$ 4 Billion by 2015."[27]

Obviously there is big money in flu vaccines...

It is time for a broad investigation of the ever-expanding 'influenza industry', including scrutiny of relationships between vaccine manufacturers and governments and other public bodies. (Consideration of the doubts around Tamiflu[28] also needs to be included in this investigation.)

This is particularly pressing in light of the ongoing calls for compulsory vaccination of medical staff[29], and continuing pressure on the general population to be vaccinated with flu vaccines of questionable benefit.[30]

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In his paper *Corruption in the Government Hospitals*[31] Professor Anwar Ul Haque of the Pakistan Institute of Medical Sciences provides an interesting perspective on flu vaccination in developing countries:

> ...the health budget in the "3rd" world countries is very small. Even most part of this lean budget is stolen away! For example useless and unwarranted vaccines like Swine Flu vaccines are sold to suck the budget intended for the poor patients.(6-30) The corrupt elements use the power and stature of World Health Organization (WHO) and armaments of sophisticated media to create panic to fool Governments and public into buying these vaccines on the expense of treatments for far more common diseases such as malaria, anemia, tuberculosis and malnourishment etc. Some manufacturer of Swine Flu vaccines had become the WHO "experts" and promoted the sale of the vaccines from the platform of WHO. In order to sale these unwarranted vaccines on mass level and thus earning billions o[f] Euros they even changed the basic definition of pandemic.(27-31)

In the conclusion to his paper on corruption in government hospitals, Professor Ul Haque says:

> **For brin[g]ing health change the doctors and other educated people of the society have to play their active role. Freedom of expression, honest and fair evaluation and strict continuing accountability must be put in place.**

Dr Keim, what steps are the NSABB taking to ensure there is appropriate political and ethical representation at the meeting to discuss bird flu research, and an objective and transparent recording of the proceedings?

In the interests of transparency, I request the matters raised in this letter be addressed in the NSABB's forthcoming statement, which you have previously indicated will be published in *Nature* and *Science* this week.

In particular, I suggest it is imperative to clarify the United States' position on lethal virus development in relation to its obligations under the Biological Weapons Convention.

Dr Keim, I would appreciate your early response to the questions raised above. I also request your advice on the progress of your discussions with Professor Palese and his colleagues regarding publication of their letter to the NSABB.

Yours sincerely

Elizabeth Hart

Note: I have initiated discussion on this topic, from a layperson's perspective, on the Bad Science Forum under the title Lethal flu virus research.... http://www.badscience.net/forum/viewtopic.php?f=3&t=27118

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17 December 2012

Ref: Docket: CDC-2012-0010
Influenza Viruses Containing the Hemagglutinin from the Goose/ Guangdong/1/96 Lineage

A submission to the
Centers for Disease Control and Prevention (CDC),
Department of Health and Human Services (HHS)
re Opposition to Lab-engineering of Potentially Lethal Pathogens

cc: Paul Keim and Michael Osterholm, NSABB
Anthony Fauci, National Institute of Allergy and Infectious Diseases, National Institutes of Health
Ron Fouchier and Ab Osterhaus, Erasmus MC
Yoshihiro Kawaoka, University of Wisconsin-Madison
Peter Palese, Mount Sinai School of Medicine
Tom Jefferson, Cochrane Vaccines Field
Philip Campbell, Nature
Bruce Alberts and Caroline Ash, Science
Fiona Godlee and Deborah Cohen, British Medical Journal

I take this opportunity to register my opposition to lab-engineering of potentially lethal pathogens.

This research pushes the limits of legitimate scientific enquiry and risks global public safety. For example, it is pertinent to note that there were 395 biosafety breaches in the United States between 2003 and 2009 – including seven laboratory-acquired infections – that risked accidental release of dangerous pathogens from high-containment labs.[1]

In regards to this matter, please refer to my open letter dated 31 January 2012 re the political and ethical implications of lethal virus development, addressed to Paul Keim of the National Science Advisory Board for Biosecurity (NSABB). The letter is accessible on the internet via this link: http://bit.ly/AfyAtQ

On the subject of H5N1, the US National Institutes of Health should be brought to account for funding research into making H5N1 more transmissible. As I ask in my letter to the NSABB, noted above, is the US breaching the Biological Weapons Convention[2] by sponsoring the development of a potentially lethal flu virus?

Likewise I question the behaviour of the scientist, Ron Fouchier, who "mutated the hell out of H5N1" and admitted he did something "really, really stupid" when the mutated H5N1 was put into the nose of one ferret, and then transferred to others.[3]

When Fouchier did something "really, really stupid", did he pause and think about the ethics of the situation? Did he fully consider the possible consequences of the dual-use research he was undertaking? Did he think about "the social responsibility of science and scientists"[4]? I suspect not.

At the European Scientific Working group on Influenza meeting in Malta in September 2011, Fouchier beat up the results of his research, saying "this is a very dangerous virus".[5] Subsequent to the furore surrounding his controversial research, Fouchier did a 180 degree turnaround on his claims about the lethality of his lab-engineered virus. Apparently the virus he created was neither as contagious nor as dangerous as people had been led to believe.[6 7 8 9 10]

It seems to me there is much fear-mongering in the flu vaccine industry, a matter I touch upon in my letter to the NSABB mentioned above. There are also potential conflicts of interest.

There are forces working very hard to set up a massive international vaccines market in developed and developing countries, and I am not convinced all these vaccine products are justifiable.[11] Relationships between the pharmaceutical industry and organisations such as the World Health Organisation, Centers for Disease Control and Prevention, National Institutes of Health and others, plus governments, should be subjected to scrutiny.

For example, I am sceptical about the World Health Organisation's Pandemic Influenza Preparedness (PIP) Framework[12] and Novartis Vaccines and Diagnostics' option to "produce enough vaccines to protect the 7 billion people on our planet".[13] Given the experience of the 2009 influenza A/H1N1 pandemic which proved to be such a "damp squib"[14] it appears to be very questionable to be investing enormous sums in flu vaccines.

Who can we trust to objectively investigate this matter and protect the best interests of the public?

Elizabeth Hart

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Dr Vincent Racaniello interviewed by Robert Herriman re Influenza H7N9 gain of function experiments

Broadcast on Dispatch Radio, 10 August 2013

Virology Blog: http://www.virology.ws/2013/08/13/influenza-h7n9-gain-of-function-experiments-on-dispatch-radio/
YouTube: http://www.youtube.com/watch?v=cefnT2u7poc&noredirect=1#at=51

Transcript prepared by Elizabeth Hart Contact: overvaccination@gmail.com

Herriman: Well welcome back to Dispatch Radio. Earlier this week there was a letter published in the scientific journal *Nature* that has caused quite a stir amongst the scientific community, and in a nutshell, it's saying that there's a group of scientists that are interested in performing controversial experiments on the bird flu that's circulating in China right now, it's called Avian Influenza H7N9, and to date it's infected 134 and killed 43. Here to try to sort this out is Dr Vincent Racaniello, he's a Professor of Microbiology and Immunology at Columbia University. Hello Vincent, and welcome back to the show.

Racaniello: Hi Robert, thanks for having me back.

Herriman: As soon as I saw the story you came to my mind immediately. So tell me if I got this right. So we've got about 20 odd virologists and they want to do some manipulations on this virus. Is that it in a nutshell?

Racaniello: Yeah, that's basically it. The H7N9 virus which, as you know, just emerged into people this year, seems to have some potential for infecting more people, so these virologists want to do experiments on it and they wrote this letter to let everybody know exactly what they want to do.

Herriman: Sure. And these experiments are called 'gain of function' experiments. In the simplest possible terms, can you try to explain what that means?

Racaniello: So a 'gain of function' simply means that you take a virus and you change it in some way so it does something new, so it does something that it didn't do before. That's all that means. It's quite simple. So you could for example take this H7N9 virus and make it resistant to an anti-viral drug, that would be a gain of function.

Herriman: Sure. And I read your post on Virology Blog the other day, and clearly you are pro gain of function experiments. And I guess my question is, and for our listening audience, why would anybody want to do this?

Racaniello: So, to really understand how this virus works, and really any other virus, we do gain of function studies all the time. We don't make a big deal of it, we don't write letters telling the world that we're going to do them because that's not the way science works. Science works by just doing your experiments. We do this because we would like to see what kinds of changes would lead to a gain of function, and what would be the consequences. So, in the case of this virus, these investigators want to make the virus drug resistant. As you know, there are a couple of anti-virals that you can use if you get influenza – Tamiflu, Relenza - and these investigators want to make the virus resistant. And the reason they want to do that is to see if a drug resistant mutant would have any properties that would make it scarier in people. So there is really a goal to these experiments. They want to know if you change the virus what might be the consequences for people. And as I said this is done all the time but these virologists decided to tell the world about it.

Herriman: And that's probably because it's kind of déjà vu to 2011 when they, they individually did the H5N1 experiments which caused all kinds of controversy.

Racaniello: Yes, as you know as a result of that there was a moratorium on that H5N1 transmission research for a while, and part of the goal of that moratorium was to try and increase telling the public what you're doing with this virus. And I think these authors being part of that whole scenario decided it might be a good idea to tell the public ahead of time what they're going to do.

Herriman: So they're sort of playing politics with this?

Racaniello: I suppose, in a way...

Herriman: Yes...

Racaniello: ...as a scientist I wouldn't do this because I think you get into trouble. As you know the press has boiled up over those letters and the headlines are incredible...

Herriman: Sure.

Racaniello: ...and I think it's better to do the experiment, and if it works out publish it, and then explain why you did it afterwards.

Herriman: OK. Now there's plenty of arguments against performing such experiments. Some people are saying, and I want you to respond to each one, that these engineered strains could be accidentally or deliberately released from the lab, sparking a flu pandemic. What do you say to that?

Racaniello: I think this is very, very unlikely. The way I view it is, you have to balance what you might get from an experiment versus the potential danger. And in this case the potential benefits far outweigh the dangers. These experiments would be done under high containment, the likelihood that a virus would escape is really, really low. Plus, whenever you do a gain of function, the virus always loses something in exchange. And in the H5N1 gain of function experiments where they adapted them to ferrets, to aerosol transmission, that was a gain of function. What those viruses lost was their ability to cause disease when they spread by that route. So I don't worry about a dangerous strain getting out at all, I think that likelihood is really negligible.

Herriman: OK. And another critique is, some are saying that the animal models, such as ferrets, yeah, they can provide some information as far as risk of transmissibility and pathogenicity. However, how do you extrapolate that to humans?

Racaniello: Right. Now this is something I have always contended, you cannot make predictions about what will happen in humans based on an experiment in an animal like a ferret, or a guinea pig or whatever your chosen animal model is. But you still have to do these experiments because they provide you other kinds of information. For example, the ferret transmission experiment, with H5N1, they, the results of those were a series of genetic changes that allowed the virus to transmit in the air from one ferret to another. Those don't predict the changes that might be important in humans. However, those changes give you an idea about how it works to make a virus better to transmit in the air. In other words what is the function of these changes? And I think the function is conserved(?) between animals and humans. So they do provide a lot of information. They're not predictive, but they provide what we call mechanistic information about very specific aspects of human disease.

Herriman: Yeah. In kind of a follow-up to that, I saw in one AP report this week, one scientist, who is definitely a critic of the experiments, said, you know, we tried this with the H5N1 two years ago, you know, we got nothing out of it, you know, should we do all this work if it's not actually going to make a difference?

Racaniello: Well I would totally disagree with the conclusion that we got nothing out of it. I would send that scientist to my blog, I've got a series of posts about what we learned from those H5N1 experiments. We learned a great deal. We were shown things that we'd never understood before about what makes a virus able to transmit in the air. So I argue that we're going to learn a lot from these types of experiments.

Herriman: Yeah, and if you weren't aware Vincent, I just saw a press release from Hong Kong today, and they have a suspected H7N9 case in China right now, and so that may be number 135.

Racaniello: Yeah, I saw that on your blog this morning.

Herriman: Oh, great! Fantastic, got to love that... Anyway, Vincent Racaniello, Dr Racaniello, has a fantastic blog called Virology Blog. If you have any remote interest in the science of virology, this is the place to go. He did a fantastic article concerning these gain of function experiments, and he

makes a very strong case for performing them. And I just found it great. And also if you're interested he has a very good Facebook page too, it's called This Week in Virology, am I correct?

Racaniello: Yes, that's right facebook.com/thisweekinvirology

Herriman: Yeah, and that's a good place to go too for information and… Alright Vincent, well I appreciate you coming on and clearing the air on this and telling us what you think, I appreciate it.

Racaniello: It's always a pleasure. Could I plug my Coursera course?

Herriman: I'm sorry?

Racaniello: I teach a course on Coursera and I'd like people to know about that.

Herriman: Oh OK, go ahead, go for it.

Racaniello: I teach a virology course, it's free on the Coursera site. It's coursera.org and you can find a link to that on my blog virology.ws. It just started last week, it's free, and we take you step by step through understanding virology, so I think you'll like it.

Herriman: That's great. And I have a lot of Facebook fans that are very interested in these kind of topics, so hopefully that'll get spread around the globe, because it is a global thing. Alright, Dr Vincent Racaniello from Columbia University, thanks for joining us on the show.

Racaniello: Robert, always great to talk to you, thank you.

Herriman: Alright, same here. Bye bye.

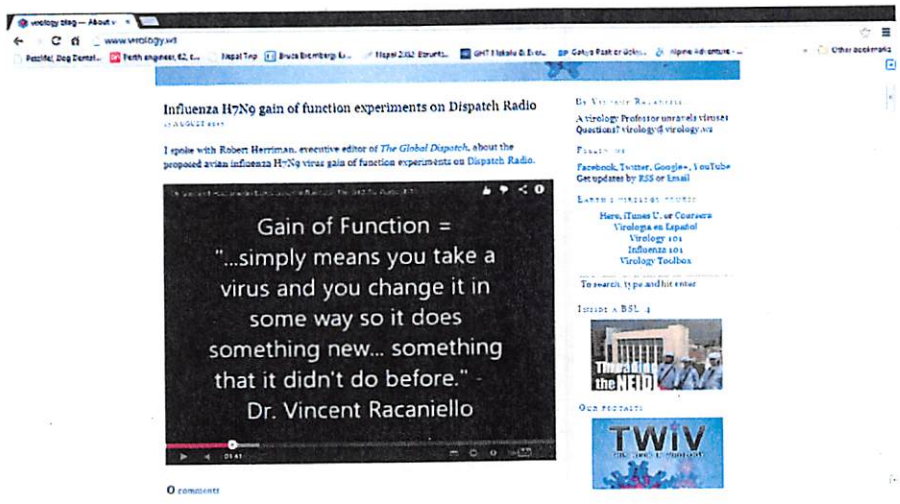
Racaniello: Bye.

Other commentator: Alright, well, fantastic as always Bob. And it's really awesome to know that Dr Racaniello is following your work as much as you're following his.

Herriman: That's fantas…that caught me off guard a little bit. So I'm really pleased, and considering that this particular story has got a lot of, I mean a lot of the stuff we talk about here on the show doesn't get necessarily the coverage it should, this has got probably more coverage than it should have, and not being a virologist by training, it's good to read and listen to what Dr Racaniello has to say, and even reading the comments on his blogs, because not everybody agrees.

Other commentator: Yeah, it's fantastic.

Screen shot of Dr Vincent Racaniello's description of 'gain of function' virus research



The Vagueness and Costs of the Pause on Gain-of-Function (GOF) Experiments on Pathogens with pandemic potential including influenza virus

Michael J. Imperiale[1] and Arturo Casadevall[2]

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Since the spring of 2012 there has been a raging controversy in scientific circles on the wisdom of carrying out so called 'gain-of-function' (GOF) experiments with pathogens of pandemic potential (PPP) such as influenza virus [1]. Although the phrase 'GOF' has been much criticized because of its inexactness, the terminology has been adopted by many including the media to mean experiments where the result is a change in virulence or host tropism for a PPP. The nugget of the debate is a disagreement over the practical value of such experiments relative to the information that they produce with opponents arguing that risk, whether from intentional release or, more likely, laboratory accidents, outweighs any knowledge gained [1]. Some anti- and pro-GOF experiment proponents have organized themselves into two camps known as the Cambridge Working Group (CWG, (http://www.cambridgeworkinggroup.org/) and Scientists for Science (SFS, http://www.scientistsforscience.org/), respectively, that have issued statements. However, these groups are heterogeneous and their members have varied views on the problem. Both authors have signed the CWG statement and one author (MJI) has also co-signed the SFS statement because both authors see important benefits for GOF work involving PPP, are nonetheless concerned about safety issues, and most importantly strongly support the common call for discussion. However, neither author has supported the idea of a moratorium on this type of research [1, 2].

In October 2014, the White House announced that the US Government (USG) was implementing a "pause" of new funding for research involving GOF experiments with three respiratory viruses, influenza virus, MERS coronavirus, and SARS coronavirus, if that research could be "reasonably anticipated" to result in enhanced pathogenicity or increased transmissibility (http://www.whitehouse.gov/blog/2014/10/17/doing-diligence-assess-risks-and-benefits-life-sciences-gain-function-research). They also asked that ongoing experiments which fall into this category be voluntarily stopped. During the pause, the USG has asked both the National Science Advisory Board for Biosecurity (NSABB) and the National Academies to engage in discussions aimed at how to assess the risks and benefits of GOF research. We ourselves have been calling for such deliberations and welcome that aspect of the White House announcement [1]. The events at the CDC this summer, in which a highly pathogenic avian influenza strain was accidentally shipped to a USDA lab, and in which *B. anthracis* spores were taken out of a lab without proper disinfection, heightened concern both in the scientific community and in the public about whether research with dangerous pathogens is being carried out with appropriate safety measure in place. These accidents, together with a growing chorus of scientists who are worried about GOF experiments [3], seem to have precipitated the government action.

Pauses and moratoriums are blunt instruments in science and carry the potential for unintended consequences. We recognize that the pause is a response from well-meaning government officials who are tasked with trying to find ways to minimize potential dangers from GOF experiments. We note, however, that depending on what interruption of work is counted, this is at least the third pause/moratorium in this field with the first being voluntary, the second requested by the USG [4, 5], and the third being the current 'pause'. We have numerous concerns with this third stoppage that include the timing of the announcement relative to the ongoing debate, the vagueness in the wording of the statement, and the potential effects on the fields of influenza virus and coronavirus research. Each concern will be discussed separately.

The timing of this 'pause' is perplexing given that one might have expected this action to follow a concerted effort to explore the issues rather than to precede detailed discussions. Many have drawn the analogy between the current situation and that surrounding the advent of recombinant DNA technologies. However, there are significant differences: the discussions at Asilomar preceded a self-imposed moratorium by molecular biologists working on recombinant

DNA technology [6]. It seems that this should have been the case now: the NSABB could have been deliberating on this topic in the two years that have passed since the GOF debate began with the publication of the two manuscripts describing mammalian transmission of H5N1 influenza virus [7, 8]. Instead, it did not even meet and this created a vacuum of discussion that may have contributed to the current crisis. In contrast, the government has responded to the heightened controversy by reactivating the NSABB while simultaneously calling for a pause of GOF work before a meaningful discussion. Although this course of action seems to emphasize safety and caution, it carries significant risks that we will discuss below. It is also unclear to us why the pause is necessary given that the government is already presumably providing an extra layer of review of GOF experiments that followed the prior moratoriums (http://www.phe.gov/s3/dualuse/Documents/us-policy-durc-032812.pdf) and has asked institutions to do the same (http://www.phe.gov/s3/dualuse/Documents/durc-policy.pdf).

We are concerned that the wording of the pause is vague and could have unintended consequences. First, the pause has no end date. Will the NSABB and the Academies be nimble enough to make concrete recommendations that are broadly acceptable within months? Given the pace at which these committees generally function we worry that this will not be the case. Having the pause drag on for too long will not only affect research progress, but also the careers of the scientists engaged in that research. Second, we worry about the meaning of "reasonably anticipated." Obviously this phrase is very subjective, and similar wording in the definition of dual use research of concern (DURC) has already made assessments of what constitutes DURC very problematic for journals and authors [9]. At one extreme cautious researchers could over interpret the vague wording and stop experiments that were not intended for inclusion in the pause order. For example, albeit somewhat extreme, any time one grows an RNA virus in the laboratory, even in cell culture, the error prone nature of the viral RNA polymerase results in each progeny genome containing more or less one mutation. Any scientist versed in RNA virus biology could 'reasonably anticipate' some of these mutations would impose a gain of function on the virus. However, if one does not select for that function, it is extremely unlikely that that mutant will overtake the population. We therefore suggest that the intent of the experiment must be considered before making a determination of whether it should be paused.

The pause will almost certainly have a disruptive effect on several laboratories at a time when information derived from GOF experiments is beginning to bear fruit in pandemic preparedness. In this issue of *mBio*, Stacey Schultz-Cherry and colleagues describe how mutational information from GOF is producing actionable information on surveillance studies and selection of strains for vaccines (insert ref). The pause means that some information from GOF experiments will cease to become available, with potential negative consequences on preparedness. Ongoing experiments will stop and the vagueness of the wording raises the possibility that other work will not be done due to an abundance of caution. For example, there is tremendous need for rodent models of coronaviruses with pandemic potential including the agents responsible for MERS and SARS. Such models could greatly facilitate the discovery of new drugs and vaccines. However, developing such models requires changing the host tropism of the virus and as such they fall under experiments of concern despite the fact that human viruses often lose virulence as they adapt to other species. The current pause affects two dozen studies that include experiments to develop rodent models of coronavirus research [10]. In this regard, the reader may want to listen to a story on National Public Radio in which researchers discuss how the pause is affecting coronavirus research (http://www.npr.org/blogs/health/2014/11/07/361219361/how-a-tilt-toward-safety-stopped-a-scientists-virus-research).The inclusion of this work is an example of how pauses and moratoriums can be blunt instruments with major unintended consequences.

Finally, we worry that work being carried out by graduate students and postdoctoral fellows will be put on hiatus, causing disruption to their plans for completing their training. Although some will argue that this is a small price to pay for ensuring safety, we worry that this could have a tremendous effect downstream as investigators may be discouraged from resuming such studies in the future. Furthermore, bright young scientists who have a choice of what research to pursue may avoid this area of investigation because of its controversy, unpredictability, and increased restrictions. Research output is not like a factory line that can be shut down and re-started depending on supply and demand. Instead research output is dependent on the presence of ongoing projects by dedicated scientists who carried them out in good faith, hoping to generate useful information. When students and postdoctoral fellows stop such projects they inevitably move to other problems and it may be difficult to jump start GOF experiments once laboratories cease doing that type of work. As such, we are more concerned about pausing ongoing projects than delaying the start of new lines of investigation. Given that a healthy research enterprise is humanity's best defense against future threats from these respiratory pathogens, the pause could hurt future progress by discouraging the best and the brightest from joining this field. Hence, this pause, which is presumably intended to safeguard society from laboratory accidents and unintentional releases, could have the paradoxical effect of leaving humanity more vulnerable to future pandemics by virtue of the information that was not obtained.

As we have written previously, understanding the pathogenicity of these viruses is necessary if we want to develop new therapies and vaccines, and ensure useful surveillance [1, 2]. Clearly, the research must be performed under biocontainment conditions that minimize the risk of accidental release. The discussion that the White House is asking for must occur because the status quo is not acceptable. We call on the government to provide clarity regarding what truly should be paused and for how long. We call on the NSABB and the NAS to move rapidly on this issue, to consider whether the current biosafety practices put in place after the prior moratoriums are sufficient, and if found to be so, to state so without a need for new layers of mandates for what is already a highly supervised field. To repeat ourselves [1], we must get this right.

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From: Steven Salzberg
Sent: Monday, November 10, 2014 7:57 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Cc: Steven Salzberg <salzberg@jhu.edu>
Subject: NSABB Public Comment

Dear NSABB:
I am writing to express my deep concerns about the gain-of-function research that has been conducted by Ron Fouchier, Yoshihiro Kawaoka, and other senior influenza researchers.

I have a longstanding interest in influenza research and vaccine design, and I am one of the co-founders of the Influenza Genome Sequencing Project, an NIAID-funded effort that has sequenced over 10,000 isolates since 2004. I have published scientific papers on the flu virus (Ghedin et al., Nature (2005), 1162) as well as commentaries (see my 2008 Nature commentary).

Gain-of-function research on the flu has created new, dangerous strains that would never occur in nature. There is no evidence that these provide any benefit in predicting the natural evolution of the flu, help to design vaccines, or aid surveillance in any way. Fouchier and colleagues have made arguments that amount to little more than hand waving, such as "this will aid our understanding of the flu." Bluntly speaking, that is nonsense.

I write a widely-read column at Forbes magazine and have just recently posted an article expressing my opposition to gain of function research (http://www.forbes.com/sites/stevensalzberg/2014/10/20/should-we-allow-scientists-to-create-dangerous-super-viruses/). I wrote about it a year ago as well (http://www.forbes.com/sites/stevensalzberg/2013/08/08/scientists-will-create-a-deadly-new-flu-strain-just-to-prove-they-can/), in a piece that now has over 50,000 views.

As I wrote in my Forbes column, we have enough problems simply keeping up with the current flu outbreaks - and now with Ebola - without scientists creating incredibly deadly new viruses that might accidentally escape their labs. Fouchier and Kawaoka's research hasn't changed our ability to respond to a pandemic, not even slightly. Nor has it changed our strategy for vaccine design - and I can't see that it ever will.

Gain-of-function research on viruses is both dangerous and irresponsible. The benefits are minimal if not zero. (And note that I am strongly in favor of investing in research on better treatments for influenza and other viruses, as well as better surveillance.) I strongly support a permanent ban on this research. Please shut it down and keep it shut down.
Sincerely,
Steven Salzberg
--
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From: Charles Stack [mailto:cstack3@uic.edu]
Sent: Wednesday, November 12, 2014 3:26 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: NSABB Public Comment

Dear Madame or Sir:

I am writing to express my personal and professional concerns about ongoing "Gain of Function" (GOF) experimentation performed by some academic researchers in the USA and internationally.

As a practicing epidemiologist with 30 years of experience in disease management and infection control, I understand the powerful forces that nature brings to bear on all of us. The public health community must constantly be on guard for novel, emerging infectious agents, or mutated agents capable of causing pandemics. Recent examples of these include the H1N1 swine influenza pandemic, emergence of MERS, and ongoing outbreak from the Ebola virus.

The rationale for performing GOF experiments on dangerous pathogens including avian influenza types H5N1 and H7N9 is weak at best. Researchers say that this will inform the scientific community about specific genetic mutations to be vigilant against; however, nature itself is the world's largest laboratory, and the odds of creating a mutation in the lab that will be identical to a natural mutation are vanishingly small.

Other reasons given for GOF research (scientific curiosity etc.) are hollow, since the risk of accidental release of a mutated pathogen into society far outweighs any insights we might obtain from this experimental work. The funding would be better spent on field surveillance for emergence of dangerous pathogens in animal and human hosts, research for a "universal" influenza vaccine, and improved vaccination of vulnerable populations. Humanity cannot afford to have a lab-originated pandemic occur when we have enough problems with naturally occurring emergent pathogens.

Therefore, I support ongoing efforts of the NIH to suspend funding for GOF experimentation until all of the scientific, ethical, and safety issues can be thoroughly discussed in an open forum. As a Charter Member of the Cambridge Working Group, I support Dr. Marc Lipsitch and my colleagues in their work to inform the public about the true risks of GOF research and evaluate the safety of these procedures.

Thank you for your consideration of my statement, and I wish you well in your upcoming deliberations.

Sincerely,
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Ralph Baric, Ph.D. & Mark Denison, M.D.

November 12, 2014

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NSABB Public Comment
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RE: Gain of Function Pause and Implications for Coronavirus Animal Model Development

To NSABB members,

As virologists engaged in genetic studies of coronavirus replication, pathogenesis, evolution, receptor recognition, adaptation, and vaccine and therapeutic interventions, we express our profound concerns regarding the recent US Government directive to "*temporarily halt all new funding for experiments that seek to study MERS-CoV and SARS-CoV using gain of function strategies that might increase pathogenesis and transmissibility in mammals"*. The term, "gain of function" (GOF)" has become so broadly over-used and encompassing that it now poses a serious risk to block development of new public health intervention strategies to combat the ongoing MERS-CoV outbreak. Additionally, this decision will significantly inhibit our capacity to respond quickly and effectively to future outbreaks of SARS-like or MERS-like coronaviruses, which continue to circulate in bat populations and camels. To our disappointment, the recent NSAAB meeting (Oct 22) which was called to initiate a deliberative process toward a standardized policy on GOF studies did not include a single nationally or internationally recognized coronavirologist, especially one who regularly performs genetic studies on SARS-CoV and MERS-CoV. Rather, the initial meeting focused almost exclusively on the risks – and much less so on the benefits - of "gain of function" (GOF) transmission studies of influenza viruses. The inclusion of SARS-CoV and MERS-CoV, as best we can glean, is based on the pandemic potential and respiratory transmission of the natural isolates and fails to recognize the substantial biological differences that exist between myxoviruses and coronaviruses***.*** We would note that studies to enhance transmissibility have never been conducted using a coronavirus. ***In fact, model systems to perform such studies in coronaviruses do not exist.***

We would like to present several experimentally-validated positions that document: 1) the ongoing emergence and high mortality of the MERS-CoV without a vaccine or therapeutic; 2) no transmission models for SARS-CoV or MERS-CoV; 3) critical need for animal models and lack of safe alternatives to animal testing for emerging coronavirus therapeutic and vaccine design; and 4) the benefits of the few GOF related studies that have been performed using MERS-CoV and SARS-CoV. All of these will be highly negatively impacted if not aborted by a pause on MERS-CoV and SARS-CoV research, to the great detriment of global health preparedness.

1. Emerging coronaviruses in nature do not observe a mandated pause. Phylogenetic studies supports the hypothesis that all currently known Human CoVs have emerged in the past ~800 years. Since 2003, three new emerging coronaviruses have circulated the globe – 2 human and one mammalian: Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV; 2003), Middle Eastern Respiratory Syndrome Coronavirus (MERS-CoV; 2012) and Porcine Epidemic Diarrhea Virus (PEDV; 2012). Since 2012, PEDV has caused millions of deaths in young piglets to the detriment to the US porcine industry. SARS-CoV had every characteristic of a pandemic virus. Fortunately it was controlled by aggressive public health interventions. However, that success was highly likely predicated on the fortuitous presence of biological vulnerabilities of SARS-CoV: a low R_0 and a requirement for symptomatic disease for transmission (Fraser PNAS 2004). Any confidence in the elimination of risk for future SARS-like CoV outbreaks is not well supported. In fact, recent studies demonstrate the existence of heterologous bat strains of a SARS-like CoV that are capable of using human receptors for docking and entry (Ge et al., Nature 2013). Thus the threat of a SARS-CoV or SARS-like virus still exists and is it unknown if existing public health or medical countermeasures would control disease outcomes or the pandemic potential of these heterologous isolates.

MERS-CoV, in contrast to SARS-CoV, poses an ongoing serious risk to US and global public health. MERS-CoV was initially identified in April 2012, and approximately 929 documented infections and 372 deaths have been reported in 23 countries, including the United States (proMed Mail.org). Of great concern, the pace of new cases once again is increasing and mortality remains ~40%. Dromedary camels and bats are thought to represent intermediate and primary hosts, respectively. It is possible that human infections have occurred for years, since camel sera from the early 1990's neutralize human MERS-CoV strains. Millions of dromedary camels are distributed widely from equatorial Western and Northern Africa through the Middle East. These camels are also traded across the globe. Camel-to-human and human-human transmission has been repeatedly documented during the outbreak. Thus it is likely that sporadic disease has occurred for years in Africa and the Middle East. In severe cases, MERS-CoV infection results in acute respiratory distress syndrome (ARDS), a clinically challenging severe end stage lung disease resulting in mortality rates of 30-50%. Importantly, milder illness and subclinical infections have also been identified and asymptomatic spread also appears to occur in human populations. Thus the MERS CoV outbreak has all of the features that made SARS-CoV so formidable in 2003, important additional features that complicate control and termination of the epidemic, specifically ongoing camel-human transmission and lower level infections with human-human transmission. *These features demonstrate the significant pandemic potential of MERS-CoV and argue that investment in diagnostics, basic research, vaccine design, and therapeutic testing is critical to prevent significant economic losses, and importantly, continued human morbidity and mortality worldwide.*

2. Transmission models have never been developed for SARS-CoV and MERS-CoV. In fact, MERS-CoV does not replicate in mice, guinea pigs and ferrets. The block in MERS-CoV replication is robust, driven by sequence differences that impede spike-receptor interactions and the presence of a glycosylation site which presents a large bulky carbohydrate moiety at the virus receptor binding interface, preventing binding and entry (Cockrell A, 2013, JV). Attempts to adapt by passage or engineer by structure-guided redesign have failed to isolate MERS-CoV host range mutants in multiple laboratories. If developed, the significant differences in primary sequence and carbohydrate presentation seen in the mouse, ferret, guinea pig DPP4 virus-receptor binding interface, which are substantially different in the human DPP4 receptor, will select for mutants that gain animal DPP4 usage while losing significant affinity for the human DPP4 receptor, attenuating pathogenesis. ***Thus, the classic flu transmission model systems which use identical receptor moieties across mammalian species simply don't exist for MERS-CoV.*** SARS-CoV does not replicate in the guinea pig, and replication in the ferret is limited, resulting in minimal disease phenotypes. SARS-CoV binding to the ferret angiotensin 1 converting enzyme 2 receptor (ACE2) is weak, requiring adaptive changes to enhance replication efficiency in this species. While it is possible to select for virus mutants that could use the ferret receptor more efficiently, human receptor usage will likely suffer substantially in these ferret adapted viruses. Moreover, passage experiments have not been reported, because research laboratories have focused their studies in the mouse model, which more accurately and faithfully reproduces the human disease condition (see below). In addition, no one has reported or attempted to develop a SARS-CoV transmission model.

3. A critical need for MERS-CoV animal model development. *Mouse adaptation of human viruses is a common practice, viewed safe as these viruses oftentimes replicate less efficiently in human cells.* For instance, we note that the mouse adapted influenza PR8 strain is fully attenuated, and won't revert even after repeated passage in humans (Beare and Hall, 1971; Beare A et al., 1975). The US government directive halts all animal virus passage studies with influenza, MERS-CoV, and SARS-CoV, including in mice. Unlike influenza, the science behind emerging CoV inclusion has never been openly discussed or debated in an open forum. Therefore, this decision is potentially dangerous and likely based on misinformation, especially troubling in light of the ongoing epidemic and complete lack of therapeutics and vaccines for MERS-CoV. The development of drugs and vaccines require robust small and large animal models of human disease. Further, the FDA will likely apply a three animal rule for the emergency use of drugs and vaccines in an outbreak setting for any newly emerging coronavirus. Please note the following facts:

i) Lack of robust animal models for MERS-CoV disease. In the current MERS-CoV models, which include various primate species and camels, infection severity is limited and the

disease outcomes do not reflect clinical disease seen in severe human infections, e.g., those at most risk for fatal disease outcomes. Mortality in these animals is very low and some models like the marmoset do not appear reproducible across laboratories, and acquiring these rare animals is difficult. MERS-CoV does not replicate in mice, hamsters, ferrets or guinea pigs or any other readily affordable and malleable small animal model species. High-throughput drug and vaccine testing is seriously constrained in primates or camels, because of ethical concerns, lack of facilities for large animal testing, and cost, so most candidate therapies are sitting on a shelf and not being evaluated. ***Thus, mouse models represent the only viable alternative.*** Mouse models under development include mice transduced with Adenovirus vectors encoding the DPP4 receptor, or transgenic mouse lines; (Perlman, 2014) however these models appear to support virus replication without serious clinical disease and do not replicate the end stage lung disease ARDS phenotypes reported in human populations. Vector and transgene induced inflammation further complicate immune readouts as well.

ii) In vivo passage is essential to the development of robust, safe, small animal models of MERS-CoV human disease. Many human and animal respiratory viruses have been adapted to mice. This requires iterative passage to select for multiple mutations that afford alternative species receptor usage, increased virus replication, increased yields/cell and enhance severe clinical disease outcomes. In SARS-CoV, 6-9 mutations are selected in 4-5 genes; the spike glycoprotein receptor binding domain mutations in combination with 2 or more other mutations regulate lethal outcomes (Roberts et al., 2006). Critically, ***no evidence link coronavirus in vivo mouse passage with increased human ris***k. These outcomes also reflect well-described results in many virus systems that serial passage in one species usually attenuates virus pathogenesis in the original species. Mice infected with wildtype or mouse-adapted SARS-CoV do not transmit these viruses to co-housed naive animals. In fact, serial passage in alternative hosts is an accepted strategy that has been widely used to attenuate many human viruses, resulting in live-attenuated viruses that have saved hundreds of millions of lives since the late 1950's.

ii) Mouse adaptation of SARS-CoV. Based on the new criteria for GOF outlined in the US Government directive, three gain of function experiments have been performed with SARS-CoV since 2003 and none have been performed with MERS-CoV***.*** Wildtype SARS-CoV replicates poorly and does not produce clinical disease or pathology in mice. Doubly-inactivated, vectored and recombinant protein vaccines provide robust protection in this model ( ). However, two groups have shown that serial *in vivo* passage rapidly selects for mouse-adapted strains that produce more severe clinical disease and death in young mice, and ARDS and death in aged mice. In aged mice, the LD_{50} drops significantly and disease vulnerabilities and outcomes phenocopy those seen in aged human populations.

Correlates of protection are key metrics used in vaccine development and therapeutics must effectively reduce peak virus titers seen in human patients. Importantly, these correlates can vary depending on virus replication efficacy and the severity of disease pathology noted in humans and in animals. For example, correlates needed to reduce virus titers from 10^5 to 10^3 or 10^8 to 10^6 (two logs) might be substantially different. Vaccines can also elicit protective or pathogenic responses, which can only be identified using animal models. Thus, robust animal models are key to human health.

4. SARS-CoV and MERS-CoV Gain of Function Experiments. Based on influenza virus transmission studies, the underlying assumption appears to be that all GOF studies pose grave public health risk. This represents a very negative over-simplification of a classical, critical and essential genetic approach to defining pathogenesis, virulence, and mechanisms of therapeutic and vaccine efficacy. This is particularly the case for coronaviruses.

Implications in model development. Importantly, vectored and doubly inactivated vaccines work well in virus replication mouse models, but fail to protect against the lethal challenges, especially in aged immunosenescent animals that recapitulate severe lung pathologies. More seriously, doubly inactivated vaccines induced a Th2 immune pathology associated with massive influxes in the numbers of eosinophils and neutrophils; effectively causing a gain in virus pathogenic potential in an unpredictable manner (Bolles et al., 2012). The resulting increased immune pathology can sometimes progress to fatal disease and similar findings have been reported in primates. Thus under the most literal interpretation, experiments to unravel mechanism must cease immediately and be subject to

review. If in vitro correlates of protection (e.g., neutralization titers, T cell responses, etc.) and minimal animal models are used to justify human vaccine use, the surprising outcome would have been that the existing data would have supported the use of doubly inactivated vaccines in human populations, potentially enhancing serious disease outcomes and death in a SARS outbreak setting. This revelation was absolutely dependent on the availability of a robust animal model of human disease. In a second example, Deng X, et al 2013 used GOF approaches to re-engineer an alphavirus, sindbis virus, to express the SARS-CoV papain like protease, designing a safer BSL2 virus surrogate pathogenesis model for rapid drug screening. Insertion of the SARS gene into sindbis attenuated pathogenesis in wildtype but not especially designed mutant mice. Sindbis causes systemic disease, viremia, and replicates in multiple organs but is most tropic for the brain and CNS. SARS-CoV is a pneumoenteric pathogen. Under identical conditions, drugs that were highly efficacious in the surrogate model, failed to protect animals from lethal SARS-CoV challenge (PMC4178736). Thus, results in surrogate models should be evaluated cautiously.

ii) Zoonotic SARS-CoV. Emerging viruses exist in swarms of highly heterologous but related viruses, thus, future outbreaks could be derived from other precursor strains which are antigenically and genetically distinct. Antigenic variation could obviate the potency and efficacy of SARS-CoV vaccines and immunotherapeutics or erode the therapeutic potency of antiviral drugs. To address this issue, the spike glycoproteins of several zoonotic SARS like viruses (e.g., civet, raccoon dog and bat) have been incorporated into the wildtype SARS molecular clone, producing chimeric viruses that encode natural variation in the S glycoprotein (PMC1933338, PMC2588415, PMC3977350, PMID:24172901). These recombinants can use the human, bat and civet receptor, some produce lethal disease with ARDS in aged mice, and demonstrate a 5-100+ fold reduction in neutralization by sera targeting the epidemic SARS-CoV S glycoprotein. Vaccines using the SARS S glycoprotein do not protect against lethal heterologous spike challenge, especially in aged animals; thus, current SARS vaccines will fail to protect against these precursor strains should they seed future outbreaks. In fact, the doubly inactivated vaccines don't protect but do stimulate the Th2 immune pathology noted above (PMC3209347). Similarily, one strain appears resistant to the existing panel of broadly neutralizing human monoclonal antibodies. It should be noted that none of these strains are transmissible in the mouse and most replicate poorly in primary human airway epithelial cells. For surveillance and the development of public health intervention platforms, these data have huge implications, demonstrating that existing vaccines require reformulation. These outcomes could not have been predicted from in silico sequence information, biochemical assays, neutralization assays with surrogate viruses or surrogate in vivo models of human disease. Animal models can lie, however, their reliability is oftentimes directly proportional to their capacity to replicate human disease.

Lack of Safe Alternatives to Animal Testing. Concerns around influenza virus transmissibility studies have now encompassed any gain of function study performed with certain high path viruses in mammals. Various groups have suggested that "ethical" and safer alternative approaches exist that provide equivalent information in the absence of risk. These include the use of pseudotyped defective viruses, recombinant protein biochemical assays, and dynamic modeling of biological processes. These approaches are not robust surrogates of disease models. For example, we note that virus particles breathe, thus some immune epitopes are quaternary in design and are only formed in intact virus particles (PMC4178732). Essentially their existence is entirely dependent on the conformational ensemble that exists in a mature virus preparation, not necessarily in pseudotypes or in recombinant proteins (PMC3358852; PMC4136251). Thus, neutralization and biochemical assays using pseudotype particles or recombinant proteins can provide misinformation. While vaccine and therapeutic potential can be predicted using biochemical assays, dynamic modeling simulations and in vitro neutralization assays and T cell killing assays, these studies are subject to error and protective efficacy can only be evaluated in the context of an animal model of human disease. If these animal models are not robust, correlates of protection may change or be over-interpreted as manufacturers move their products into human populations.

Expert Recommendations. ***First*** and foremost, we argue that it is premature to include the emerging coronaviruses under these restrictions, as scientific dialogue that seriously argues the biology, pros, cons, likely risks to the public, and ethics of GOF have not been discussed in a serious forum. ***Second,*** we recognize the potential dangers of transmission models and encourage open

diaglog and discussion. ***Third,*** we propose that the development of a graduated system be considered that captures perceived risk as a function of the significant biological differences that exist between viruses. As such, we note the: a) significant barriers and difficulties in developing emerging CoV transmission models (which don't yet exist); and b) differences in virus-receptor engagement and host range restrictions that would likely occur should someone actually decide to develop transmissibility models for these particular emerging coronaviruses. ***Forth,*** we note that reverse genetic approaches that employ loss of function strategies (gene inactivation/deletion) almost universally result in severe attenuation and that mouse-adapted models are key to reduced public health risk. These scientific approaches should be encouraged, not discouraged. ***Fifth,*** developing a regulatory framework that over-reaches and hampers these traditional genetic strategies are not in the public interest, as these basic studies in pathogenesis provide gateway discoveries for future treatment strategies. In the case of the emerging coronaviruses, the lack of targeted scientific discourse detracts from the credibility of the process.

We live in unprecedented times, as four highly pathogenic emerging viruses (e.g., H5N1, H7N9, MERS-CoV, Ebola) are currently circulating and causing severe disease in human and animal (PEDV) populations. Decades of research on emerging pathogens have revealed a common pattern; specifically, recurrent introductions of zoonotic strains into human populations, the emergence of mutations that promote adaptation and then transmissibility in the new host, and virus spread throughout the target populations. Influenza viruses and coronaviruses are examples of viruses that crossed and rapidly adapted to new species, resulting in high mortality and disruption of global economy. The pandemic potential of these viruses is clear, but they also are vulnerable in the early stages of an outbreak to public health intervention methods. For public health preparedness, a well-defined and rapidly implemented program of research is needed including the availability of robust small and large animal models of human disease. GOF experiments are a documented, powerful tool to understand viral pathogenic mechanisms, to attenuate virus pathogenesis, to identify new paradigms of disease causation. We are willing to participate at any level in discussions regarding this important new pathogenic human coronavirus.

Sincerely,

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From: Andrew Snyder-Beattie
Sent: Tuesday, November 25, 2014 6:26 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: NSABB Public Comment

Hello!

Some conservative back-of-the-envelope calculations might help this discussion.

If we assume there is a 1 in 10,000 chance per year of an accident occurring that results in a pandemic, and that the pandemic is typical for flu (infecting some 20% of the world's population), and has a case fatality rate of 0.05%, we get some 700 deaths per year in expectation.

Should we condone an experiment in which 700 people were expected to die per year?

Of course, some of these experiments might push the case fatality rate up by orders of magnitude. The utility of these experiments will need to be exceptional in order to justify thousands of deaths per year (in expectation).

All the best,
Andrew

--
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From: Laura H. Kahn [mailto:lkahn@Princeton.EDU]
Sent: Tuesday, November 25, 2014 10:26 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: NSABB Public Comment

Attn: Carolyn Mosby

NSABB Public Comment

In 2004, the National Academy of Sciences report, "Biotechnology Research in an Age of Terrorism," listed seven "experiment of concern" in which altered microbial agents could pose significant public health risks if released from the laboratory.

The experiments include:

1. Make a vaccine ineffective
2. Confer resistance to antibiotics or antiviral agents
3. Enhance a pathogen's virulence or make a non-virulent microbe virulent.
4. Increase transmissibility of a pathogen
5. Alter the host range of a pathogen
6. Enable a pathogen's ability to evade diagnostic or detection modalities
7. Enable weaponization of a biological agent or toxin

Gain of function studies clearly fall into one or more of these categories and should not be supported by the NIH. I have written about this issue in my online column in the Bulletin of the Atomic Scientists.

Sincerely,

Laura Kahn
http://www.nap.edu/catalog/10827/biotechnology-research-in-an-age-of-terrorism
http://thebulletin.org/going-viral

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NSABB Meeting
November 25, 2014 - 11:00am to 1:00pm

Written public comment submitted by

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Our written comment for the meeting of the NSABB is submitted in the form of an article that has been accepted for publication in *mBio*, the flagship journal of the American Society for Microbiology. It consists of 15 pages including this cover.

Moratorium on Research Intended to Create Novel Potential Pandemic Pathogens

ACCEPTED MANUSCRIPT TO APPEAR in *mBio*

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ABSTRACT: We applaud the US government's funding pause on gain-of-function experiments that create potential pandemic pathogens while deliberation about risks and benefits of such experiments occurs. The risks of some such experiments, which create transmissible strains of highly virulent influenza strains, are so large that a quantitative risk assessment will almost certainly find them unacceptably high. Other types of experiments covered by the moratorium may have different risk profiles. We discuss benefit assessment and emphasize the need for weighing concrete benefits of portfolios of approaches excluding and including PPP experiments against the unique risk of PPP experiments. Other risks, including biosecurity risks in general, and biosafety risks of experiments on coronaviruses and experiments to enhance pathogenicity, should also be quantified. The US plays a leadership role as funder of much of the PPP research at the moment and must seek significant international input to arrive at appropriate policy decisions.

MAIN TEXT

Research on highly pathogenic organisms is crucial for medicine and public health, and we strongly support it. This work creates a foundation of new knowledge that provides critical insights around the world's most deadly infectious diseases, and it can lay groundwork for the future development of new diagnostics, medicines and vaccines. Almost all such research can be performed in ways that pose negligible or no risk of epidemic or global spread of a novel pathogen. However, research that aims to create new potential pandemic pathogens (PPP)[1] – novel microbes that combine likely human virulence with likely efficient transmission in humans -- is an exception to that rule.

While this research represents a tiny portion of the experimental work done in infectious disease research, it poses extraordinary potential risks to the public.

Experiments that create the possibility of initiating a pandemic should be subject to a rigorous quantitative risk assessment and a search for safer alternatives *before* they are approved or performed. Yet a rigorous and transparent risk assessment process for this work has not yet been established. This is why we support the recently-announced moratorium on funding new "gain-of-function" experiments that enhance mammalian transmissibility or virulence in SARS, MERS and influenza viruses. This realm of work roughly corresponds with the work we have termed PPP above. Because the term "gain of function" in other contexts can be used to describe techniques of scientific research that have nothing to do with the creation of novel potential pandemic pathogens, we think the term can be too broad and can mislead. Throughout this commentary we focus on research designed to create PPP strains of influenza, the type of research that initially attracted attention leading to the moratorium and for which the most discussion has already occurred. Other types of gain-of-function research on influenza, and studies intended to enhance pathogenicity or transmissibility of MERS and SARS coronaviruses, may or may not fit the definition of PPP research that we established, and further clarification is needed and ongoing. As we discuss near the end of this article, it will be essential to clarify the different risks and benefits entailed by different types of experiments covered by the funding pause.[2]

The purpose of this research funding pause is to complete "a robust and broad deliberative process…that results in the adoption of a new US government gain-of-function research policy"[3]. The moratorium would stop new funding for:

> *"research projects that may be reasonably anticipated to confer attributes to influenza, MERS, or SARS viruses such that the virus would have enhanced pathogenicity and/or transmissibility in mammals via the respiratory route. The research funding pause would not apply to characterization or testing of naturally occurring influenza, MERS, and SARS viruses, unless the tests are reasonably anticipated to increase transmissibility and/or pathogenicity."*[3]

The new US government policy also encourages the currently-funded US government and non-government research community to join in adopting a voluntary pause on research that meets this gain of function definition. Some 18 NIH research projects have been identified that possibly meet that definition[2]. The moratorium does not apply to the larger infectious disease research portfolio supported by the US government. The announced moratorium does not affect disease surveillance or vaccine development programs. During the moratorium, a deliberative process will occur that will be led by the National Science Advisory Board for Biosecurity and the National Academy of Sciences. This process is intended to produce "recommendations for risk mitigation, potential courses of action in light of this assessment, and propose methodologies for the objective and rigorous assessment of risks and potential benefits that might be

applied to the approval and conduct of individual experiments or classes of experiments." [3]

In this commentary, we discuss key elements of risk analysis and offer an example of an approach that could be taken. We describe benefit analysis, offering an account of the kinds of benefits that are relevant and our own view of those at this point. We note other factors that are important to consider. And we argue that a moratorium is the right approach until a rigorous, objective and credible risk assessment process can be established.

RISK ANALYSIS

Risk assessment for GOF work should be quantitative, objective, and credible. Extensive qualitative arguments have been made on both sides of this issue, and these arguments have not provided sufficient clarity or evidence to resolve concerns or identify a consensus path forward. Quantitative assessments should now be performed so as to provide specific calculations and information to inform decisions. It is also important for these risk assessments to be objective. Given the stakes in this process, the risk assessment process should be directed by those without a clear personal stake in the outcome. Just as peer review of science is performed by those without a direct stake in the outcome, so too should these risk assessments be performed in the same way. The credibility of the risk assessment will depend both on the rigor of the quantitative process and the perceived objectivity of the process.

The record of laboratory incidents and accidental infections in biosafety level 3 (BSL3) laboratories provides a starting point for quantifying risk. Concentrating on the generation of transmissible variants of avian influenza, we provide an illustrative calculation of the sort that would be performed in greater detail in a fuller risk analysis. Previous publications have suggested similar approaches to this problem.[1, 4]

Insurers and risk analysts define risk as the product of probability times consequence. Data on the probability of a laboratory-associated infection in US BSL3 labs using Select Agents show that 4 infections have been observed over < 2,044 laboratory-years of observation, indicating at least a 0.2% chance of a laboratory-acquired infection[5] per BSL3 laboratory-year. An alternative data source is from the intramural BSL3 labs at the National Institutes of Allergy and Infectious Diseases (NIAID), which report in a slightly different way – 3 accidental infections in 634,500 person-hours of work between 1982 and 2003, or about 1 accidental infection for every 100 full time person-years (2000 hours) of work. [6]

A simulation model of an accidental infection of a laboratory worker with a transmissible influenza strain estimated about a 10-20% risk that such an infection would escape control and spread widely.[7] Alternative estimates from simple models range from about 5% to 60%. Multiplying

Probability of an accidental laboratory-acquired infection per lab-year (0.2%) or full-time worker-year (1%)
X
Probability the infection leads to global spread (5%-60%)

provides an estimate that work with a novel, transmissible form of influenza carries a risk of between 0.01% and 0.1% per laboratory-year of creating a pandemic, using the Select Agent data, or between 0.05% and 0.6% per full-time worker-year using the NIAID data.

Readily transmissible influenza, once widespread, has never before been controlled before it spreads globally, and influenza pandemics historically have infected about 24-38% of the world's population[8, 9]. The case-fatality ratio of a novel strain is of course unpredictable. The worst case might be a case-fatality ratio similar to that of avian H5N1 influenza in people, which approaches 60%.[10] A greatly attenuated version of the same virus might have a case-fatality ratio of "only" 1%.
Again, multiplying

Pandemic attack rate (24%-38%)
X
Global population (~7 billion)
X
Case-fatality ratio (1%-60%)

would produce an estimate of between 2 million and 1.4 billion fatalities from a pandemic of a highly virulent influenza strain.

Putting all these numbers together, the Select Agent data suggest that a laboratory-year of experimentation on virulent, transmissible influenza might have an 0.01%-0.1% chance of killing 2 million-1.4 billion, or an expected death toll of 2000-1.4 million fatalities per BSL3-laboratory-year. From the NIAID data, for each full-time person-year of BSL-3 work we might expect a toll of between 10,000 and over 10 million.

These numbers should be discussed, challenged, and modified to fit the particularities of specific types of PPP experiments. For creation of novel, transmissible, virulent influenza strains, they may overstate the risk for the following reasons: 1) most work is done in BSL3+, which may be safer than BSL3; 2) control measures, including vaccination and antiviral prophylaxis of laboratory workers, might reduce the risk of infection and of spread, although none of these is perfect; 3) the human case-fatality ratio of an avian influenza strain that gains transmissibility could be below 1%; 4) transmissibility in laboratory animals does not necessarily indicate transmissibility in humans[11, 12]; 5) novel strategies of molecular biocontainment[13], if employed, might reduce the risk of human transmission of a strain used in transmission experiments in other mammals.

On the other hand, these numbers may understate the risk because 1) the Select Agents calculation includes in its numerator only BSL3 labs, but in the denominator BSL3 as well as BSL2 and BSL4 "registered entities" as separate figures for BSL3 are not publicly available[5]; 2) the rate of accidents is calculated for US labs, while GOF experiments are performed in many countries; if this work expands to some of the many countries with less stringent standards than the US[14], risks could be higher; 3) the costs of an accidental pandemic considered here are deaths only, but additional losses would include scientific credibility, nonfatal health outcomes, economic and educational losses, etc.

The illustrative calculations above show that approximate risk estimates are possible for creation of PPP strains of influenza. During the deliberative process initiated with this moratorium, the risk assessment approach that is established should be able to provide calculations that reflect these and other available probability and consequence estimates and take into account the range of modifying factors including those just described. The risk assessment process should also be able to provide calculations related to PPP experiments where the risks are harder to calculate given more limited data, such as enhancement of coronavirus pathogenicity in small mammals.

BENEFIT ANALYSIS

On the surface, analyzing the benefits of PPP experimentation would seem more difficult. In the cumulative process of knowledge acquisition that is science, it is hard to see far ahead where a particular type of research may lead. On the other hand, scientists make judgments about the relative merits of experimental approaches on a daily basis in their roles as investigators and grant reviewers. Doing and funding science is a process of severe winnowing (especially severe in today's tight funding climate) in which we choose to pursue one approach and not to pursue others based on judgments of which approaches are expected to have lowest cost, highest probability of success, and greatest yield of valuable findings, among other considerations. Implicit in this process is the idea of opportunity cost. In prioritizing the week's or the year's research work, we do not judge in isolation whether a particular experiment should be done or not done. We decide how to allocate our time and funding among possible approaches, devoting resources to the portfolio of efforts that seems most promising. Similar prioritizations are made by funders when they decide which kinds of research will be funded, and which research will not.

The analysis of benefits of PPP experiments should follow this familiar approach. The choice is not: do PPP experiments or do nothing. Rather, the appropriate question is: within a portfolio of scientific and public health activities designed to understand and combat influenza or a coronavirus (or, perhaps, a broader subset of infectious diseases), what are the benefits of including PPP approaches compared to the benefits of expanding other parts of the portfolio to use the resources in another way? From the perspective of public health and the practical goal of preventing and treating flu,

alternative approaches include those which, like PPP experiments, seek to enhance our scientific understanding of biology, pathogenesis and transmission. Alternatives also include efforts to develop treatments and prevention measures, including surveillance, through means other than improving our basic biological understanding of influenza.[4] This approach is shown graphically in Figure 1, which also depicts the risks of PPP research. Such risks should be weighed against the risks of alternatives, which are typically much smaller or even negligible. Figure 1 embodies the idea PPP research should be a component of our research portfolio only if devoting resources to PPP studies at the expense of alternatives has net benefits that outweigh the unique risks of PPP studies.

This comparative approach to benefits should be informed by a hardnosed look at the benefits that are readily achievable by PPP experiments, not to hypothetical outcomes that could someday lead to unspecified benefits. We acknowledge the possibility that PPP experiments may lead to benefits we cannot today envision. But so could the experiments that are done in their place if support for PPP is reallocated to other scientific approaches. The possibility of unanticipated benefits is surely a reason to do science, but it is not a reason to favor PPP approaches over others, unless some specific case can be made for the unique yet unanticipated benefits of PPP work. Such a case seems hard to imagine for benefits that are by assumption unanticipated.

For example, it has been suggested that mutations or phenotypes identified through PPP experiments could be used to sort through the massive diversity of nonhuman influenza strains to prioritize those that should trigger countermeasures, including pre-pandemic vaccine manufacturing. While this might be is possible in principle, there are many practical barriers to achieving public health benefits of this sort from PPP studies.[15] Lists of mutations, and even phenotypes, associated with PPP studies, can be compiled and compared against isolates of influenza from birds and other nonhuman sources[16]. We know that these lists are unreliable and can even be misleading: the mutations in hemagglutinin identified by two prominent PPP experiments on H5N1 do not reliably confer human receptor specificity even on other H5N1 viruses[17]. The E627K mutation in the PB2 gene, known as a virulence and transmissibility determinant before GoF experiments[16, 18, 19], found repeatedly in GoF experiments in H5N1[20, 21], and used for pandemic risk assessment in H7 viruses[16], was found in some isolates of the H1N1pdm strain in 2009, leading to concern about possible increased virulence and transmissibility. Yet it conferred neither trait in this genetic background.[22]

At the present time, the high levels of epistasis – dependence of phenotype on the genetic background on which a mutation is found – make prediction of pandemic risk for any given strain more of an art than a science. Indeed, the very presumption that we will see human cases of an incipient pandemic before that pandemic occurs has never been met in practice[23]: we have never observed zoonotic cases of any flu virus before it caused a pandemic. This is not to deny that PPP experiments provide any useful data for surveillance and prioritization. Rather, it is to say that other approaches can also identify

such predictors (as in the case of the PB2 mutation[11, 13, 14]) and that the ability to use markers of putative transmissibility or virulence to make reliable predictions remains far in the future.[23] The fact that some analysts consider mutations identified in PPP experiments when assessing threats of viruses found in surveillance does not mean that the use of such mutations improves the predictions, a claim for which we have no evidence because no pandemic strain has ever been identified in advance. The analysis of benefits of PPP creation should reflect this state of science.

According to some proponents, the most valuable scientific finding of experiments to make ferret-transmissible mutants of influenza A/H5N1 is the definitive proof that such variants could be produced with a small number of mutations. This could not be definitively proven without doing the PPP experiment to manufacture a potentially pandemic variant of H5N1[24]. While it is now undeniable that a ferret-transmissible mutants of influenza A/H5N1 can be created experimentally, the impact on scientific opinion about the risk of a pandemic from H5N1 has been hard to gauge. Prior to the gain-of-function experiments there was a wide range of expert opinion on the likelihood of an H5N1 pandemic [25]. Some influenza experts questioned whether H5N1 was a major pandemic threat. After the publication of the experiments producing potentially pandemic H5N1, one prominent member of this group, Peter Palese, noted the shortcomings of the ferret model for humans and correctly concluded that the question of whether H5N1 can transmit efficiently in people remains unsettled[26], as it must until the phenomenon is directly observed in nature. From a practical perspective, responsible policy makers and public health leaders should have been planning for the possibility of H5N1 pandemic before PPP experiments on H5N1 were undertaken. In some countries of the world they were making stockpiling vaccines against H5N1[27, 28] and making plans for nonpharmaceutical [8] interventions in the event of a pandemic. The same remains true after the experiments. We have observed no discernible influence of the H5N1 PPP experiments on H5N1 policy preparations.

CALCULATING OTHER FACTORS

During the moratorium, progress should also be made in calculating the risks associated with potential deliberate misuse of PPP strains and with potential deliberate misuse of the information that is created and published following PPP experimental work. This calculation should take into account the possibility of deliberate theft and dissemination by either persons working within a lab or theft by those outside the lab. While the probability of this is likely to be very low for most scientists and most laboratories, it is not zero. There is precedent of scientists using pathogens from their own labs to cause harm. And as with potential accidents, while the probability may be very low, the consequences could be very high.

This assessment should also take into account the possibility that scientists may deliberately misuse the knowledge gained and published following the experiments by recreating the novel PPP strains in another laboratory using methods from published papers and then purposefully disseminating it. This possibility is typically dismissed out of hand by many scientists. But before dismissing that possibility, an analysis by an assembly of experts in the best position to make that judgment should be conducted. What is the possibility that individuals or groups who would seek to carry out such an act would develop the capacity and skill to carry it out? Given that once knowledge is published, it will be available forever, these questions are not just about the possibility of this happening in today's world, but also anytime in the future. Despite the inherent uncertainties in trying to answer these questions, they should be answered with the best possible expertise.

Similarly, the moratorium should be used as a time to answer, or at least be addressing, another major issue as well: the international approach to funding, authorizing and overseeing PPP. An accident or deliberate act involving PPP anywhere in the world could conceivably impact the public around the world. Therefore, the community of nations has an abiding interest to set common rules for how this work will be pursued. However at this point, few countries have begun any kind of deliberative process on an approach to research with these unique dangers. Country X should have the right to know if this work is going on in Country Y, and if yes, what is being done to ensure it is done with the greatest safety and security. But currently, the way Country X finds out about PPP work being done elsewhere in the world is when it is published in a science journal. Given the prestige that some scientists have received for pursuing PPP research, it would be surprising if scientists from countries around the world did not increasingly pursue it. As comparatively less experienced labs decided to pursue this work, this will increase potential dangers.

A MORATORIUM IS THE RIGHT STEP

There are prominent scientists who agree that there are potential serious dangers to this work and agree that a risk assessment process is needed, but who are opposed to a moratorium being imposed while such a the risk assessment process is undertaken. They believe that a moratorium should be avoided for reasons that include the potential damage it can do to the funding and work of that lab, as well as to the careers of those involved in the work.

We have a different view. A substantial number of scientists agree there are extraordinary potential consequences of the work.[15] There is no rigorous, objective, credible risk assessment process to judge the risks and benefits of proceeding with it. We believe that the responsible course is to take a research pause until such a risk assessment process is established which creates a stronger basis for decisions and actions. This is not solely a scientific issue. It is a scientific, public health and safety issue, and it is an issue where the public itself has an abiding interest.

We have no interest in stopping scientists from doing their work or preventing laboratories from receiving funding. The narrow and defined area of GOF research intended to create novel potential pandemic strains should be put on pause until the risk assessment process is completed. The same laboratories and scientists whose work has been stopped by the moratorium are free and able to pursue all other avenues of infectious disease research except for that narrowly defined by the GOF definition in the new policy; to the extent that other activities not meeting the narrow definition in the pause have been included in letters to principal investigators ordering or requesting work stoppage, the boundaries of the funding pause should be quickly clarified to allow important alternative work on flu to continue. We note that there are over 250 NIH-funded projects listed as active with titles containing MERS, SARS, coronavirus, or influenza[29] of which 18 have been affected by the funding pause. The number that remain on pause may be further reduced by negotiations between investigators and the NIH that are now underway that will define which projects truly are within the scope of the moratorium vs. those that do not meet its terms and can resume.

The character and scope of the risk assessments that are applied is important. To establish methodologies and approaches for risk assessment and risk mitigation for this context, it would be valuable to start with a global assessment of the risks and benefits of this realm of research, identifying the common aspects of risk and benefit within PPP experiments and other approaches covered in the funding pause. For example, any risk assessment should include estimates of the probabilities of accidental infection and extensive spread, as well as estimates of the impacts of these events should they occur. The specific values of these estimated parameters will differ for different types of experiments. It will then be necessary to set standards and expectations for the quality and characteristics of risk-benefit assessments for individual experiments, for example to distinguish coronavirus research from influenza research, enhancements of pathogenicity from enhancements of transmissibility, and other important distinctions. Given that the term “risk assessment” is used to mean different things by different people, an agreement on an approach to individual risk assessments would be needed to ensure rigor and credibility. Once this kind of analytic structure is established, individual risk assessments on GOF experiments that meet the definition in the new USG policy[3] should become the norm before such experiments are funded. Crucially, this process should be quantitative, rather than relying on unquantified and unverifiable assurances that particular laboratories are safe.

CONCLUSIONS

The results of this risk assessment process are not only important to the US Government -- which had been a major funder of PPP experiments -- but also to other funders, regulators, and investigators worldwide who consider such experiments. Our support for the funding pause and associated deliberative process does not indicate that we

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would support a permanent end to all experiments subject to the pause. There may be research endeavors that are subject to the moratorium that have a risk-benefit profile sufficiently favorable to justify their resumption, once risks and benefits have been explicitly set forth. After two years of debate, we think the balance is evidently unfavorable for experiments to enhance avian influenza transmissibility, but other classes of experiments may be different. In the meantime, the moratorium is an appropriate and responsible step while dedicated and rigorous efforts are made to understand the risks and benefits of this work.

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29. US National Institutes of Health. http://projectreporter.nih.gov/reporter.cfm accessed November 18, 2014.

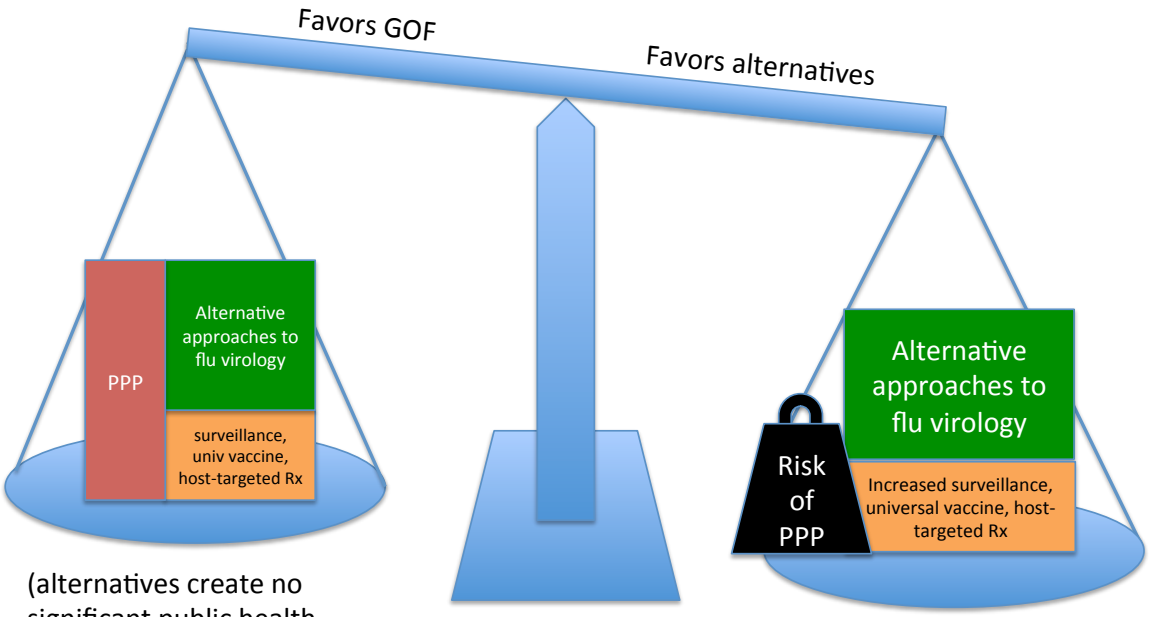

FIGURE: Weighing risks and benefits. The benefits (squares) of spending a fixed quantity of resources on a portfolio of activities including PPP research (red), other approaches to influenza virology (green), and other public health activities to defeat influenza (yellow), should be weighed against the benefits a portfolio in which the other activities are expanded to use the resources freed by not supporting PPP activities, reflecting the opportunity cost of the PPP research. If there are net benefits to including PPP activities in the portfolio, then they should be weighed against the net risks created by PPP experiments, which in the case of influenza transmissibility enhancement we have argued (see main text, RISK ANALYSIS) are exceptionally high. The balance may differ for other activities, but this comparison of benefits of portfolios with and without gain-of-function experiments is the appropriate comparison, with any net benefits weighed against net risks.

From: Gary R. Whittaker [mailto:gary.whittaker@cornell.edu]
Sent: Tuesday, December 02, 2014 8:56 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: NSABB Public Comment

Dear NSABB members

I would like to offer the following comments regarding the gain-of-function funding and research pause for influenza, MERS-CoV and SARS-CoV. I am writing from both the perspective of a researcher who has studied these, and other viruses, for many years, and also from the general perspective of a researcher interested and engaged in issues of biosafety.

My principal comment is that the guidelines still lack clarity and encompass an over-broad category of "gain-of-function". As noted by Dr. Steele during the public comment following the recent teleconference, many "gain-of-function" experiments that are currently covered by the pause are of low or negligible risk. Further clarity is urgent needed to avoid unnecessary burden and impact on both researchers and administrators. As noted by Dr. Denison during the discussion period, there is a large body of evidence that adaptation of (human corona)viruses to mice results in "gain-of function" and increased pathogenesis in that particular species, but likely the opposite effect in humans. The same reasoning clearly applies to influenza viruses adapted to mice (which are not a natural host). Without more specific wording, the current guidelines include any mammal. I would suggest that studies of influenza, MERS-CoV and SARS-CoV in mice (or other rodents) are inherently low risk and can be exempted until final guidelines are made. During the teleconference, Dr. Lipsitch commented that there are alternative ways to make mouse models (i.e. introduction of a transgene into the mouse) but I would argue that robust mouse models of human virus infection need both modification of the animal and adaptation of the virus. In fact, the introduced virus will always adapt to the new species by a process of natural selection anyway. So virus adaptation is likely a necessary part of development of any animal model, whether it is intended by the researcher or not. Another area of clarity may be to identify "designer" gain of function, whereby a researcher may make targeted changes in a recombinant virus in a "high risk" species (arguably gain-of function in humans), rather that the virus adapting naturally to a "low risk" species (arguably loss of function in humans).

A similar situation arises with influenza in other species. As a professor at a major veterinary college, I would like to note that it is important to consider that (non-human) influenza is an important natural pathogen in certain animal species; principally horses and dogs. As with mice, I would argue that adaptation studies on equine or canine influenza viruses in these species pose little or no risk to humans, and can be included in an interim exemption. The present guidelines could be interpreted to include these important veterinary pathogens with negligible risk to human health, and so have an unnecessary impact on veterinary surveillance and research. In contrast experiments in other species, where there is the realistic likelihood of cross-species transmission into humans (e.g. pigs and ferrets), might be considered high risk.

In summary the designation of "any influenza…." and "(any) mammal" is overly broad, and needs further clarity. I ask that new, risk-based, interim guidelines be issued as soon as possible, and not within the currently projected one year time-line of the deliberations. Lack of clarity in the overly broad current guidelines is imposing significant burden on both researchers and administrators. This comes in addition to the comprehensive federal and institutional biosafety regulations and oversight already in place. An

unnecessary pause on funding or research may have significant consequences for junior faculty and graduate students who need to consider tenure and promotions deadlines, or need to complete dissertations. The current guidelines many also unnecessarily impact many other researchers who are struggling to maintain their research labs in times of limited financial resources. I would ask that new interim guidelines issued within 30 days for the benefit of all involved parties.

Gary Whittaker
Professor of Virology

C4127 VMC
Microbiology & Immunology
Cornell University
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http://instruct1.cit.cornell.edu/research/whittakerlab/

From: Derrin Culp
Sent: Tuesday, January 06, 2015 11:48 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Public Comments Regarding the "Deliberative Process"

January 6, 2015
TO MEMBERS OF THE NATIONAL SCIENCE ADVISORY BOARD FOR BIOSECURITY:
I am a retired private citizen and a non-scientist who never worked in any microbiological enterprise. However, I have published brief pieces addressing insider threats in pathogen research and the risks of a "brain drain" from further regulation of GOF research. Having watched the October 22 NSABB meeting and the subsequent National Academies GOF workshop in their entirety, I am submitting some observations regarding the upcoming risk/benefit assessment.
1. The USG's regulatory frameworks for biosafety and biosecurity already contain express restrictions or "guidelines" on the kinds of facilities in which research with enumerated pathogens may occur and on the security screening and monitoring of scientists working with certain pathogens. Certain Select Agents (and maybe other organisms as well) are restricted to BSL-3 and BSL-4 facilities, and scientists and facilities that handle "Tier I Select Agents" are subject to personnel screening and monitoring that do not apply to other pathogens.
Nobody in the USG or the microbiology profession will assert that the existing biosafety and biosecurity regimes are fool-proof, and there is hardly a soul who would claim that LAIs, other biosafety accidents, or security breaches cannot occur, even absent human error. Anyone who acknowledges that there is even an infinitesimal risk that a dangerous pathogen may escape laboratory containment is engaging in an implicit—and perhaps unconscious—risk/benefit assessment: something COULD possibly go wrong, but the harm that could possibly arise is not great enough to further restrict research.
If you have not already, it would be worthwhile to establish whether the USG, in deriving its existing restrictions or "guidance" for microbes that must/should be researched only within particular biosafety level laboratories, engaged in any kind of deliberate risk/benefit assessments:

- Did the USG consider the potential impacts on scientific progress and public health that might arise from limiting research with certain pathogens to the highest BSL labs?

- Did the USG rigorously analyze the potential consequences of an organism escaping BSL-3 or BSL-4 containment, regardless of how small they believed that risk to be?

- Likewise, in the 2011-2012 rulemaking process for the Select Agent Regulations, what kind of risk/benefit assessment (if any) did the USG perform to determine that the incremental biosecurity protections included in the FINAL Rule (1) need apply only to Tier 1 pathogens, AND (2) could be significantly less restrictive than the protections proposed in the DRAFT Rule?

The answers to these questions potentially could help guide your deliberations regarding the contemplated risk/benefit assessment, but also clarify if you are on the verge of potentially establishing a novel (and substantially more onerous) precedent for pathogen research risk/benefit assessment—a precedent that may have implications for research far beyond the scope of NSABB's charge in this instance.
2. Much of the debate about the "risk" side of the research that is subject to the Pause consists of disagreements about the adequacy of the existing biosafety regime and what the available data on LAI and other biosafety failures are--or are NOT--telling us about the actual pandemic potential of laboratory manipulated organisms. That is critical, but I urge you not to give short shrift to the

biosecurity element of the risk component. If it is a legitimate line of inquiry to consider whether certain types of research with PPP should occur only in, say, BSL-4 laboratories, it is equally legitimate to consider whether those types of research should (a) entail more restrictive personnel screening and monitoring than those currently applicable to research with TIER 1 Select Agents (b) be done only in military or classified environments, and (c) be authorized and funded subject to explicit restrictions on publication and dissemination of findings . Conceptually simple engineering fixes on the biosafety side should not squeeze out conceptually difficult enhancements on the biosecurity side.

3. I commend to you a forthcoming article from the American Academy of Arts and Sciences, tentatively titled "Insider Threats: Lessons from Amerithrax." This article arises from an Insider Threats Workshop held last May as part of the Academy's Global Nuclear Futures Project. Perhaps the administrator of the project, Francesca Giovannini (fgiovannini@amacad.org) might be willing to provide NSABB with a non-attribution draft.

4. I urge you to resist pressures to propose solutions to the international dimensions of research with potential pandemic pathogens. The numerous speakers at the two meetings who emphasized the need for an international response are undoubtedly correct, but that doesn't mean NSABB has the time or the wherewithal to derive a practical solution. If you even think about trying to lay out the parameters of an international regime to address this issue, you will fritter away the resources you need to define the choices available to the USG in terms of the domestic facilities over which it has direct jurisdiction. Conversely, you will do an immense service for USG policy makers if you simply clarify the international dimensions of the issue and propose further lines of investigation.

The USG has a limited number of tools (military, diplomatic, economic, trade) with which to influence the behavior of sovereign governments, and the USG always exercises those tools in the context of multiple, often conflicting foreign policy objectives. Far better for NSABB to spell out the implications for the United States of other nations allowing certain kinds of research to proceed without the level of protections deemed necessary in the USA, than to propose **how** the USG should attempt to bring other nations in line with our approach. I urge you to consult with experts who previously have gone down this road and who can give you a realistic sense of the challenges and limited potential payoffs: the University of Pennsylvania's Harvey Rubin and his proposed "Global Governance Structure for Infectious Disease" (http://www.istarpenn.org/category/istar-news-on-the-move/); DHS's Gerald Epstein (http://csis.org/files/media/csis/events/061130_btr_brief.pdf); and Princeton's Chris Chyba (who within the last decade has written and spoken on "Proposed International Regimes for Regulating Biotechnology Research").

5. Finally, I urge you to request a presentation on the history of nuclear weapons accident risk assessment from Eric Schlosser, the author of Command and Control: Nuclear Weapons, the Damascus Accident, and the Illusion of Safety. Schlosser's five-decade appraisal of how bench scientists, bureaucrats and political officials discussed and assessed risk in an enterprise where the consequences of a safety failure could be of the catastrophic magnitude that preoccupies supporters of the Pause, is a compelling argument NOT to have great faith in scientists' or government officials' assertions that an accidental or deliberate release of a modified pathogen is "extremely unlikely" to occur in the United States or Europe.

Thank you for your attention to these comments.

Sincerely,

Derrin Culp
48 Ogden Avenue
White Plains, NY 10605

From: Dustin Phillips
Sent: Monday, January 12, 2015 11:11 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Please permanently halt research to turn avian flu into a more deadly virus.

I wanted to contact you and express my concern with research to turn avian flu into a more virulent strain. Though, undoubtedly, much can be learned from such research, there are safer ways to research the virus without the, albeit low, risk of worldwide catastrophe. Even small risk is not worth taking when the states are so high.

Thank you for hearing me,

Dustin Phillips
Louisville, KY

From: Richard Adams
Sent: Monday, January 12, 2015 12:05 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Cc: Ana Adams
Subject: stop gain-of-function research on pathogenic viruses.

To Whom it may concern,

As a deeply concerned citizen and a human being, I demand a permanent halt to gain-of-function research on pathogenic viruses. Such 'science' is hubris, and it is a fantasy that the results can be 100% contained, for sure, forever. Whether by accidental release, or by serving as a recipe base for bio-terrorists, the risks from this technology, which is in essence the same as bio-weapons research, is not worth any supposed research benefits.

http://www.forbes.com/sites/stevensalzberg/2015/01/12/flu-experiments-may-cause-2000-deaths-per-year/

Thank you very much,

- Richard Adams

Richard S. Adams

From: Kimball Ward
Sent: Monday, January 12, 2015 1:04 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: It's time to end Gain of function research

I have followed this issue since the publication of the gain of function (GOF) research on the influenza virus by Fouchier and Kawaoka, including the temporary hiatus after publication when many people, both in and out of the scientific and human immune system research communities. The hearings in December were especially telling: despite many of the panelists being in favor of GOF research, the presenters against made much more convincing arguments.

The primary reason this type of research should be discontinued:

1) The risks outweigh the potential gains.
We know lab viruses can escape containment because it has happened. Professor Lipsitch gave an excellent presentation on this point. He also provided other less risky methods for doing research that would yield similar results.

2) The main claim of GOF research supporters is that it will prepare us to deal with a future Flu pandemic.
This argument was fairly convincingly debunked this year. We deal with the flu virus every year, and attempt to predict how strains will mutate and which will be prevalent the following year so we can prepare effective vaccines. Despite all our knowledge, the vaccine we prepared was fairly poor in terms of overall effectiveness. That gives little reason to believe that GOF research will somehow accurately predict what the virus will do in the future; instead, we risk creating the kind of nightmare we claim to be trying to prevent.

The great thing about science is that we can make informed decisions based on the available evidence. In this case, the evidence against GOF research is far outweighed by the evidence for. For these reasons, I am writing to you to express my desire to see an extended hiatus on GOF experiments.

Kimball Ward

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All our actions take their hue from the complexion of the heart, as landscapes their variety from light.
~ Francis Bacon

From: Billie Sellers
Sent: Tuesday, January 13, 2015 10:53 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Re: Gain of Function Research on Pathogenic Viruses.

Please recommend a permanent halt to gain-of-function research on pathogenic viruses. This research must be banned.
It is far too dangerous to continue and is totally unnecessary.

From: Shannon Scott
Sent: Thursday, January 15, 2015 4:02 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: GOF

Please please PLEASE make the "pause" on the gain of function research on the Avian flu virus permanent. If it's extremely risky and not any more or less effective at helping us then please don't keep doing it, ditch GOF research for less risky and more cost effective methods.

From: Daniel O'Connell
Sent: Sunday, January 18, 2015 11:22 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Gain-of-Function Research

Please recommend a permanent halt to gain-of-function research on pathogenic viruses. This research is unnecessary and risky.

Daniel O'Connell
Albany, Oregon

From: Peter Murakami [mailto:pmurakam@jhu.edu]
Sent: Wednesday, January 21, 2015 7:38 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject:

Dear NSABB,
I am writing to you to urge you to permanently halt to gain-of-function research on pathogenic viruses. Work like that of virologists Ron Fouchier and Yoshi Kawaoka pose much greater risks than rewards. There are far better things to do with our research funds.

Sincerely,
Peter Murakami

Baltimore, Maryland

From: Denise Hein
Sent: Saturday, January 24, 2015 6:36 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: GOF studies

I am contacting you to recommend a permanent halt on further GOF studies.

Please advise me how to get my voice heard on this issue.

Thank you.
Denise Hein

Detailed response to Fouchier's criticism of my mBio comments

To: *The National Science Advisory Board for Biosecurity, in advance of the May 5, 2015 meeting*

From: *Lynn C. Klotz, PhD*
Senior Science Fellow
Center of Arms Control and Non-proliferation,
Washington D.C., USA

My comments on Dr. Fouchier's calculation that "1 LAI would be expected to occur less frequently than once every 1 million years" were published on April 14 2015 in mBio. Fouchier's response to my comments was also published there.

His response is problematic in several ways. In addressing the problems, I will quote frequently from my comments and from his response to make sure it is clear what was said.

The biggest problem is that Dr. Fouchier does not once address my calculation of potential fatalities and fatality burden that employs his low probability of an undetected or unreported LAI escaping from his laboratory. Instead, he chooses to argue against my peripheral comments that his probability is likely much too low. His focus unfortunately pulls attention away from my calculation that finds intolerable potential fatalities and fatality burden.

Detailed comments on Fouchier's criticism

I am numbering the comments to keep each point separate.

1. Dr. Fouchier seems to misunderstand my arguments that his formula, $y=1/P_1$, is too simple. I questioned the meaning of his calculation:

> "Does it give us the elapsed time for a 10% chance that an LAI occurs? Does it give us elapsed time for a 50% chance, or an 80% chance? In this regard, the elapsed time for a 100% chance is infinite, as we can never be absolutely certain that an LAI will occur."

Solving the equation $E = 1 - (1-P_1)^{yn}$ for y, gives $y = (1/n) \times \log(1-E) / \log(1-P_1)$, a better equation for calculation of elapsed time when likelihood or chance of an escape must be considered. Derivations of these equations may be found in the Appendix at the end. The equations and derivations are not necessary to understand the arguments here. They are included only to further document some of my arguments for those who have a basic understanding of algebra and elementary probability.

In reply, Fouchier writes:

> "In calculations of the probability of a community LAI ("E"), Dr. Klotz further assumes that transmission studies in the Erasmus MC facility will be performed for a period ("y") of 1 million years. I am hopeful that our research enterprise will have reached solid conclusions on determinants of airborne transmission a bit sooner."

Rhetorical quip aside, neither his one-million-year calculation result nor my questioning of it implies or assumes in any way that research must be performed for a given number of years. My questioning and my alternative equation are simply a comment on his methodology. It was he who brought up this time frame in the first place: "1 LAI would be expected to occur less frequently than once every 1 million years." Elsewhere in my comments, I assume the research enterprise will be concluded in ten years, as does he.

2. I agree with Fouchier that probability of escape from a laboratory "is the key challenge in this debate." Acknowledging this uncertainty, I use the words 'arguments' and 'likely': "arguments as to why the Fouchier value for P_1 is likely much too low."

Dr. Fouchier writes:

> "Dr. Klotz suggests that incidents at the U.S. CDC laboratories and the long history of escape of LAI agents and other escapes from laboratories show that my estimates of the likelihood of LAIs occurring at the Erasmus MC facility are too low."

The CDC's shipping of an H5N1 contaminated sample to USDA and similar incidents shows the importance of not underestimating human error, especially if one considers the influenza lab at the CDC to be one of the top federal labs in the country. Although biosecurity measures have improved greatly over the years, human nature has not. Laboratory accidents will happen and laboratory workers will get infected, not realize it or not admit it, and take the infection home. The Achilles' heel in Fouchier's argument is that no number of safety procedures can provide for human error.

While the history of escapes should make us worry that the probability may be much higher, the difference between Fouchier and me is moot here since I employ his low probability in my calculations.

3. Dr. Fouchier writes:

> "Dr. Klotz proposes to multiply the low likelihood of LAIs by 300, based on an estimated 30 laboratories involved in the "whole research enterprise" for 10 years, and assumes that part of this research enterprise may lack the rigorous safety practices in place at Erasmus MC. Both assumptions are wrong, to the best of my knowledge; just over a handful of laboratories have worked on airborne transmission of avian influenza viruses, each of which has rigorous safety practices in place."

Our disagreement here is because we define "whole research enterprise" differently. I define it as research on pathogens subject to the NIH funding pause (influenza and SARS category pathogens). He defines it as only influenza research. I implied that the whole research enterprise includes the other pathogens by picking the number 15 for NIH's 15 projects subject to the pause. Perhaps I should have been explicit by listing the pathogen categories as I have just done. In addition, some of the laboratories throughout the world conducting this research that are not funded by NIH may have lax safety standards.

Thus, both of my assumptions are likely correct.

4. Dr. Fouchier writes:

> "Another key aspect is that Dr. Klotz estimates the likelihood of onward transmission from a case of LAI as 0.1 (10%), in contrast to my justification for an adjusted likelihood of $<1 \times 10^{-5}$, based on the specific conditions under which the research is performed, without providing a rationale for that important deviation"

I certainly do provide a rationale for the 10% through references (8) and (9) to risk assessment studies:

> "Summarizing the literature, Lipsitch and Inglesby estimate the probability that a community LAI leads to a global spread (pandemic) to be 5 to 60%. This range is consistent with the 5 to 15% range found by Merler and coworkers (8) and with the 1 to 30% range found in a focused risk assessment (9) for infection spread beginning on crowded public transportation."

Furthermore, there is a rather arcane subject in probability theory, branching theory, which allows prediction of the likelihood of uncontrolled spread of any pathogen based on its R_o value and the variance to mean of the R_o. A large variance to mean would occur due to super spreaders, for instance some people infected with SARS. For a wide range of R_o values, Lipsitch and coworkers have calculated the probability of uncontrolled spread (see figure 4a in their study). For a single infected individual with $R_o = 2$, the probability ranges from 10% (spread of R_o's) to 80% (uniform R_o).

Thus, the pandemic likelihood from a single infected individual is potentially large. I suspect that future risk assessments will confirm that once a highly contagious potential pandemic pathogen escape occurs, the probability of an uncontrolled outbreak is significant.

Fouchier mentions vaccination and antivirals as factors that reduce onward transmission. Antivirals would not be prescribed for undetected LAIs. Vaccines may reduce viral replication in the index case, but active virus may still be present when the infected person leaves the laboratory potentially infecting unvaccinated persons. The annual flu vaccine is sometimes less than 50% effective, so it is unclear if vaccinated laboratory workers are protected by the laboratory vaccine strain.

I would classify vaccination and antivirals, effective or not, as inside laboratory measures. But if an LAI escape occurs, clearly these measures were not effective in preventing the undetected or unreported LAI.

Again, we come back to the probability of escape from a laboratory as the key challenge in this debate.

Once an undetected or unreported LAI from a highly contagious pathogen escapes from the laboratory, it is out of Fouchier's control. Its global spread will depend on the reproductive number, R_o, and other factors external to Fouchier's laboratory.

Fouchier claims that "the viruses are ferret-adapted rather than human adapted," which could lead to a lower R_o in humans. Among the different mutated viruses presumably under development in his

laboratory, some could be highly transmissible and deadly in humans. We will never know for testing them on humans is, fortunately, unethical. Of course if one escaped…

The argument of being ferret adapted and not human adapted is misleading. First, it cannot be proved. Secondly, Fouchier's own work may have already brought an avian H5N1 virus far closer to successful replication in humans. If such a virus escaped from his laboratory, it may well adapt within the individuals in the early transmission chain and then take off in a big way. Dr. Fouchier and the field do not have the knowledge to know just how short of a successful virus they have engineered. That is why they are doing this work.

5. Dr. Fouchier concludes

> "Finally, Dr. Klotz describes the (apocalyptic) scenario of an influenza pandemic with 140 million fatalities based on a 10% case-fatality rate in 20% of the world's population. These numbers not only ignore the scientifically justifiable counterarguments raised before (2) but also are at odds with the documented influenza pandemics of the past. In my view, the "gain-of-function" debate has suffered from the apocalyptic scenarios that are provided as factual whereas they provide estimates that are far beyond the observed worst cases (8)."

It is estimated that the 2009 pandemic influenza infected 20% of the world population. The 1918 H1N1 "Spanish" flu killed perhaps 2% of its victims. The H5N1 avian influenza virus, the subject of Fouchier's research, kills about 50% of those who are infected through direct contact with poultry. The scenario I use as an example represents a combination of these three <u>real</u> events. While this scenario has not yet and may never occur in nature, it is a possible scenario perhaps more likely from a laboratory escape.

Since the consequences of most scenarios, even one on a par with seasonal influenza– several hundred thousand deaths – would be catastrophic and unacceptable, it behooves us to be exceedingly careful in deciding which potential pandemic pathogen research should be allowed. For much of this research, the potential risk far outweighs the potential benefits.

APPENDIX

Derivation of equations for years to a lab escape

Let P_1 be the yearly probability of escape of a pathogen from a single lab. The first question to be asked is "What is the probability of at least one escape from one of the n labs conducting research on the pathogen for y years.

The probability of no escapes in y years for a single lab is

$$\text{prob (no escape)} = (1-P_1)^y \qquad (1)$$

For y years *and* n labs

$$\text{prob (no escape)} = (1-P_1)^{yxn} \qquad (2)$$

The probability, E, of at least one escape in y years from one of the n labs is

$$E = 1 - (1-P_1)^{yxn} \qquad (3)$$

How much risk are we willing to tolerate; that is, what value of E is too high a risk? E=1%, E=10%? E=50%? E=80%? The level of risk we are willing to tolerate is subjective. A related question is: At our risk tolerance level, how many years y of research in the N labs will it take to exceed our risk toleration? Solving equation (3) for y, will allow this question to be answered.

$$\log (1-E) = \log(1-P_1)^{yxn} = y \times n \times \log(1-P_1)$$

$$y = (1/n) \times \log(1-E) / \log(1-P_1) \qquad (4)$$

Checking the limit for equation (4): If there is no likelihood of escape $P_1=0$, log(1)=0, and as expected $y=\infty$.

Some examples of the use of equation (4):

N =	30						
					y		
			E =		E =		E =
p_1	p_N		0.01		0.5		0.99
0.1	0.958		0.0		0.22		1.46
0.01	0.260		0.0		2.3		15.3
0.002	0.058		0.2		11.5		76.7
0.001	0.030		0.3		23.1		153.4
0.0001	0.003		3.3		231.0		1,535
1.00E-06	3.000E-05		335.0		23,105		153,506

TABLE A. Some sample values for N=30 labs. The body of the Table is years to at least one escape.

For instance, if the probability of escape from a single lab in a single year is 0.0001 or 0.01% (a reasonable estimate), and we will tolerate only a 1% chance of escape, E=0.01, over the 30-lab research enterprise, the number of years for at least one escape is only 3.3 years.

Another observation about equation (4), the number of years of research, y, which must elapse before we reach our risk-tolerance level is inversely proportional to the number of labs, n.

From: Charles Stack [mailto:cstack3@uic.edu]
Sent: Thursday, April 23, 2015 1:17 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Cc: Marc Lipsitch <mlipsitc@hsph.harvard.edu>
Subject: Comments for NSABB Meeting

Dear Madam or Sir:

I have watched the Gain of Function (GOF) controversy for years and became a charter member of the Cambridge Working Group in order to have some influence upon this research.

I agree with Dr. Marc Lipstich and colleagues that the risks of GOF experiments are far too grave considering the marginal usefulness of the discoveries that might be made.

University of Wisconsin Prof. Kawaoka and others claim that GOF research will help to develop new vaccine strains, but this is only true if a pandemic virus in the wild emerges with the same genetic profile of the GOF strain. The odds against this happening are astronomical. Vaccine manufacturers have already said that it would not be economical to produce vaccines unless a strain is circulating and identified in the wild.

Public health is undergoing wrenching financial changes, so we must be prudent with how research dollars are spent. Instead of shot-in-the-dark GOF research, I advocate for increased field surveillance for emerging viruses and other pathogens. Such surveillance may have shown that the recent Ebola virus outbreak in West Africa could have been predicted, based upon seroepidemiology of the human population and culture testing of the indigenous biota. We are performing this surveillance after the fact and need to be out in front of emerging threats.

Thank you for your consideration of my comments,

Charles R. Stack, MPH
DrPH Candidate
Estelle Goldstein Memorial Scholar
UIC School of Public Health
www.uic.edu/sph/

Certified Leader, 2013 Chicago Training
Climate Reality Leadership Corps
http://climaterealityproject.org/leadership-corps/

cstack3@uic.edu

COMMENTS ON THE NSABB DRAFT FRAMEWORK DATED 6 APRIL 2015
Marc Lipsitch, DPhil
Harvard T.H. Chan School of Public Health
mlipsitc@hsph.harvard.edu
Comments dated April 24, 2015

Overall I believe that the Draft Framework dated 6 April 2015 contains much that is of value, that it makes mostly appropriate recommendations for the structuring of the risk and benefit assessment, and that it appropriately mentions alternative approaches, human error, and the importance of including scenarios where countermeasures may and may not be effective, as well as both scenarios involving accidents and those involving malevolent action.

However one essential element appears to be missing and to suffer from vague and contradictory directions. This is the question of *what exactly* is being assessed for its risks and benefits, what are the *components* of those risks and benefits, and *in comparison to what* are they being assessed?

1. What is being assessed? To calculate, say, the risks of GOF experimentation, it is necessary to specify which pathogen(s), investigated by how many laboratories, for what period of time, at what biosafety level, among other inputs. The risk presented by one laboratory for one year will be multiplied by approximately a factor of 6 if, say, 2 laboratories work for 3 years under the same conditions. Evidence about the rate of laboratory-acquired infections (LAI) is obtainable with denominators of laboratory-years or full-time laboratory-worker-years [1].

RECOMMENDATION: Most importantly, the Framework should specify some unit of research. Specifically, because LAI are the precipitating events for most of the scenarios of greatest concern, I recommend that the unit of analysis be the high-containment laboratory-worker-year or laboratory-year, to facilitate data assimilation. Biosafety conditions should also be specified.

2. What are the components of these risks? An essential aspect of a risk assessment on this topic is to clearly separate the two components: (a) probability of an adverse outcome, and (b) magnitude or consequence of this outcome. For GOF, the probabilities of LAI are not extraordinary, but the consequences may be in certain cases.

RECOMMENDATION: Analysis would be clarified greatly by specifying that these calculations should be described separately and then appropriately combined to estimate risk.

3. In comparison to what are these being assessed? The RA/BA process is intended to aid the USG in making a decision: whether to fund GOF research, and under what conditions. Two possible decisions would be to resume GOF funding using ordinary biosafety review and no additional review, or to stop funding such work for a defined period of time or permanently. In the event of the latter decision, the USG research portfolio on influenza would not be expected to change in overall size, but only in composition.

Note: This issue is particularly confused and contradictory in the current Draft and thus

needs clarifying. Guiding principle 1 in the draft states "The possible risks and benefits of not doing this work also need to be thoroughly examined." As I understand this instruction, it involves either a trivial point (risks of doing = benefits of not doing, and vice-versa) or more likely an instruction to compare GOF to no GOF, without considering the alternatives that would be undertaken. That however contradicts Guiding Principle 2. This needs to be clarified.

Note: This decision is highly consequential. In a medical context, where risk-benefit analysis is commonly employed, very different conclusions would follow from evaluating an antibiotic treatment for a life-threatening condition, where the antibiotic carries a 1/10,000 risk of causing liver failure if the alternatives were (a) no treatment or (b) treatment with another antibiotic of similar efficacy without the risk of liver failure. Similarly, an unrealistic comparison of GOF research to "not doing GOF research" might have a very different risk-benefit profile from the actual comparison, which is replacing GOF in the research portfolio with other approaches, holding the budget constant.

RECOMMENDATION: I recommend that the Framework specify that the RA/BA should compare the risks and benefits of

(1) A USG influenza research portfolio of a fixed budget including GOF and non-GOF research, with composition determined by peer review and other existing mechanisms

vs.

(2) A USG influenza research portfolio of the same budget including only non-GOF research, with composition determined in the same way apart from the removal of GOF research.

Specific comments

1) lines 49-50: SARS and MERS are no longer at issue. Same on lines 262-4. They are not still in the funding pause. They should perhaps be included in the RA/BA but this is not accurate.

2) Overall comments on guiding principles: apart from the comment above, these are sensible and comprehensive.

3) lines 268-279 All of these are important considerations but of particular concern are experiments reasonably anticipated to result in a virus that is readily transmissible, not known to be currently circulating in humans, and virulent in humans. Increasing one of these in the absence of others may not be of such concern, but the three properties together are of special concern. This was much discussed at the NAS meeting, and in particular David Relman's remarks emphasized the cruciality of the combination.

4) lines 282-334 This list is in general quite appropriate and comprehensive, with exceptions described here and in the next comment. The comment "Opportunity costs might also be considered" (l. 333) is ambiguous. If it means opportunity costs of doing GOF as opposed to other, alternative (and generally much safer) approaches, the alternatives MUST be considered (line 180 ff.). If it means something else, it should be spelled out.

5) One category of risk not included and very important is reputational and credibility risk for science. If in the face of ongoing laboratory mishaps at the nation's most prestigious laboratories, the US Government decides to fund and approve experiments to create novel pathogens with pandemic potential, and there is an accident involving serious outcomes following accidental infection, the credibility of science as a whole will suffer, leading the public to question the quality of public stewardship of biomedical funding, and indeed to question the reliability of scientific and medical advice regarding risk. This should be explicitly considered as an independent category of harm that could result from an accident. In an era of science skepticism related to issues from climate change to vaccine safety, this could be harmful to science's ability to inform policy, not to mention to science funding.

6) lines 347-352. Scientific knowledge is a benefit of all scientific research, including GOF and alternative approaches to virology. Scientific knowledge has appropriately been characterized as having unpredictable outcomes. This is a reason to do science, but not a reason to choose one (risky) scientific approach in preference to other (low-risk) approaches. The question of what unique scientific knowledge can be generated is only appropriate if it is asked both of GOF and of alternative types of scientific study that would be foregone -- that is, opportunity costs must be properly accounted for.

7) lines 354-385. The emphasis on comparison against alternatives mentioned in lines 373 and following is welcome, but should cover all of these points.

8) lines 387-391 Informing policy decisions. There is an important distinction between research that can *inform* policy decisions and research that can *uniquely improve* policy

decisions. Policy makers may well use information in decisions that does not make those decisions better. Evidence for a benefit should be evidence that GOF results *uniquely improve* policy decisions. The term uniquely is important because the phenotypes and in most cases the mutations found in the GOF influenza experiments to date were all known to be important for mammalian adaptation of influenza viruses before the GOF studies. Improvement vs. informing is crucial because the ability to predict influenza pandemic risk is agreed by a wide range of scientists with varying views on GOF to be a long-term future aspiration without evidence that such predictions can be validated by experience to date [2].

9) Line 441: accurate is a strange term to use for a hypothetical scenario. A critic could say that some aspect of a hypothetical scenario is not "accurate" because exactly that condition does not exist, but this would be crippling as a constraint on scenario generation. Credible is a good word; plausible or realistic might be other appropriate modifiers.

10) lines 410-480. Again this list is appropriate and comprehensive. Point 14 is ambiguous, and should read "For comparison against the risks of GOF research, scenarios should be generated involving the above categories (where appropriate) involving alternative, non-GOF approaches.

11) line 534-76. This list is appropriate and comprehensive. It should be emphasized that these should be applied to both GOF and alternative approaches.

13) lines 543-4. I this instance (as in all, but especially here) citation counting may be misleading. This work has been extremely controversial and therefore unusually visible. That has prompted acceptance of multiple papers by prestigious journals and much commentary (including criticism of the safety and security aspects) which has significantly contributed to citation counts. For papers as new as these one might argue that citation counts are not good indicators of scientific importance, but rather (in the short term) of visibility and controversy.

1. Lipsitch M, Inglesby TV (2014) Moratorium on research intended to create novel potential pandemic pathogens. MBio 5.
2. Russell CA, Kasson PM, Donis RO, Riley S, Dunbar J, et al. (2014) Improving pandemic influenza risk assessment. Elife 3: e03883.

These references are included for convenience of the NSABB and form part of my formal comments.

GUEST EDITORIAL

Moratorium on Research Intended To Create Novel Potential Pandemic Pathogens

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Research on highly pathogenic organisms is crucial for medicine and public health, and we strongly support it. This work creates a foundation of new knowledge that provides critical insights around the world's most deadly infectious diseases, and it can lay groundwork for the future development of new diagnostics, medicines, and vaccines. Almost all such research can be performed in ways that pose negligible or no risk of epidemic or global spread of a novel pathogen. However, research that aims to create new potential pandemic pathogens (PPP) (1)—novel microbes that combine likely human virulence with likely efficient transmission in humans—is an exception to that rule. While this research represents a tiny portion of the experimental work done in infectious disease research, it poses extraordinary potential risks to the public.

Experiments that create the possibility of initiating a pandemic should be subject to a rigorous quantitative risk assessment and a search for safer alternatives before they are approved or performed. Yet a rigorous and transparent risk assessment process for this work has not yet been established. This is why we support the recently announced moratorium on funding new "gain-of-function" (GOF) experiments that enhance mammalian transmissibility or virulence in severe acute respiratory syndrome (SARS), Middle East respiratory syndrome (MERS), and influenza viruses. This realm of work roughly corresponds with the work we have termed PPP above. Because the term "gain of function" in other contexts can be used to describe techniques of scientific research that have nothing to do with the creation of novel potential pandemic pathogens, we think the term can be too broad and can mislead. Throughout this commentary, we focus on research designed to create PPP strains of influenza virus, the type of research that initially attracted attention, leading to the moratorium and for which the most discussion has already occurred. Other types of gain-of-function research on influenza and studies intended to enhance pathogenicity or transmissibility of MERS and SARS coronaviruses may or may not fit the definition of PPP research and further clarification is needed and ongoing. As we discuss near the end of this article, it will be essential to clarify the different risks and benefits entailed by different types of experiments covered by the funding pause (2).

The purpose of this research funding pause is to complete "a robust and broad deliberative process . . . that results in the adoption of a new [U.S. Government] gain-of-function research policy" (3). The moratorium would stop new funding for the following:

> . . . research projects that may be reasonably anticipated to confer attributes to influenza, MERS, or SARS viruses such that the virus would have enhanced pathogenicity and/or transmissibility in mammals via the respiratory route. The research funding pause would not apply to characterization or testing of naturally occurring influenza, MERS, and SARS viruses, unless the tests are reasonably anticipated to increase transmissibility and/or pathogenicity. (3)

The new U.S. Government (USG) policy also encourages the currently funded U.S. Government and nongovernment research community to join in adopting a voluntary pause on research that meets this gain-of-function definition. Some 18 NIH research projects that possibly meet that definition have been identified (2). The moratorium does not apply to the larger infectious disease research portfolio supported by the U.S. Government. In particular, it does not affect disease surveillance or vaccine development programs. During the moratorium, a deliberative process will occur that will be led by the National Science Advisory Board for Biosecurity and the National Academy of Sciences. This process is intended to produce "recommendations for risk mitigation, potential courses of action in light of this assessment, and propose methodologies for the objective and rigorous assessment of risks and potential benefits that might be applied to the approval and conduct of individual experiments or classes of experiments" (3).

In this commentary, we discuss key elements of risk analysis and offer an example of an approach that could be taken. We describe benefit analysis, offering an account of the kinds of benefits that are relevant and our own view of those at this point. We note other factors that are important to consider. And we argue that a moratorium is the right approach until a rigorous, objective, and credible risk assessment process can be established.

RISK ANALYSIS

Risk assessment for GOF work should be quantitative, objective, and credible. Extensive qualitative arguments have been made on both sides of this issue, and these arguments have not provided sufficient clarity or evidence to resolve concerns or identify a consensus path forward. Quantitative assessments should now be performed so as to provide specific calculations and information to inform decisions. It is also important for these risk assessments to

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be objective. Given the stakes in this process, the risk assessment process should be directed by those without a clear personal stake in the outcome, just as peer review of science is performed by those without a direct interest in the outcome. The credibility of the risk assessment will depend both on the rigor of the quantitative process and the perceived objectivity of the process.

The record of laboratory incidents and accidental infections in biosafety level 3 (BSL3) laboratories provides a starting point for quantifying risk. Concentrating on the generation of transmissible variants of avian influenza, we provide an illustrative calculation of the sort that would be performed in greater detail in a fuller risk analysis. Previous publications have suggested similar approaches to this problem (1, 4).

Insurers and risk analysts define risk as the product of probability times consequence. Data on the probability of a laboratory-associated infection in U.S. BSL3 labs using select agents show that 4 infections have been observed over <2,044 laboratory-years of observation, indicating at least a 0.2% chance of a laboratory-acquired infection (5) per BSL3 laboratory-year. An alternative data source is from the intramural BSL3 labs at the National Institutes of Allergy and Infectious Diseases (NIAID), which report in a slightly different way: 3 accidental infections in 634,500 person-hours of work between 1982 and 2003, or about 1 accidental infection for every 100 full-time person-years (2,000 h) of work (6).

A simulation model of an accidental infection of a laboratory worker with a transmissible influenza virus strain estimated about a 10 to 20% risk that such an infection would escape control and spread widely (7). Alternative estimates from simple models range from about 5% to 60%. Multiplying the probability of an accidental laboratory-acquired infection per lab-year (0.2%) or full-time worker-year (1%) by the probability that the infection leads to global spread (5% to 60%) provides an estimate that work with a novel, transmissible form of influenza virus carries a risk of between 0.01% and 0.1% per laboratory-year of creating a pandemic, using the select agent data, or between 0.05% and 0.6% per full-time worker-year using the NIAID data.

Readily transmissible influenza, once widespread, has never before been controlled before it spreads globally, and influenza pandemics historically have infected about 24 to 38% of the world's population (8, 9). The case-fatality ratio of a novel strain is of course unpredictable. The worst case might be a case-fatality ratio similar to that of avian H5N1 influenza virus in people, which approaches 60% (10). A greatly attenuated version of the same virus might have a case-fatality ratio of "only" 1%.

Again, multiplying the pandemic attack rate (24% to 38%) times the global population (~7 billion) times the case-fatality ratio (1% to 60%) would produce an estimate of between 2 million and 1.4 billion fatalities from a pandemic of a highly virulent influenza virus strain.

Putting all these numbers together, the select agent data suggest that a laboratory-year of experimentation on virulent, transmissible influenza virus might have an 0.01% to 0.1% chance of killing 2 million to 1.4 billion, or an expected death toll of 2,000 to 1.4 million fatalities per BSL3-laboratory-year. From the NIAID data, for each full-time person-year of BSL-3 work, we might expect a toll of between 10,000 and over 10 million.

These numbers should be discussed, challenged, and modified to fit the particularities of specific types of PPP experiments. For creation of novel, transmissible, virulent influenza virus strains, they may overstate the risk for the following reasons: (i) most such work is done in BSL3+ labs, which may be safer than BSL3; (ii) control measures, including vaccination and antiviral prophylaxis of laboratory workers, might reduce the risk of infection and of spread, although none of these is perfect; (iii) the human case-fatality ratio of an avian influenza virus strain that gains transmissibility could be below 1%; (iv) transmissibility in laboratory animals does not necessarily indicate transmissibility in humans (11, 12); and (v) novel strategies of molecular biocontainment (13), if employed, might reduce the risk of human transmission of a strain used in transmission experiments in other mammals.

On the other hand, these numbers may understate the risk because (i) the select agent calculation includes in its numerator only BSL3 labs, but in the denominator, BSL3 as well as BSL2 and BSL4 "registered entities" as separate figures for BSL3 are not publicly available (5); (ii) the rate of accidents is calculated for U.S. labs, while GOF experiments are performed in many countries; if this work expands to some of the many countries with less stringent standards than those in the United States (14), risks could be higher; and (iii) the costs of an accidental pandemic considered here are deaths only, but additional losses would include scientific credibility, nonfatal health outcomes, economic and educational losses, etc.

The illustrative calculations above show that approximate risk estimates are possible for creation of PPP strains of influenza virus. During the deliberative process initiated with this moratorium, the risk assessment approach that is established should be able to provide calculations that reflect these and other available probability and consequence estimates and take into account the range of modifying factors, including those just described. The risk assessment process should also be able to provide calculations related to PPP experiments where the risks are harder to calculate given more limited data, such as enhancement of coronavirus pathogenicity in small mammals.

BENEFIT ANALYSIS

On the surface, analyzing the benefits of PPP experimentation would seem more difficult. In the cumulative process of knowledge acquisition that is science, it is hard to see far ahead where a particular type of research may lead. On the other hand, scientists make judgments about the relative merits of experimental approaches on a daily basis in their roles as investigators and grant reviewers. Doing and funding science constitute a process of severe winnowing (especially severe in today's tight funding climate) in which we choose to pursue one approach and not to pursue others based on judgments of which approaches are expected to have the lowest cost, highest probability of success, and greatest yield of valuable findings, among other considerations. Implicit in this process is the idea of opportunity cost. In prioritizing the week's or the year's research work, we do not judge in isolation whether a particular experiment should be done or not done. We decide how to allocate our time and funding among possible approaches, devoting resources to the portfolio of efforts that seems most promising. Similar prioritizations are made by funders when they decide which kinds of research will be funded and which research will not.

The analysis of benefits of PPP experiments should follow this familiar approach. The choice is not between doing PPP experiments and doing nothing. Rather, the appropriate question is, within a portfolio of scientific and public health activities designed

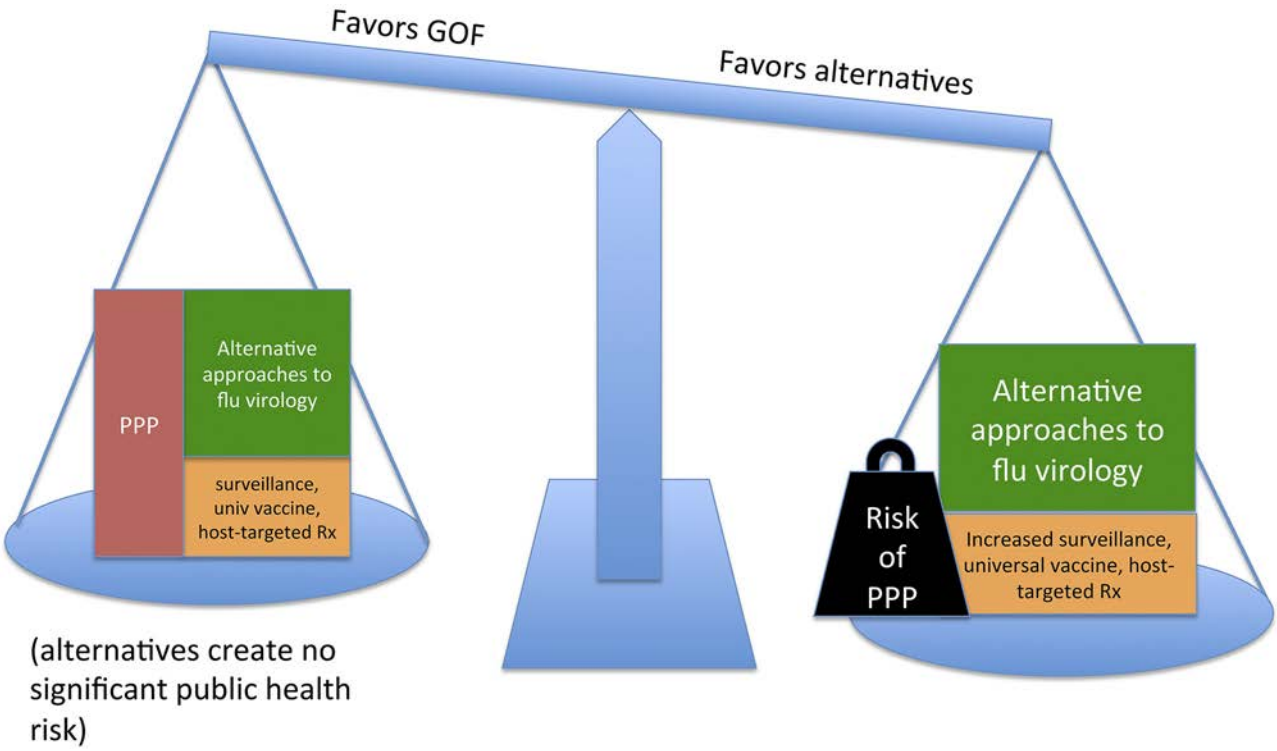

FIG 1 Weighing risks and benefits. The benefits (squares) of spending a fixed quantity of resources on a portfolio of activities, including PPP research (red), other approaches to influenza virus virology (green), and other public health activities to defeat influenza (yellow), should be weighed against the benefits of a portfolio in which the other activities are expanded to use the resources freed by not supporting PPP activities, reflecting the opportunity cost of the PPP research. If there are net benefits to including PPP activities in the portfolio, then they should be weighed against the net risks created by PPP experiments, which in the case of influenza transmissibility enhancement, we have argued (see the main text, Risk Analysis) are exceptionally high. The balance may differ for other activities, but this comparison of benefits of portfolios with and without gain-of-function experiments is the appropriate comparison, with any net benefits weighed against net risks. univ, universal.

to understand and combat influenza or a coronavirus (or, perhaps, in our portfolio of infectious disease countermeasures more broadly), what are the benefits of including PPP approaches compared to the benefits of expanding other parts of the portfolio to use the resources in another way? From the perspective of public health and the practical goal of preventing and treating flu, alternative approaches include those which, like PPP experiments, seek to enhance our scientific understanding of biology, pathogenesis, and transmission. Alternatives also include efforts to develop treatments and prevention measures, including surveillance, through means other than improving our basic biological understanding of influenza (4). This approach is shown graphically in Fig. 1, which also depicts the risks of PPP research. Such risks should be weighed against the risks of alternatives, which are typically much smaller or even negligible. Figure 1 embodies the idea that PPP research should be a component of our research portfolio only if devoting resources to PPP studies at the expense of alternatives has net benefits that outweigh the unique risks of PPP studies.

This comparative approach to benefits should be informed by a hard-nosed look at the benefits that are readily achievable by PPP experiments, not hypothetical outcomes that could someday lead to unspecified benefits. We acknowledge the possibility that PPP experiments may lead to benefits we cannot today envision. But so could the experiments that are done in their place if support for PPP is reallocated to other scientific approaches. The possibility of unanticipated benefits is surely a reason to do science, but it is not a reason to favor PPP approaches over others, unless some specific case can be made for the unique yet unanticipated benefits of PPP work. Such a case seems hard to imagine for benefits that are by assumption unanticipated.

For example, it has been suggested that mutations or phenotypes identified through PPP experiments could be used to sort through the massive diversity of nonhuman influenza virus strains to prioritize those that should trigger countermeasures, including prepandemic vaccine manufacturing. While this is possible in principle, there are many practical barriers to achieving public health benefits of this sort from PPP studies (15). Lists of mutations, and even phenotypes, associated with PPP studies can be compiled and compared against isolates of influenza viruses from birds and other nonhuman sources (16). We know that these lists are unreliable and can even be misleading: the mutations in hemagglutinin identified by two prominent PPP experiments with H5N1 do not reliably confer human receptor specificity even for other H5N1 viruses (17). The E627K mutation in the PB2 gene, known as a virulence and transmissibility determinant before GOF experiments (16, 18, 19), found repeatedly in GOF experiments in H5N1 (20, 21), and used for pandemic risk assessment in H7 viruses (16), was found in some isolates of the H1N1pdm strain in 2009, leading to concern about possible increased virulence and transmissibility. Yet it conferred neither trait in this genetic background (22).

At this time, the high levels of epistasis—dependence of phenotype on the genetic background in which a mutation is found—make prediction of pandemic risk for any given strain more of an art than a science. Indeed, the very presumption that we will see human cases of an incipient pandemic before that pandemic occurs has never been met in practice (23): we have never observed zoonotic cases of any flu virus before it caused a pandemic. This is not to deny that PPP experiments provide any useful data for surveillance and prioritization. Rather, it is to say that other approaches can also identify such predictors (as in the case of the PB2 mutation [11, 13, 14]) and that the ability to use markers of putative transmissibility or virulence to make reliable predictions remains far in the future (23). The fact that some analysts consider mutations identified in PPP experiments when assessing threats of viruses found in surveillance does not mean that the use of such mutations improves the predictions, a claim for which we have no evidence because no pandemic strain has ever been identified in advance. The analysis of benefits of PPP creation should reflect this state of science.

According to some proponents, the most valuable scientific finding of experiments to make ferret-transmissible mutants of influenza A/H5N1 is the definitive proof that such variants could be produced with a small number of mutations. This could not be definitively proven without doing the PPP experiment to manufacture a potentially pandemic variant of H5N1 (24). While it is now undeniable that ferret-transmissible mutants of influenza A/H5N1 can be created experimentally, the impact on scientific opinion about the risk of a pandemic from H5N1 has been hard to gauge. Prior to the gain-of-function experiments, there was a wide range of expert opinion on the likelihood of an H5N1 pandemic (25). Some influenza experts questioned whether H5N1 was a major pandemic threat. After the publication of the experiments producing potentially pandemic H5N1, one prominent member of this group, Peter Palese, noted the shortcomings of the ferret model for humans and correctly concluded that the question of whether H5N1 can transmit efficiently in people remains unsettled (11), as it must until the phenomenon is directly observed in nature. From a practical perspective, responsible policy makers and public health leaders should have been planning for the possibility of an H5N1 pandemic before PPP experiments on H5N1 were undertaken. In some countries of the world, they were stockpiling vaccines against H5N1 (26, 27) and making plans for nonpharmaceutical (8) interventions in the event of a pandemic. The same remains true after the experiments. We have observed no discernible influence of the H5N1 PPP experiments on H5N1 policy preparations.

CALCULATING OTHER FACTORS

During the moratorium, progress should also be made in calculating the risks associated with potential deliberate misuse of PPP strains and with potential deliberate misuse of the information that is created and published following PPP experimental work. This calculation should take into account the possibility of deliberate theft and dissemination by either persons working within a lab or theft by those outside the lab. While the probability of this is likely to be very low for most scientists and most laboratories, it is not zero. There is a precedent of scientists using pathogens from their own labs to cause harm. And as with potential accidents, while the probability may be very low, the consequences could be very high.

This assessment should also take into account the possibility that scientists may deliberately misuse the knowledge gained and published following the experiments by recreating the novel PPP strains in another laboratory using methods from published papers and then purposefully disseminating it. This possibility is typically dismissed out of hand by many scientists. But before dismissing that possibility, an analysis by an assembly of experts in the best position to make that judgment should be conducted. What is the possibility that individuals or groups who would seek to carry out such an act would develop the capacity and skill to carry it out? Given that once knowledge is published, it will be available forever, these questions are not just about the possibility of this happening in today's world but also anytime in the future. Despite the inherent uncertainties in trying to answer these questions, they should be answered with the best possible expertise.

Similarly, the moratorium should be used as a time to answer, or at least be addressing, another major issue as well: the international approach to funding, authorizing, and overseeing PPP. An accident or deliberate act involving PPP anywhere in the world could conceivably impact the public around the world. Therefore, the community of nations has an abiding interest to set common rules for how this work will be pursued. However, at this point, few countries have begun any kind of deliberative process on an approach to research with these unique dangers. Country X should have the right to know if this work is going on in country Y, and if so, what is being done to ensure it is done with the greatest safety and security. But currently, the way country X finds out about PPP work being done elsewhere in the world is when it is published in a science journal. Given the prestige that some scientists have received for pursuing PPP research, it would be surprising if scientists from countries around the world did not increasingly pursue it. As comparatively less experienced labs decided to pursue this work, this will increase potential dangers.

A MORATORIUM IS THE RIGHT STEP

There are prominent scientists who agree that there are potential serious dangers to this work and agree that a risk assessment process is needed but who are opposed to a moratorium being imposed while such a risk assessment process is undertaken. They believe that a moratorium should be avoided for reasons that include the potential damage it can do to the funding and work of that lab and to the careers of those involved in the work.

We have a different view. A substantial number of scientists agree that there are extraordinary potential consequences of the work (15). There is no rigorous, objective, credible risk assessment process to judge the risks and benefits of proceeding with it. We believe that the responsible course is to take a research pause until such a risk assessment process is established, which creates a stronger basis for decisions and actions. This is not solely a scientific issue. It is a scientific and public health and safety issue, and it is an issue in which the public itself has an abiding interest.

We have no interest in stopping scientists from doing their work or preventing laboratories from receiving funding. The narrow and defined area of GOF research intended to create novel potential pandemic strains should be put on pause until the risk assessment process is completed. The same laboratories and scientists whose work has been stopped by the moratorium are free and able to pursue all other avenues of infectious disease research except for that narrowly defined by the GOF definition in the new policy; to the extent that other activities not meeting the narrow

definition in the pause have been included in letters to principal investigators ordering or requesting work stoppage, the boundaries of the funding pause should be quickly clarified to allow important alternative work on flu to continue. We note that there are more than 250 NIH-funded projects listed as active with titles containing MERS, SARS, coronavirus, or influenza (28), of which 18 have been affected by the funding pause. The number that remain on pause may be further reduced by negotiations between investigators and the NIH, which are now under way, that will define which projects truly are within the scope of the moratorium and which do not meet its terms and can resume.

The character and scope of the risk assessments that are applied are important. To establish methodologies and approaches for risk assessment and risk mitigation for this context, it would be valuable to start with a global assessment of the risks and benefits of this realm of research, identifying the common aspects of risk and benefit within PPP experiments and other approaches covered in the funding pause. For example, any risk assessment should include estimates of the probabilities of accidental infection and extensive spread, as well as estimates of the impacts of these events should they occur. The specific values of these estimated parameters will differ for different types of experiments. It will then be necessary to set standards and expectations for the quality and characteristics of risk-benefit assessments for individual experiments, for example, to distinguish coronavirus research from influenza research, enhancements of pathogenicity from enhancements of transmissibility, and other important distinctions. Given that the term "risk assessment" is used to mean different things by different people, an agreement on an approach to individual risk assessments would be needed to ensure rigor and credibility. Once this kind of analytic structure is established, individual risk assessments for GOF experiments that meet the definition in the new USG policy (3) should become the norm before such experiments are funded. Crucially, this process should be quantitative, rather than relying on unquantified and unverifiable assurances that particular laboratories are safe.

CONCLUSIONS

The results of this risk assessment process are important not only to the U.S. Government, which had been a major funder of PPP experiments, but also to other funders, regulators, and investigators worldwide who consider such experiments. Our support for the funding pause and associated deliberative process does not indicate that we would support a permanent end to all experiments subject to the pause. There may be research endeavors that are subject to the moratorium that have a risk-benefit profile sufficiently favorable to justify their resumption once risks and benefits have been explicitly set forth. After 2 years of debate, we think the balance is evidently unfavorable for experiments to enhance avian influenza virus transmissibility, but other classes of experiments may be different. In the meantime, the moratorium is an appropriate and responsible step while dedicated and rigorous efforts are made to understand the risks and benefits of this work.

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Improving pandemic influenza risk assessment

Abstract Assessing the pandemic risk posed by specific non-human influenza A viruses is an important goal in public health research. As influenza virus genome sequencing becomes cheaper, faster, and more readily available, the ability to predict pandemic potential from sequence data could transform pandemic influenza risk assessment capabilities. However, the complexities of the relationships between virus genotype and phenotype make such predictions extremely difficult. The integration of experimental work, computational tool development, and analysis of evolutionary pathways, together with refinements to influenza surveillance, has the potential to transform our ability to assess the risks posed to humans by non-human influenza viruses and lead to improved pandemic preparedness and response.

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Influenza pandemics arise when antigenically novel influenza viruses enter and spread extensively in the human population. By this definition, there have been five influenza pandemics in the last 100 years, the worst of which cost 50 million lives worldwide (***Johnson and Mueller, 2002***). Of these pandemics, three likely arose from the introduction of genes from avian viruses into the human population (1918—H1N1, 1957—H2N2, 1968—H3N2 (***dos Reis et al., 2009***; ***Neumann et al., 2009***, ***Worobey et al., 2014***)), one arose from the introduction of a swine virus (2009—H1N1 (***Smith et al., 2009***)), and one was likely due to the unintended reintroduction of a previously widespread human virus that had not been seen in humans for two decades (1977—H1N1 (***dos Reis et al., 2009***, ***Nakajima et al., 1978***, ***Palese, 2004***)). However, the viruses responsible for these pandemics represent only a tiny fraction of the total diversity of influenza A viruses that exist in nature (***Webster et al., 1992***). Assessing which viruses pose the greatest risk of causing the next human pandemic is an enormous challenge.

Pandemic influenza risk assessment faces a fundamental problem: a paucity of empirical data on the differences between pandemic viruses and their immediate ancestors from non-human hosts. The challenge was clearly articulated by Harvey Fineberg in his analysis of the US government's response to the 1976 swine influenza scare (***Fineberg, 2009***): 'The first lesson is to avoid over-confidence about scientific insights. Major flu pandemics arise on average only about three times every century, which means scientists can make relatively few direct observations in each lifetime and have a long time to think about each observation. That is a circumstance that is ripe for over-interpretation.'

Core elements of current approaches to pandemic preparedness and mitigation, such as the development of vaccines and stockpiling of antiviral drugs, require detailed virological and

immunological data on viruses with perceived pandemic potential and ample lead time for production (***Jennings et al., 2008***, ***Keitel and Piedra, 2014***). The substantial diversity of known influenza viruses in non-human hosts, and the frequent identification of new viruses, makes extensive experimental testing and development of pandemic preparedness measures against all viruses unfeasible. Thus, there is a need for continuing attempts to assess the pandemic risks posed by non-human viruses in order to prioritize viruses of concern for pandemic preparedness planning. Currently, influenza pandemic risk assessment is largely driven by a simple idea: animal viruses that cause sporadic human infections are thought to pose a greater pandemic risk than viruses that have not been documented to infect humans (***Figure 1***). This intuitively attractive idea does not have direct empirical support, as none of the viruses that caused the 1918, 1957, 1968, or 2009 pandemics was detected in humans before they emerged in their pandemic form (***Smith et al., 2009***). This is largely due to a lack of surveillance (1918, 1957, and 1968 pandemics) and to the mistaken assumption that virus subtypes already circulating in humans were unlikely to cause pandemics (2009 pandemic) (***Peiris et al., 2012***). However, increased surveillance has probably improved the chance that the next pandemic virus will be identified prior to sustained human-to-human transmission.

If it is true that influenza surveillance has the possibility of identifying potential pandemic viruses before they begin to spread extensively between humans, then improving the basis for assessment of the risks posed by those viruses is an important goal. The level of public health concern about identified non-human influenza viruses should be a function of the potential of each virus to gain the ability to transmit efficiently from human to human and the severity of disease that such a virus would cause should it become pandemic. These two high-level phenotypes are each determined by the interaction of a number of biochemical traits of the virus during human infection (***Figure 2***) (***Chou et al., 2011***, ***Hatta et al., 2001***, ***Kobasa et al., 2004***, ***Labadie et al., 2007***, ***Yen et al., 2011***), the state of immunity to that influenza virus in human populations at the time of emergence (***Miller et al., 2010***, ***Xu et al., 2010***), and by environmental factors such as temperature and humidity (***Shaman et al., 2011***).

Currently, the primary tool that uses multiple data streams for assessing pandemic risk is the Influenza Risk Assessment Tool (IRAT) (***Cox et al., 2014***, ***Trock et al., 2012***). The IRAT integrates existing knowledge, including information on virus transmissibility and disease severity, with expert opinion about potential pandemic viruses to assign relative risk scores to those viruses. The IRAT is useful for identifying key gaps in knowledge,

Figure 1. Evidence for concern and actions to mitigate influenza pandemics. Types of evidence that have been, or could be, used to justify specific preparedness or mitigation actions prior to evidence of sustained human-to-human transmission, largely based on the authors' interpretation of national and international responses to H5N1, H7N9, and H3N2v outbreaks (***Epperson et al., 2013***, ***WHO, 2011***). Red indicates largely sufficient, orange partly sufficient, yellow minimally sufficient, gray insufficient. * high pathogenicity phenotype as defined by the World Organization for Animal Health (OIE) (***OIE, 2013***).

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focusing risk management efforts, and providing clear documentation of decision rationales. However, to be used optimally, the IRAT requires a substantial amount of experimental data about virus phenotypes including information on receptor binding, transmissibility in laboratory animals, and antiviral treatment susceptibility. In the absence of phenotype data, preliminary assessments with the IRAT must rely on extrapolations from related viruses, which are prone to subjective interpretation.

The biochemical traits that determine virus phenotypes are themselves determined by the genetic sequence of the virus (***Figure 2***). In theory, it might eventually be possible to predict virus phenotype directly from virus sequence data. However, the complexities of the relationships between sequences and traits and from traits to disease phenotypes, make the prediction of pandemic potential from genomic sequence a tremendous challenge. Here, we discuss ways in which laboratory experiments, together with computational and theoretical developments, could improve genotype-to-phenotype prediction and, in conjunction with enhanced surveillance, improve assessment of the risks posed to humans by non-human influenza viruses.

Experimental approaches

One goal of experimental studies on non-human influenza viruses is to identify general virus traits that are likely to affect transmissibility between humans, and then relate those traits to specific virus sequence changes. For obvious reasons, direct experimental assessment of human-to-human transmission of potential pandemic viruses is not feasible. However, influenza viruses that have caused pandemics in humans have been shown to transmit efficiently in animal models (most commonly ferrets) (***Chou et al., 2011***, ***Yen et al., 2011***), thus animal models are thought to be useful for examining the genetic changes in viruses that facilitate human-to-human transmission. For example, several studies have shown that genetic changes in the neuraminidase (NA) and matrix (M) gene segments acquired by the virus lineage responsible for the 2009 H1N1 pandemic increased transmissibility in animal models (***Chou et al., 2011***, ***Lakdawala et al., 2011***, ***Yen et al., 2011***), suggesting that these changes may have played a role in enhancing the virus's transmissibility in humans and hence paved the way for pandemic emergence. When animal experiments provide quantitative measures of virus traits, these can be integrated into quantitative measures of risk assessment such as the IRAT (***Trock et al., 2012***).

Recently, several high-profile and controversial gain-of-function (GoF) studies have attempted to go beyond the characterization of existing viruses to prospectively identify new mutations in avian H5N1 viruses that enhance the ability of these viruses to transmit between ferrets by the airborne route (***Chen et al., 2012***, ***Herfst et al., 2012***, ***Imai et al., 2012***, ***Zhang et al., 2013***). Important questions about the relative risks and benefits of these studies have been debated extensively elsewhere (***Fauci, 2012***; ***Fouchier et al., 2013***; ***Lipkin, 2012***; ***Casadevall and Imperiale, 2014***; ***Lipsitch and Galvani, 2014***); here, we focus on scientific considerations.

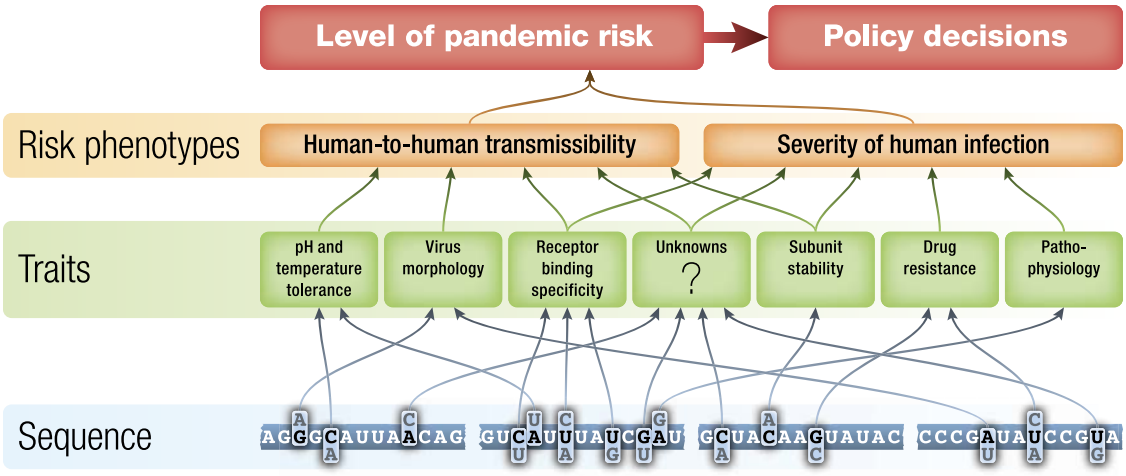

Figure 2. Schematic of potential relationships from virus genetic sequence to level of public health concern/pandemic risk. Pandemic risk is a combination of the probability that a virus will cause a pandemic and the human morbidity and mortality that might result from that pandemic. Arrows represent possible relationships between levels and are not intended to summarize current knowledge.

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Because of the vast size of genetic space, such studies cannot possibly delineate all genetic variants of a virus that might be transmissible—after all, there are more than 10^{18} different possible five-mutation variants of any given hemagglutinin (HA), which is more than what can reasonably be assayed experimentally and the vast majority will not facilitate transmissibility. A more modest goal is to attempt to associate classes of genetic or phenotypic traits with transmissibility. Transmissibility traits identified by GoF studies to date include some that were already known (such as switching receptor binding from avian-like α2,3 sialic acid to human-like α2,6 sialic acid linkages (***Yamada et al., 2006***) and lowering the optimal temperature for viral polymerase activity (***Massin et al., 2001***)), as well as some that are new, such as increasing HA stability and reducing glycosylation on HA's globular head (***Herfst et al., 2012***, ***Imai et al., 2012***). Whether these traits are either necessary or sufficient for transmissibility among humans or even other mammalian animal models remains unclear. For example, a recent study of an avian H5N1 virus found that by reassorting its internal genes with those of a 2009 pandemic virus, the virus could be rendered transmissible in guinea pigs (which have both α2,6 and α2,3 sialic acid in the upper respiratory tract) despite retaining a preference for binding α2,3 sialic acid. However, when mutations identified in earlier ferret GoF experiments were used to switch the receptor specificity to α2,6 sialic acid, transmissibility was lost (***Zhang et al., 2013***).

A key question for efforts to assess pandemic risk of non-human viruses is the degree to which certain substitutions are general markers for a phenotype, or whether the impacts of those mutations are dependent on genetic context and/or specific non-human host. Some mutations have been shown to be strong markers for phenotype for well-defined collections of viruses—for instance, the NA mutation H275Y consistently confers oseltamivir resistance on N1 neuraminidases (although the impact of the mutation on surface expression of NA, and thus virus fitness, varies dramatically) (***Baz et al., 2010***, ***Bloom et al., 2010***). Similarly, the PB2 E627K substitution adapts the viral polymerase to mammalian cells in some viruses (***Long et al., 2013***) but not others (***Herfst et al., 2010***), while other viruses have adapted to mammals via different substitutions in PB2 (***Jagger et al., 2010***, ***Mehle and Doudna, 2009***; ***Zhu et al., 2010***). In many cases, the effect of mutations can be highly sensitive to genetic context—for instance, the effects of cytotoxic T-lymphocyte escape mutations on nucleoprotein (NP) function depend on the stability of the parent protein, which can be affected by at least dozens of other mutations (***Gong et al., 2013***). Similar patterns of context dependence have recently been shown for receptor binding specificity substitutions in H5N1 viruses (***Tharakaraman et al., 2013***). Therefore, even when phenotypic traits of interest can be identified, clear genetic markers for these traits are only present in some cases.

The utility of experimental studies for informing surveillance for higher-risk viruses hinges on the question of whether virus traits associated with risk of infection and transmission in humans possess clear genetic markers. If a trait only arises from a limited number of specific mutations or combination of mutations, then experimentally delineating these mutations would be helpful for surveillance. For these cases, it is important and useful for the community to have access to collections of interpretable genotype to phenotype traits such as in the H5N1 genetic changes inventory (http://www.cdc.gov/flu/avianflu/h5n1-genetic-changes.htm) as well as computational tools to quickly connect new sequences to the body of available mutation annotation knowledge (FluSurver: http://flusurver.bii.a-star.edu.sg/). On the other hand, if a trait can be conferred by a large number of different mutations or combinations of mutations, then it will be less effective to monitor specific mutations. In such cases, it may be more beneficial to focus on the broader biochemical properties of viruses or their proteins. Developing laboratory capacity for rapid phenotype assessment would therefore be a valuable complement to high-throughput sequencing of new viruses. Moving forwards, if such biochemical traits can be clearly delineated and reliably modeled, then computational simulation of proteins could be used to predict phenotype from sequence, even for sequences from viruses that have never been experimentally tested.

Computational predictions

Computational methods present an attractive adjunct to experimental studies because they have higher throughput, have shorter turnaround times, are cheaper, and are safer than experimental work with whole virulent viruses. The main drawback of computational methods is the largely unknown accuracy of their predictions—a drawback that is exacerbated by the lack of an established framework for validating the accuracy of the numerous computational prediction methods that populate the literature.

The elements of influenza pandemic risk assessment that are most amenable to computational prediction are those that correspond to well-defined, quantifiable molecular-scale traits such as receptor-binding preference, antiviral susceptibility, antigenicity of HA and NA, and possibly T-cell epitopes. Higher-level phenotypes such as transmissibility, that integrate phenomena at a range of scales, are not yet sufficiently well understood to be reasonable targets for computational predictions. A variety of computational methods shows promise for genotype-to-phenotype prediction including molecular dynamics simulations that combine high- and low-fidelity models (***Amaro et al., 2009***) and statistical learning approaches that use protein structure, dynamics, and sequence data to predict the phenotypic consequences of mutation (***Kasson et al., 2009***). However, better prospective validation of these tools against experimental data, particularly for exploring context dependency of genetic changes, is essential before these tools can be reliably used for informing public health decisions or policymaking (***Figure 1***).

Making substantial progress in the development of computational tools and the assessment of their accuracy will require collaboration between experimental and computational scientists to produce consistent testing and validation data. One possible mechanism to spur cooperation would be a series of regular community assessment exercises similar to Critical Assessment of protein Structure Prediction (CASP) (***Moult et al., 2011***). In a CASP-like exercise, one or more experimental groups would generate quantitative phenotype data for a set of viruses, for example the relative binding of α2,3-sialoglycans and α2,6-sialoglycans, pH profile of viral activation, or sensitivity to oseltamivir, and challenge computational groups to predict that virus phenotype data from the genetic sequences of the viruses tested. The quantitative experimental data would be held under embargo while the exercise runs. Computational groups would complete predictions for these targets, the experimental data set would then be released, and a meeting would be held to assess the performance of different methods to define avenues for improvement.

Ideal experimental data sets for CASP-like exercises include thermophoretic or interferometric measurements of HA binding affinities to α2,3- and α2,6-sialoglycans (***Xiong et al., 2013***) and multi-method characterizations of viral pH activation shifts for sets of point mutants in HA (***Galloway et al., 2013***, ***Thoennes et al., 2008***). Reliable computational prediction of biochemical traits from genetic data would be a major accomplishment. However, it should be recognized that further major developments, particularly computational prediction of total virus fitness in new hosts, would still be required for realizing the utility of computational tools in policymaking.

Evolutionary theory and modeling

In addition to the genotype–phenotype relationship itself, there is a need for better understanding of the evolutionary mechanisms and pathways that allow adaptive mutations controlling host range to appear and rise in frequency. These mechanisms act in reservoir hosts, in intermediate hosts (if any), and in humans or other potential hosts; they also act at multiple scales, as viruses compete for replication within hosts and transmission between hosts (***Park et al., 2013***, ***Russell et al., 2012***). Developing better phylodynamic model frameworks (***Grenfell et al., 2004***) for modeling virus host transfer and adaptation will require collaboration between theoreticians and experimentalists.

Specific goals would be to determine realistic parameters for mutation/selection processes (***Illingworth et al., 2014***) and virus population bottlenecks at transmission (***Wilker et al., 2013***) and to generate high-resolution data sets to test and train mechanistic models. Such data-driven mechanistic models could shed light on additional constraints to virus genetic change, such as fitness valleys that separate virus genotypes adapted to one species or another, or conflicts in selection acting at different biological scales. For example, at the most simple level of understanding of the role of receptor binding, avian to mammalian host switching is often assumed to only require a binary change in receptor specificity from α2,3 to α2,6 sialic acid and to be directly related to binding affinity. However, in addition to the α2,3 and α2,6 linkages, there is a tremendous variety in the structures of oligosaccharides displaying the sialic acids and in the structure of the sialic acids in different avian hosts (***Gambaryan et al., 2012***, ***Jourdain et al., 2011***). The binding specificity for each receptor variant form may affect the potential for different viruses to cross the species barrier or make the difference between causing severe or only mild disease. Rich experimental data sets that provide insights on such factors will improve the power of evolutionary models to interpret experimental and field data.

Surveillance methodology

Detection of the genetic changes and phenotypes of concern relies on systematic characterization

of influenza viruses circulating in wild and domesticated animal populations. If there are virus traits that correlate with genetic markers observed to increase risk in humans, or that can be computationally inferred from genetic sequence data, it could be possible to monitor those markers in surveillance and adjust risk assessments prior to emergence in humans. However, the acquisition of samples entering existing surveillance networks is largely ad hoc, exhibits substantial variation by host and geographical region, and only a small proportion of the data end up in the public domain (***Figure 3***). Making non-human influenza surveillance more systematic by using statistical analysis to determine appropriate levels of coverage by geographic region and host species would facilitate the early detection of viruses of concern and also have the potential to facilitate detection of evolutionary and epidemiological patterns of virus activity that warn of potential emergence events.

There are large regions of the world and many animal populations for which little or no surveillance is performed but where significant animal influenza diversity can be inferred to exist. Systematic assessments of surveillance by geographic area and host species, similar to efforts for malaria (***Gething et al., 2012***, ***2011***, ***Hay et al., 2010***, ***Sinka et al., 2012***) and dengue (***Bhatt et al., 2013***), would help to identify major gaps where surveillance is either non-existent or unlikely to be sufficient for timely detection of viruses of concern. For enhancing surveillance, prioritizing among these gaps will require substantial improvements in understanding animal host ecology to identify hotspots for virus transmission within and among animal species. Similar efforts are required to better understand what aspects of the human–animal interface facilitate transmission of viruses between animals and humans, particularly in animal production and domestic animal settings, and the human biological and epidemiological factors that promote chains of transmission of newly introduced viruses.

One motivation for changing existing surveillance systems is to increase their power to rapidly detect changes in patterns of non-human influenza virus activity. Substantial changes, such as

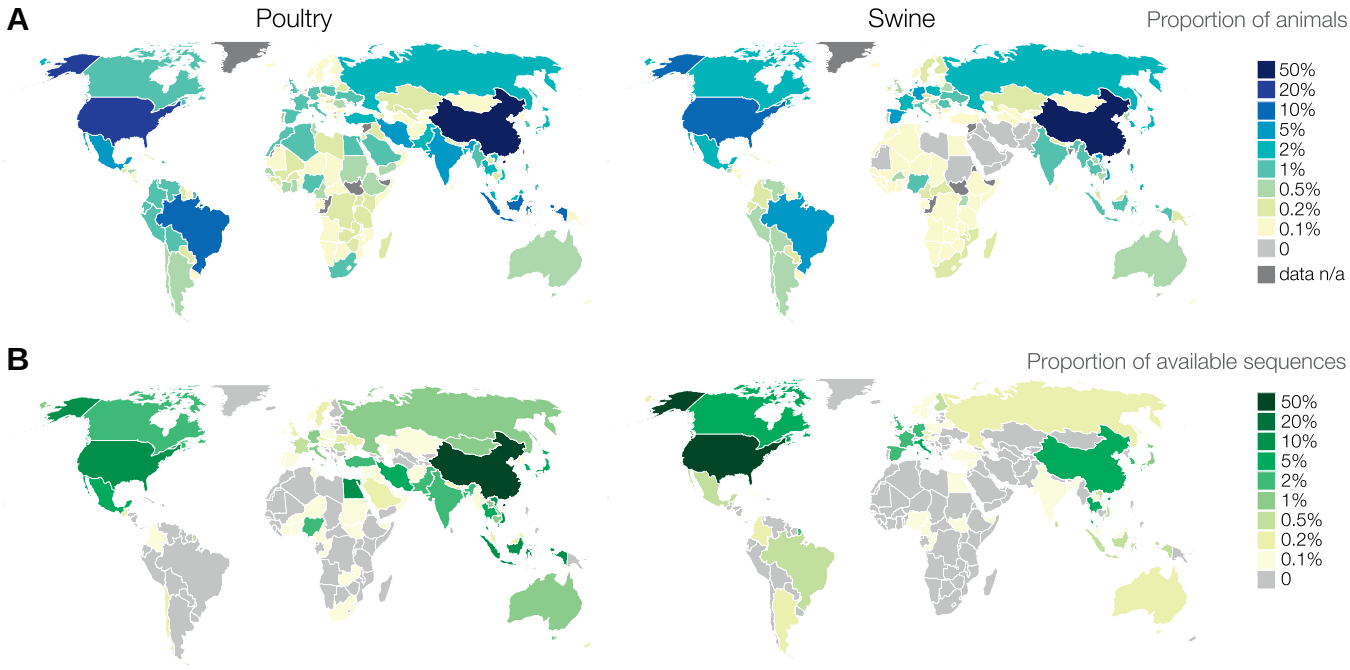

Figure 3. Geographic distribution of publicly available influenza virus genetic sequence data in comparison to poultry and swine populations.
(**A**) Proportions of worldwide animal population by country (data from the Food and Agriculture Organization of the United Nations).
(**B**) Number of unique influenza viruses for which sequence data exists in public databases from poultry or swine by country. Numbers of influenza virus sequences are not representative of influenza virus surveillance activities. Information regarding surveillance activities is not readily available. Virological surveillance, even if robust, may result in negative findings and is not captured in these figures. Most countries do not sequence every influenza virus isolate and some countries conduct virological surveillance without sharing sequence data publicly. Sequences deposited in public databases can reflect uneven geographic distribution and interest regarding viruses of concern such as H5N1 and H9N2.
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the sudden proliferation of a previously rare virus subtype or of a virus with an H9N2 internal gene cassette (**Gao et al., 2013**, **GarcÌa-Sastre and Schmolke, 2014**; **Guan et al., 1999**), could indicate the emergence of new viral variants in non-human hosts that should be prioritized for further study even before the detection of human infections of zoonotic origin (**Vijaykrishna et al., 2011**). To be useful from a human health perspective such detection systems would require sampling of animals with no obvious signs of infection, routine assessment of particular genetic signatures or full genome sequencing, and near real-time sharing of these data; these activities all present potential financial, political, and logistical constraints.

Further development of surveillance infrastructure in some geographic locations and host species is likely to be unpopular or unfeasible due to economic disincentives for disease detection. However, the geographic movements of many non-human influenza hosts, via migration or trade, make it possible to identify surrogate sources of information. For example, by linking virological and serological data, it has been possible to make inferences about swine influenza virus activity in some parts of mainland China based only on the data from Hong Kong (**Strelioff et al., 2013**).

A systematic, open, and timely global surveillance system based on viral sequence data would be a powerful tool in pandemic risk assessment. Viral sequences, with associated metadata and systematic recording of virus negative sample results, provide a rich source of information beyond the simple presence or absence of particular strains. Phylodynamic reconstructions from even a relatively small number of samples are capable of revealing lineages that are proliferating (**Grenfell et al., 2004**, **Pybus and Rambaut, 2009**). Phylogenetic methods can be used to reveal gaps in surveillance (**Smith et al., 2009**, **Vijaykrishna et al., 2011**). Genetic similarity between viruses in different locations or host species can identify drivers of transmission between populations (**Faria et al., 2013**, **Lemey et al., 2014**).

Data on negative samples would provide valuable denominators for estimating the prevalence of infection: tracking infection rates through time would give a window into transmission dynamics and allow investigation of mechanisms underlying virus circulation. The Influenza Research Database (IRD) (http://www.fludb.org) includes an animal surveillance database that contains negative test data but the amount of data is extremely limited compared to the global scale of ongoing surveillance activities. Standards should be developed for consistently recording these relevant associated metadata, so that the number of animals tested, the setting in which sampling took place, and the motivation for sampling associated with genetic data can be submitted in a consistent form to public data repositories, along with all sequence submissions.

Conclusions

It is currently not possible to predict which non-human influenza A virus will cause the next pandemic. Reducing the impact of the next pandemic will rely on early detection and mitigation strategies that slow the early spread to allow more preparatory work to be done. The integration of further experimental data with computational methods and mathematical models in conjunction with refinements to surveillance methodology will increase the feasibility of genotype-to-phenotype based assessments, increase the power of tools for more objectively assessing pandemic risk and decrease the time required for assessing the pandemic threat posed by extant non-human influenza A viruses—all of which can inform strategies to help mitigate the impact of the next pandemic.

Even as risk assessment capabilities improve, scientific insights into non-human influenza viruses must not give way to complacency that the most substantial threats have been identified and characterized. Despite the perceived risks of highly pathogenic H5N1 viruses, the emergence of the 2009 H1N1 pandemic virus in humans, the increasing incidence of human infection with H7N9 viruses in China since 2013, and the first documented human infections with H6N1 (**Wei et al., 2013**) and H10N8 (**Chen et al., 2014**) viruses highlight the importance of remaining vigilant against as-yet unrecognized high-risk viruses and the value of surveillance for influenza viruses in humans. Beyond further scientific investigations and refinement of surveillance capacity, the development of local surveillance-based outbreak response capacity worldwide remains essential. The first wave of the 2013 H7N9 outbreak in China demonstrated the value of swift coordinated action, including the timely dissemination of surveillance data, to limit further incursions of new viruses into the human population. Without developing similar response capacities in other areas at high risk of new virus introductions, we are only building expensive systems for watching the next pandemic unfold.

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WHERE SHOULD THE RED LINES BE DRAWN ?

1. **Making Ebola, Lassa or other hemorraghic fever viruses transmissible by coughing or sneezing**
2. **Making HIV transmissible by coughing, sneezing or skin contact**
3. **Making Ebola or rabies transmissible by mosquitos**
4. **Making highly-pathogenic avian influenza viruses transmissible between humans**
5. **Increasing the transmissibility of SARS and MERS viruses between humans**
6. **Making influenza viruses resistant to vaccines and antiviral drugs**
7. **Creating chimeric viruses that could be anticipated to have pandemic potential**
8. **Recreating extinct or eradicated viruses**
9. **Making drug-susceptible bacteria resistant to antibiotics**
10. **Making group A streptococcus (*S. pyogenes*) resistant to penicillin**
11. **Making malaria (*P. falciparum*) resistant to artemisinin combination treatment**
12. **Increasing toxin production of *Pertussis* or *Clostridium difficile***

HI-PATH AVIAN INFLUENZA VIRUSES

SOME IMMEDIATE RED LINES CAN BE DRAWN

1. **GOF experiments *to obtain mammalian transmissibility* of HPAI viruses by respiratory droplets**

Strain	Known human cases (dead-end infections)	Deaths	Mortality
H5N1	826	440	53%
H7N9	>640	224	35%

Other dead-end infection strains (121 cases):
H5N6, H6N1, H7N2, H7N3, H7N7, H9N2 & H10N8

2. **GOF experiments with chimeric influenza viruses**

H1N1 1918-like and analogs

3. **Human pandemic influenza viruses**

Engineering H1N1, H2N2, H3N2 *to totally escape vaccine control*

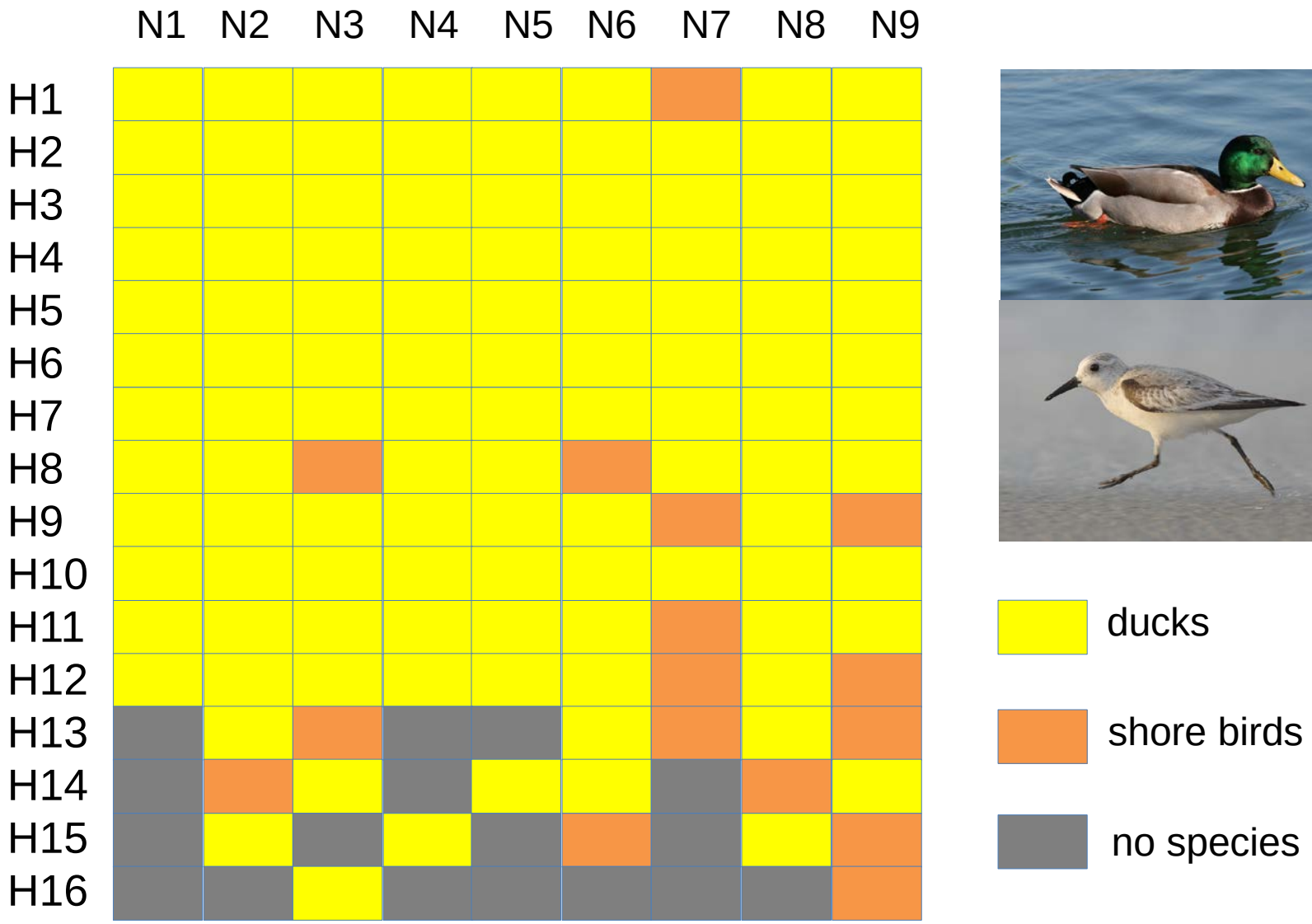
4. **GOF experiments *to increase transmissibility or pathogenesis* of human respiratory viruses**

SARS-CoV	8422	916	11%

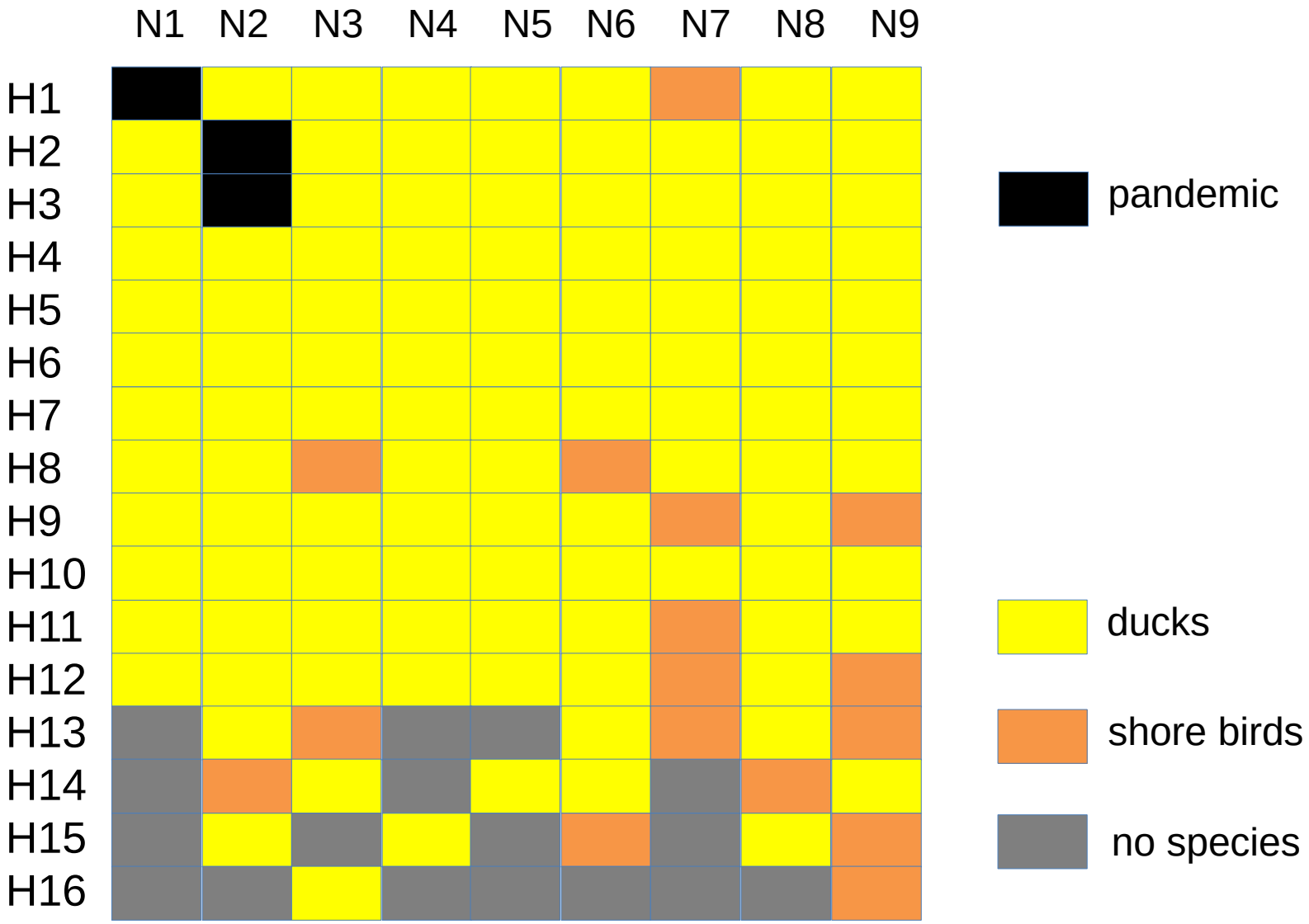
Foundation for Vaccine Research
NSABB Meeting, May 5, 2015

Sources WHO & CDC

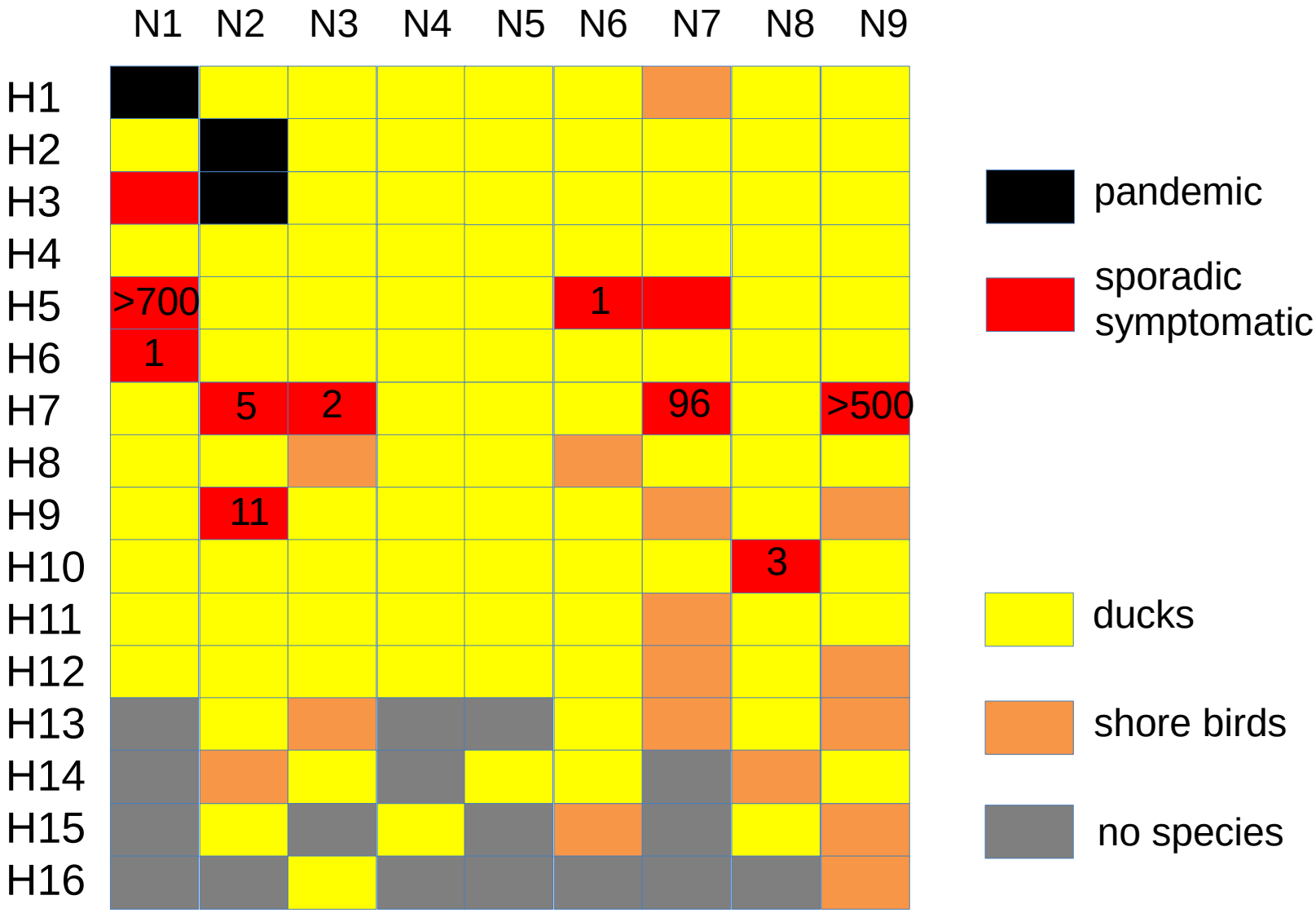
Aquatic bird reservoir of 127 H,N flu combinations



Only 3 pandemic H,N combinations in 100 years



Spillovers occur – all dead-end infections



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Infectious Diseases Society of America

August 10, 2015

[Submitted electronically to nsabb@od.nih.gov]

Samuel L. Stanley, MD
Chairman of the NSABB
Office of Science Policy
National Institutes of Health

IDSA Recommendations to the NSABB to consider during the Risk Benefit Assessment Process of Gain-of-function Research

Dear Dr. Stanley,

The Infectious Diseases Society of America (IDSA) is pleased to offer recommendations to the National Science Advisory Board for Biosecurity (NSABB) as it works with Gryphon Scientific to assess the risk and benefits of gain-of-function (GOF) research on pathogens with pandemic potential.

Ongoing technological advances in the life sciences increasingly offer critical new capabilities for understanding and managing human-microbe interactions. The goals of these efforts include health promotion and disease prevention. At the same time, these same capabilities, especially the means of manipulating genomes and, therefore, the properties of bacteria, viruses, and other infectious agents, pose important risks. Efforts to study and/or predict the natural evolution and emergence of pathogenic microbes by deliberately creating pathogens in the laboratory with enhanced disease-causing and transmission-promoting properties pose the greatest concern. Examples of this gain of function research include the recent creation of highly pathogenic avian influenza viruses with altered host range, enhanced transmissibility, and/or the ability to evade certain forms of human immunity.

ID specialists will be among the physicians who will respond to care for affected individuals in any microbial disease outbreak, be it of natural or human origin—either accidental or deliberate. ID specialists are also among those leading research efforts to counter these disease threats. Accordingly, ID specialists are especially well-positioned to understand the risks and benefits posed by potentially dangerous experiments involving pathogenic microbes and can be valuable advisors for those who will need to undertake complicated risk-benefit analyses (RBA).

IDSA applauds the NSABB for its recent efforts to develop a framework to guide the assessment of risk and benefit of GOF research. The framework highlights key considerations on how to structure this assessment, addresses and evaluates possible alternative approaches, includes the issue of human error or malevolent action, and finally considers the effectiveness of medical countermeasures. We are happy to see

that Gryphon Scientific's risk benefit approach significantly improves on the specificity of the framework, addressing several of our concerns with the draft framework. We offer below six additional points for NSABB and Gryphon Scientific to consider as you work together to assess the risk and benefit of GOF research and develop final recommendations to the U.S. Government (USG).

1. Focus on the GOF experiments of special concern

IDSA remains concerned that the NSABB framework's broad definition of GOF may inadvertently capture areas of research that pose a lower risk to the public. For example, while the NSABB recognizes the benefit of research aiding the development or selection of new or more effective vaccines, its framework still targets influenza vaccine production methods that rely on adaptation of viruses for growth in culture as GOF research. The adaptation and manipulation of wild type influenza virus for growth in eggs or mammalian cell lines are critical to vaccine manufacturing. This approach to produce high growth vaccine candidates has been practiced since the 1940s, and is essential to protect the public from both seasonal and pandemic influenza.

IDSA strongly urges the NSABB to narrow its definition of GOF research to be considered for RBA to avoid this inadvertent capture of low risk research, which is not mentioned in the original White House description of the types of research that should be included in the deliberative process. We recommend that the RBA process focus on research that is reasonably anticipated to result in a pathogen that combines high transmissibility with high pathogenicity in humans, as this combination poses the greatest risk to public health. Such research may involve enhancing either of these properties in a pathogen already possessing the other, or the simultaneous enhancement of both. Whereas other types of GOF research are of concern as well, notably that which increases resistance to known medical countermeasures, they are secondary to the above characteristics. IDSA believes that this definition strikes a balance between impeding experiments with lower risk that society has accepted for many years while ensuring that experiments of special concern are assessed appropriately.

2. Address the uncertainty in estimating both risk and benefit

The risk assessment process provided by Gryphon Scientific will have to use estimated data in the models, as it will have to make assumptions on risks and benefits. Although IDSA understands assumptions are necessary to assess risk and benefit, our society is concerned that Gryphon Scientific has not adequately addressed the uncertainty of its models. IDSA urges the NSABB and Gryphon Scientific to hold robust discussions with experts surrounding the uncertainty of its estimates of risk. We also recommend the NSABB and Gryphon Scientific ensure that its analysis of uncertainty not only include uncertainties in the outcome of the research, such as the pathogenicity changes in a GOF organism, but also the uncertainties in the assessments of likelihood of misuse of the science as well as the consequences of accidents, misuse, and regulations on the conduct of the science. Whereas Gryphon Scientific will use a qualitative assessment of the benefit of GOF research, we urge that the uncertainties around the benefits of research be explicitly considered. Finally, IDSA recommends Gryphon Scientific consider communicating specific assumptions used in its modeling as well as error due to uncertainty to assist the NSABB and other policy makers in better understanding the risk/benefit estimates.

3. Seek a wide breadth of expertise to aid in the RBA process

Gryphon Scientific has indicated that it will interview subject matter experts to obtain additional input to aid its RBA efforts. IDSA strongly supports these actions, and also urges the NSABB and Gryphon Scientific to consider seeking additional perspectives to inform the RBA process, including those of a range of experts in vaccine development, microbial risk assessment, public health response, physicians whose work is primarily clinical, as well as through engagement of the public. In addition, the moral and ethical implications surrounding GOF research have not been adequately addressed in the NSABB framework. Several experts in this field are actively engaged in the GOF debate, and their unique viewpoints can be valuable to the RBA process.

Some stakeholders have expressed concern that the experts best positioned to evaluate the risk and benefits of GOF research are in some cases the ones who are actively conducting the research. IDSA agrees this is an issue that should be considered, and strongly believes that while this RBA evaluation needs as many expert perspectives as possible, they must be transparent with all relevant interests disclosed.

4. Risk should account for the impact on the public perception of science.

One important type of risk that is not included in the NSABB framework, or by Gryphon Scientific's mandate, is the ethical, reputational, and credibility risk for science with the public. The recent laboratory mishaps at the nation's most prestigious laboratories have placed strain on the public's trust for scientific research. Should a USG funded GOF study result in an accident or a deliberate act that places the public at risk, the credibility of science as a whole may suffer. This, in turn, could lead the public to question the quality of public stewardship of biomedical funding and the reliability of scientific and medical advice on risk. This loss of public trust could significantly impair science's ability to inform evidence-based policy decisions. IDSA recommends that the NSABB consider recruiting additional perspectives, such as those with sociology and ethics expertise, to asses this risk as it develops its final recommendations.

5. Risk should account for the impact of any new GOF framework on the course of science.

The ability of humanity to protect itself against pathogens of pandemic potential rests on a vigorous and healthy scientific enterprise. Some, including IDSA members, have raised the concern that as controversy swirls around GOF types of experiments that these fields could abandon certain types of scientific approaches that are powerful tools of scientific inquiry. Furthermore, the concern has been raised that the best and brightest will avoid these areas of inquiry simply because of the weight of regulation, the uncertainty in planning careers in areas subject to moratoriums and increased scrutiny and the controversial nature of the work. If this happens, humanity will be more vulnerable to future threats. IDSA recommends that the possible risk of regulation to the scientific enterprise and, in particular, to certain fields of inquiry be factored in the overall risk-benefit analysis.

6. Consider recommendations on how to make GOF research safer

In Gryphon Scientific's assessment approach for GOF research benefit, it states that it will evaluate "other GOF experiment types" in addition to alternative approaches. IDSA believes these efforts will yield valuable information that may be useful in developing constructive recommendations on how GOF research may be conducted more safely. For example, at the

December 2014 National Academies of Science discussion on the GOF pause, one researcher presented data on how to engineer high risk influenza strains to only undergo productive infection in experimental animals, posing minimal risk to public health. This search for pragmatic solutions that lower risk of GOF has not been widely discussed in the debate, and IDSA urges that this be a more prominent component in the NSABB's final recommendations.

IDSA is committed to ensuring that the broader scientific and science policy community participates in efforts to appropriately guide gain of function research. To complement the NSABB's efforts, IDSA calls for a continued series of transparent broad discussions on gain-of-function and dual use research of concern among stakeholders, including scientists, healthcare workers, policy-makers, ethicists, and representatives from the public. These discussions include the consideration of risk-benefit methodologies, governance models, the place, if any, of classified research, social responsibilities of scientists and journal editors, increased vigilance of biosafety and security concerns, societal values, and, finally, the discussion should solicit international input.

IDSA thanks NSABB for this opportunity to comment, and looks forward to continuing to work with the U.S. Government and those who advise it to clarify the decision-making process on how and whether to undertake high-risk life science experiments. Should you have any questions or concerns about these comments, please feel free to contact Greg Frank, PhD, IDSA Program Officer for Science and Research Policy, at gfrank@idsociety.org or 703-299-1216.

Sincerely,

Stephen B. Calderwood, MD, FIDSA
IDSA President

About IDSA

IDSA represents over 10,000 infectious diseases physicians and scientists devoted to patient care, disease prevention, public health, education, and research in the area of infectious diseases. Our members care for patients of all ages with serious infections, including meningitis, pneumonia, tuberculosis, HIV/AIDS, antibiotic-resistant bacterial infections such as those caused by methicillin-resistant *Staphylococcus aureus* (MRSA) vancomycin-resistant enterococci (VRE), and Gram-negative bacterial infections such as *Acinetobacter baumannii, Klebsiella pneumoni*ae, and *Pseudomonas aeruginosa*, and, finally, emerging infectious syndromes such as Ebola virus fever, enterovirus D68 infection, Middle East Respiratory Syndrome Coronavirus (MERS-CoV), and infections caused by bacteria containing the New Delhi metallo-beta-lactamase (NDM) enzyme that makes them resistant to a broad range of antibacterial drugs.

To: The NSABB Board (in advance of the September 28, 2015 meeting)

From: Lynn C. Klotz, PhD
Senior Science Fellow and member of the Scientist Working Group on Biological and Chemical Weapons
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The following document is a first draft of a literature-research project that started in the summer of 2015. The project is in an early stage, but has gone far enough to make its point: There is an urgent need for international proactive oversight of influenza research that might increase the pathogenicity of influenza viruses. Some of this gain-of-function research may create lab-made potential pandemic influenza viruses.

Even if the probability is small for an escape from a lab in a single year for such a virus, the fact that there are a large number of research projects underway throughout the world, projects that will be conducted for many years, the overall probability of escape from at least one lab is uncomfortably high.

The Potential Pandemic Influenza Research Enterprise

In a recent Letter to the Editor titled *Danger of Potential-Pandemic-Pathogen Research Enterprises* (http://intl-mbio.asm.org/content/6/3/e00815-15.full), I argued that there are likely many labs throughout the world, many not funded by the NIH, that are developing mammal-contagious influenza viruses. Research that makes avian, mammalian, or human influenza viruses more virulent, increases their transmissibility, alters their host range, or evades countermeasures is potentially dangerous and may create potential pandemic pathogens.

Influenza viruses are more likely to fuel an uncontrollable outbreak because of their long history of doing just that. This kind of research got considerable attention in 2011 when Professor Ron Fouchier announced that his laboratory had made the H5N1 highly pathogenic avian influenza virus (HPAI) airborne transmissible by respiratory aerosols from ferret to ferret.

In the context of this analysis of recent publications reported in Pub Med (references (1) through (35)), the larger category of Experiments of Concern (EoC) is used as a guide to look for potentially dangerous research. In 2004, the National Academy of Sciences published a report *Biotechnology Research in an Age of Terrorism* (http://www.nap.edu/catalog/10827.html). The so-called Fink Committee that produced the report was asked to "consider ways to minimize threats from biological warfare and bioterrorism without hindering the progress of biotechnology, which is essential for the health of the nation." The committee recommended that the "Department of Health and Human

Services…create a review system for seven classes of experiments (the Experiments of Concern) involving microbial agents that raise concerns about their potential for misuse." Specifically, the EoC are:

> "1. Would demonstrate how to render a vaccine ineffective. This would apply to both human and animal vaccines…
> 2. Would confer resistance to therapeutically useful antibiotics or antiviral agents. This would apply to therapeutic agents that are used to control disease agents in humans, animals or crops…
> 3. Would enhance the virulence of a pathogen or render a non-pathogen virulent. This would apply to plant, animal, and human pathogens…
> 4. Would increase transmissibility of a pathogen. This would include enhancing transmission within or between species. Altering vector competence to enhance disease transmission would also fall into this class.
> 5. Would alter the host range of a pathogen. This would include making non zoonotics into zoonotic agents. Altering the tropism of viruses would fit into this class.
> 6. Would enable the evasion of diagnostic/detection modalities. This could include microencapsulation to avoid antibody-based detection and/or the alteration of gene sequences to avoid detection by established molecular methods.
> 7. Would enable the weaponization of a biological agent or toxin. This would include the environmental stabilization of pathogens."

These seven classes of experiments "will require review and discussion by informed members of the scientific and medical community before they are undertaken [proactive oversight] or, if carried out, before they are published in full detail." For experiments making deadly avian influenza viruses airborne transmissible, many scientists think they should not be carried out at all.

An excellent system for reviewing potentially dangerous experiments, *Controlling Dangerous Pathogens: A Prototype Protective Oversight System*, was developed in 2007 by The Center for International and Security Studies at Maryland (http://drum.lib.umd.edu/bitstream/1903/7949/1/pathogens_project_monograph.pdf). It recommends a tiered review, from most to least dangerous research. Paraphrased from the Maryland paper:

> International Oversight: *Activities of Extreme Concern* – An international body would be charged with approving and monitoring all research projects of extreme concern. That authority would be narrowly focused only on those ... that could put an appreciable fraction of the human species at risk, such as research with potential pandemic pathogens.
> National Oversight: *Activities of Moderate Concern* – National oversight bodies would be responsible for research activity of moderate concern, such as work with anthrax and other agents already identified as having biological weapons potential.
> Local Oversight: *Activities of Potential Concern* – Concern—This "encompasses those activities that may increase the destructive potential of biological agents that otherwise would not be considered a threat.
> No oversight: *All other research*

In my opinion, there should be two levels of local oversight. The first level is the currently employed Institutional Biosafety Committee (IBC), and the second is an outside committee. There is concern that IBCs will simply rubber-stamp research proposals from labs in their own institution, so I suggest proactive oversight by a committee outside the institution (perhaps at the state level in the US) for experiments of concern on influenza viruses that do not carry an immediate threat of an outbreak from an escape from the laboratory (e.g., vaccine viruses and other attenuated and inactivated viruses).

Because of the potential for some strains of lab-made influenza viruses to cause international outbreaks, research mutagenizing these viruses that could result in increased pathogenicity (gain of function) should be subject to external oversight. At present, there is little national and no international proactive oversight with any authority to guide or ban experiments. See for instance: Gronvall GK, Rozo M. Synopsis of Biological Safety and Security Arrangements. UPMC Center for Health Security.July 2015. Available at http://www.upmchealthsecurity.org/ourwork/publications/synopsis-of-biological-safety-and-security-arrangements.

The literature analysis

To date, only one general Pub Med search term, "avian influenza virus mutagenesis," has been used here to identify potentially dangerous research that might fall under the Experiments of Concern (EoC). To focus on the most recent research, only research over the last two years (September 1, 2013 through August 29, 2015) published Pub Med abstracts were read. Thirty-five potential EoC were identified in 136 abstracts for this single search term. Many of the 136 abstracts (136-35=101) described research that did not constitute EoC; for the most part, they did not employ live viruses.

For each of the 35 abstracts that seemed to describe EoC, parts of the full research papers were read to confirm their EoC status. Since I have only a modest grasp of molecular virology, I may have labeled a few that are not EoC, and I may have missed a few that are EoC.

The actual number of EoC research being carried out today is likely much greater than 35 because of the following:

- Only a single avian influenza search term was used; other influenza search terms would yield additional EoC. In particular, viruses that have already caused pandemics such at the 2009 H1N1 virus.
- Expanding the search back to 2012, and even before that, would yield more EoC.
- There are surely some EoC that are not yet published.
- Search terms involving other pathogens such as SARS, MERS and Ebola would yield more EoC.

A summary of the 35 EoC found from the search is provided in Table 1. Titles and citations for the reference numbers are in the reference list at the end.

Reference Number	Countries of Authors	Viruses	Biosafety Level	EOC Category
1	USA, Korea	H1N1 vaccine strain	?	2
2	**China**	Avian, human H6N1	BSL3	5
3	**China**	H5N1 HPAI	not reported	1, 3
4	USA, Egypt	H5N1 HPAI	?	1, 3
5	**China**	H9N2 avian	BSL3	3, 5
6	**Japan**, USA	H5N1 HPAI	BSL3	3, 5
7	**Netherlands**, UK	H1N1 2009	BSL2	1, 3
8	**China**	H5N1 HPAI	Not reported	3
9	**China**	H7N1 avian	BSL3	3, 5
10	**China**	H9N2, H1N1 2009, H5N1 HPAI	BSL3, BSL3+	3
11	**USA**, Japan	H5N1 HPAI	BSL3	3
12	**China**	H6N1 avian	??	3, 5
13	**Japan**, Thailand	H5N1 HPAI	BSL3	3
14	**China**	H9N2 duck	ABSL3+	3, 5
15	**France**	avian H1N1	BSL3+	3, 5
16	**USA**	A/WSN/1933 H1N1	likely BSL2	5
17	**Netherlands**	airborne trans H5N1 HPAI	animal BSL3+	3, 4
18	**China**	H7N9 HPAI	ABSL3	3
19	**Japan**	H7N9 HPAI	BSL3+	3
20	**China**	H1N1 2009 pandemic	not reported, BSL2?	3
21	**China**	H1N1 2009 pandemic	not reported, BSL2?	2
22	Spain, UK	influenza A vaccine strains	assume BSL2	3
23	**Netherl.**, Germany	HPAI H5N1	BSL3+	1
24	**USA**	H1N1 vaccine strain	assume BSL2	1
25	**USA**	H3N2	BSL2?	2
26	**Germany**	HPAI H5N1	BSL3+	1
27	**China**, USA	HPAI H5N1	BSL3, ABSL3	2
28	**Russia**	nonpath H5N2, HPAI H5N1	not reported, BSL2?	1
29	**Germany**	1968 pandemic H3N2	not reported, BSL2?	3?
30	**USA**	H1N1 vaccine strain	not reported, BSL2?	1
31	**USA**	HPAI H5N1	BSL3	3, 5
32	**UK**	HPAI H5N1	BSL3	3, 5
33	**USA**	H3N2, H1N1	not reported, BSL2?	3?
34	**USA**	HPAI H5N1	ABSL3+	3, 4
35	**USA**	human H3N2, HPAI H5N1	ABSL3+	1

Table 1: The 35 EoC. The boldface in the Countries of Authors column indicates the country where the BSL2, BSL3 research was performed. Much of that research is being carried out in Asia, particularly China.

The 35 published research listed in the Table are described briefly below. The descriptions are a combination of quotes from the Pub Med abstracts and full papers, often paraphrased to make them readily understandable with regard to EoC. The numbers, 1 through 35, at the beginning of each entry below correspond to the numbered reference citations at the end of this document. The **bold-face highlighted** descriptions are the greatest concern in my opinion because the mutated viruses are often more pathogenic than the wild-type strains and are potentially airborne transmissible from human to human.

1. Recombinant influenza viruses were made that have single or double substitutions in neuraminidase N3, N7 and N9 subtypes in a background of an H1N1 vaccine strain. N3, N7 and N9 subtypes have caused human infections. The research discovered resistance to neuraminidase inhibitors in some strains. [Comment: Mutagenesis of vaccine strains are not of the highest concern, unless there is reason to believe that the mutagenesis could make the strain virulent.]

2. Avian H6N1 virus was adapted to human receptor-binding. Receptor-binding was analyzed using isolated H6 proteins. Binding was confirmed using two avian and one human-derived H6N1 recombinant viruses. The research found two HA substitutions important to acquire the human receptor-binding. [Comment: Only one case of human H6N1 infection has been reported to date. Could increasing receptor binding in humans lead to more human cases?]

3. Site-directed mutagenesis was used to generate different patterns of stem glycans on the HA protein of an HPAI H5N1. The results indicated that some glycans were dispensable for the generation of replication-competent influenza viruses. Some combinations of glycans led to a significant decrease of

the growth rates of the mutant viruses in animal cells in comparison to wild type virus. Furthermore, most of the mutant viruses were more sensitive to neutralizing antibodies than the WT virus. [Comment: Could researchers predict results in advance? These are experiments that should be proactively reviewed, as some mutations could have increased virulence or avoided existing vaccines. The outcome is, however, reassuring]

4. Variant H5N1 viruses with five mutations in the HA gene were made. The research indicated that targeted mutation in the HA may be effectively used as a tool to develop broadly reactive influenza vaccines to cope with the continuous antigenic evolution of viruses. [Comment: Could researchers predict results in advance? These are experiments that should be proactively reviewed, as some mutations could have increased virulence or avoid existing vaccines. As viral mutant population sizes are huge, the probability of finding an adaptive mutation is pretty large for RNA viruses.]

5. The research found three mutations in HA, N and PB2 proteins that after four passages conferred high virulence to H9N2 virus in mice. Adaption in mice enhanced the viral polymerase activity and receptor-binding ability, which resulted in a virulent phenotype in mice but not a transmissible phenotype to guinea pigs. [Comment: This additional guinea pig experiment was useful to reduce concern or fear over increased host range.]

6. Mutations made in the PA protein enhanced HPAI H5N1 virus growth capability in human lung cells and increased pathogenicity in mice, suggesting that they contribute to adaptation to mammalian hosts.

7. Mutants made with substitutions in the hemagglutinin of a strain of 2009 H1N1 pandemic influenza virus revealed that single substitutions affecting the loop adjacent to the receptor binding site caused escape from ferret and human antibodies elicited after the 2009 H1N1 pandemic influenza virus infection. The majority of these substitutions resulted in similar or increased replication efficiency *in vitro* compared to that of the virus carrying the wild-type hemagglutinin. However, none of the substitutions was sufficient for escape from the antibodies in sera from individuals that experienced <u>both</u> seasonal and pandemic H1N1 virus infections. [Comment: This is the virus that infected 25% of the world population world-wide in 2009 and killed thousands of people. Any experiment that increases replication efficiency or escapes antibodies should not be carried out in BSL2.]

8. Mutant HPAI H5N1 viruses made with loss of two HA protein glycosylation sites showed increased pathogenicity, systemic spread and pulmonary inflammation in mice compared to the wild-type H5N1 virus.

9. Two mouse-adapted variants of wild-type avian H7N9 made by independent serial passages in mice confer enhanced virulence in mammals. [Comment: This virus has infected and caused fatalities in humans from direct contact with poultry. It would have been informative if the researchers had carried out a single ferret to ferret transmission experiment to see if this mouse-passaged virus has increased host range and virulence in a species (ferrets) that is perhaps a model for humans.]

10. Mouse-adapted PB2 gene reassortants with a phenylalanine-to-leucine mutation contributes to enhanced polymerase activity, enhanced replication, pathogenicity of H9N2 in mice, increased

virulence of H5N1 and 2009 pandemic H1N1. [Comment: Could increasing virulence in the 2009 pandemic flu cause a new outbreak among humans?]

11. The introduction of an arginine residue into PA of HPAI H5N1 significantly increased the viral polymerase activity in mammalian cells and its virulence and pathogenicity in mice.

12. A substitution in the PB2 protein and a substitution in the PA protein enhance virulence and expand the tropism of H6N1 virus in mice. [Comment: Only one case of human H6N1 infection has been reported to date. Could increasing virulence and tropism in humans lead to more human cases?]

13. Introduction of a single substitution into PB1 polymerase of an HPAI H5N1 increased both polymerase activity in chicken cells and the pathogenicity of the recombinant viruses in chickens. [Comment: This translates to humans.]

14. A nonpathogenic duck-origin H9N2 virus was serial-passaged in mouse lungs. Increased virulence was detectable after five passages, and a highly pathogenic mouse-adapted strain was obtained after 18 passages. There were eight amino-acid substitutions in six viral proteins. [Comment: Since serial passage was in lungs, this kind of research could lead to airborne transmission. A single ferret to ferret passage experiment should have been carried out to see if airborne transmission was achieved.]

15. A deletion in the NS segment of a duck-origin avian H1N1 virus showed both increased replication potential and an increased pathogenicity in chicken embryonated eggs and in a chicken lung epithelial cell line.

16. Mutants created in the PB2 subunit identified critical residues required for general polymerase function and specific residues preferentially required in human but not avian cells. [Comment: It is unclear what virus was used in the study. It may have been PB2 mutants reassorted into A/WSN/1933 H1N1 virus. A/WSN/1933 is a derivative of 1918 flu virus and is not around today. This is a mouse brain adapted virus so not a threat.]

17. Five substitutions proved to be sufficient to retain the airborne-transmissible phenotype of HPAI H5N1. [Comment: A large number of substitution experiments on an airborne transmissible, deadly virus were carried out in this study, and a large number of nose and throat swabs and blood samples were taken, all increasing significantly the likelihood of an LAI. This is follow-up research from the Fouchier lab.]

18. An H7N9 virus from a fatal case was used as the recombination background to study the contribution of the E627K mutation in PB2 and of other mutations to the pathogenicity of H7N9 virus infection in mammals. All the mutant viruses generated were likely to be loss-of-function mutants with regard to pathogenicity, compared to the wild-type H7N9. [Comment: The research appears to yield less pathogenic H7N9. Nonetheless, it is not possible to predict pathogenicity at the outset of the experiments. Since the background virus is a fatal case; proactively, the generated viruses could have been more virulent humans. It would have been informative if the researchers had carried out a single

ferret to ferret transmission experiment to see if this virus was more virulent in ferrets, the model for human lung.]

19. Potentially mammalian adapting amino acids were converted individually and in combination to their avian virus-type counterparts in a H7N9 virus. Several mutants were slightly more virulent in mice than the wild-type A(H7N9) virus and exhibited increased polymerase activity in human cells.

20. A single "consensus" PB2 mutation common to swine and the 2009 H1N1 pandemic virus increased pathogenicity. Mutant virus prepared by recombination of a 2009 H1N1 pandemic virus with a segment containing the single PB2 mutation significantly enhanced polymerase activity in mammalian cells. Also, the virus exhibited increased growth properties and induced significant weight loss in a mouse model compared to the wild type. [Comments: This more pathogenic virus could win the battle with the immune system, so cause significant illness.]

21. Reduced sensitivities to oseltamivir were observed in three mutant H1N1 2009 pandemic viruses. A double mutant showed a large increase of IC-50 for the drug Oseltamivir from 0.7 nM for WT to 4,000 nM for the double mutant, a 5,700-fold difference [Comment: Such a large increase in IC-50 would almost certainly make the drug unusable in humans.]

28. A non-pathogenic avian H5N2 was adapted to mice by lung-to-lung passage. Also, the reverse genetics-derived influenza virus containing the HA and NA genes of an HPAI H5N1 in the genetic background of a high-growth H1N1 vaccine strain was obtained. Antibody escape mutants using these two viruses were obtained. Monitoring of effects of HA mutations found in H5 segment escape mutants is essential for accurate prediction of mutants with pandemic potential. [Comment: While H5N2 does not appear to have caused any human infections, adapting it to mice by lung to lung passage could have made it virulent in humans and even airborne transmissible.]

29. Influenza A viruses circulating in humans from ~1950 to ~1987 featured a nonstructural (NS1) protein with a C-terminal amino acid extension present in the H3N2 1968 pandemic flu virus. This research deleted the NS1 extension in the H3N2 in order to compare the wild type H3N2 with the virus with the NS1 deletion. The replication kinetics of the wild-type H3N2 and the deletion mutant were indistinguishable in most experimental systems. However, wild-type virus out-competed the mutant during mixed infections, suggesting that the NS1 extension conferred minor growth advantages. [Comment: The resurrection/rescue of an historical pandemic virus is potentially as dangerous as a lab-made PPP if it escapes from the laboratory, provided that the virus employed is identical to or very close to the 1968 pandemic strain.]

31. A particular point mutation in the PB2 protein of HPAI H5N1 virus, PB2 627K, has been identified as a virulence and host range determinant for infection of mammals, and is present in strains capable of airborne transmission. This mutation in the PB2 gene appeared from day 4 and 5 along the respiratory tracts of mice inoculated intranasally and was complete by day 6 post-inoculation. The mutation correlated with efficient replication of the virus in mice. [Comment: This kind of experiment may be on a path to an airborne transmissible strain.]

32. This research focused on the particular PB2 point mutation in Reference 31, just above. Viruses constructed by reverse genetics were made to contain converse PB2 627K/E mutations in a Eurasian HPAI H5N1 virus and, for comparison, a historical pre-Asian HPAI H5N1 virus that naturally bears PB2 627E. Effects on viral fitness were observed in *in vitro* or *in vivo* experiments. Results suggest that the PB2 627K mutation supports viral fitness in Eurasian-lineage viruses; in contrast, the mutation carries a significant fitness cost in a historical pre-Asian virus.

34. Influenza virus entry is mediated by the acidic-pH-induced activation of HA protein. This research investigated how a decrease in the HA activation pH influences the properties of highly pathogenic H5N1 influenza virus in mammalian hosts. Viruses containing either wild-type HA or an acid-stabilizing point mutation were prepared. Wild-type and viruses with the mutation promoted similar levels of morbidity and mortality in mice and ferrets. The mutation was found to enhance the growth of an H5N1 influenza virus in the mammalian upper respiratory tract, and yet it was insufficient to enable contact transmission in ferrets. Neither virus transmitted efficiently to naive contact cage-mate ferrets. [Comment: It is fortunate that contact transmission was not found.]

35. The research focused on an antigenic cluster associated with a natural single hemagglutinin (HA) substitution that occurred between 1992 and 1995 in the H3N2 virus. Reverse-genetics experiments demonstrated that the HA mutation increases viral receptor binding avidity. The mutation does not prevent antibody binding; rather, viruses possessing this mutation escape antisera simply because the virus attaches to cells more efficiently. [Comment: The H3N2 virus has caused human infections when transmitted from swine. In a 2012 small outbreak, there was no evidence of community transmission. Nonetheless, the virus is an immune escape strain.]

While the search term was not designed to pick up the 2009 human pandemic H1N1 virus, it did pick up a few experiments involving mutagenesis of that strain. While some of this research is carried out at BSL2, it could be classified as research of great concern because that virus is airborne transmissible.

For research involving mutagenesis of vaccine strains, biosafety level was generally not reported. It is assumed that it is BSL2, as vaccine strains are attenuated or inactivated viruses. One concern is that some mutagenesis research could make a vaccine strain virulent. Researchers should be prepared to argue for the safety of their particular proposed vaccine-strain mutagenesis research to defend the lower BSL2 containment.

Several of the EoC (references 7, 10, 14, 17, 20, 21, 28, 29) are lab-made potentially dangerous influenza viruses that could spread from human to human by the airborne route.

Proactive review at the local, national, or international level that considers risk and value (benefits) should be considered before allowing any mutagenesis and related research that might result in Experiments of Concern to go forward, and under what conditions.

Conclusion

Research that employs, makes, or could make airborne transmissible strains is of the greatest concern. All this research should be subject to proactive international review and oversight. There is an urgent need for a binding international process. While the NSABB mandate is likely restricted to NIH-funded research or perhaps any research in the United States, it behooves the NSABB to urge the State Department to seek a binding international agreement for proactive review and oversight of potential pandemic research.

I would like to thank Simon Wain-Hobson for comments and insights.

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Comments on the September 28, 2015 NSABB meeting

The comments below are a written version of oral comments presented during the public comment period at the NSABB Sept 28, 2015, meeting on the "robustness" of the process for regulation of GOF research that was in place before the funding pause. By this I mean the general DURC frameworks and the HHS frameworks for H5N1 (1) and H7N9 (2) GOF research. On paper, these processes sound robust. We have some historical record of how the process works – one example of which was published by *Nature* in the case of the University of Wisconsin and the reconstruction of a 1918-like virus (http://www.nature.com/polopoly_fs/7.18249!/file/WISC_Review.pdf). Based on the characteristics of the process so far, there are several areas of concern that in my judgment make that process less than robust.

The fact that the existing DURC process did not even flag PPP research as a separate issue until a confluence of accidents at prominent labs and public activism forced the issue, is a . Ironically, the extension by HHS of the Framework to H7N9 GOF research (2) appeared in the same issue of *Nature* in as a report of GOF studies from the Fouchier lab, funded by the US Government and not captured by this framework (3); see also http://comments.sciencemag.org/content/10.1126/science.1244158 .

More specifically the present Framework for H5N1 and H7N9 GOF that was in place before the funding pause has the following issues:

1. *Expertise*. Much of the responsibility for assessing risks and benefits under the current system lies with the institutional biosafety committee. These committees are mainly composed of laboratory scientists and laboratory safety experts. These committees are essentially expert in occupational health. The difference with pandemic risk is that the risk is a public health, possibly global risk. IBCs do not traditionally include epidemiologists who might be able to identify what is a potential pandemic pathogen experiment or what the likely magnitude of risk would be[1].

   IBCs are not well designed to consider such risks. If you read the IBC minutes from the University of Wisconsin that have been posted by *Nature* magazine, it is clear that the claims of the investigator are often accepted at face value. Most IBCs also have little or no expertise in biosecurity threats. Note that I am not criticizing IBCs' fitness for their traditional task of dealing with occupational health risk of most pathogens in the lab. I am criticizing their fitness for playing the same role in managing global public health risk, an issue that uniquely arises in the potential pandemic pathogen context.

[1] Prof. Yoshi Kawaoka has informed me that the University of Wisconsin IBC includes an infectious disease physician and a representative from the state Division of Communicable Diseases. I do not know whether these areas of expertise were represented at the meeting that approved the 1918-like virus work.

2. *Disinterestedness*. The current process for oversight depends mainly on the funders and the recipients of funding. Neither of these is a disinterested party. Institutional biosafety committees very often see their role as facilitating the research that they regulate and whose indirect costs support the IBC's activities. This may be another reason why IBCs and other reviewers have been prone to accept the claims of investigators, especially on the benefits, at face value even when they are aspirational.
3. *Quantitative considerations*. To my knowledge the existing process makes no effort to quantify risk, either at the IBC level – where we have a written record, or at the HHS level.
4. S*cope*. The policy applies only to institutions applying to the USG for funding for unclassified life sciences research, not to classified research or to non-HHS-funded research.

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From: David Fedson [mailto:dfedson@wanadoo.fr]
Sent: Sunday, December 13, 2015 9:35 AM
To: Viggiani, Christopher (NIH/OD) [E]
Cc: Opal, Steven
Subject: NSABB Meeting on GOF research on January 7-8, 2016

Christopher Viggiani, Ph. D.
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Dear Dr. Viggiani,

I have reviewed the agenda of the NSABB meeting on January 7-8, 2016. At this meeting, the NSABB will discuss its Working Group's overview of progress, preliminary findings and draft working paper on Gain-of-Function (GOF) studies. The Gryphon Scientific report - "Risk and Benefit Analysis of Gain of Function Research, Final Report - December 2015" - will be presented at this meeting.

I would like to bring to your attention and that of the NSABB several important points.

1. If GOF research accidentally or deliberately creates a new highly virulent and highly transmissible influenza virus, it will spread throughout the world in a matter of months. The ensuing pandemic will be a global event, and it will require a global response.

2. Ron Fouchier has said that Mother Nature is the biggest bioterrorist. Pandemic influenza viruses can arise not only in nature but also in experimental circumstances. In a paper published 1974, Webster and Campbell described how they created in turkeys a new transmissible influenza reassortant virus that led to a 100% population die off (attachment 1). This GOF research was conducted more than 40 years ago.

3. In the event of a global pandemic caused by a highly virulent, highly transmissible influenza virus, regardless of its provenance, none of our current medical countermeasures (vaccines, antivirals) will be available to meet the needs of more than 90% of the world's people (attachment 2).

4. When a new pandemic virus appears, the most important question to ask is "what next?" In 2013, Professor Steven Opal at Brown University and I published a paper on GOF research in which we addressed this question. We described an approach to treating pandemic patients using widely available, inexpensive generic drugs that target the host response to infection, not the virus itself (attachment 3).

5. In late 2014, physicians in Sierra Leone treated approximately 100 patients with Ebola virus disease with a combination of a statin (atorvastatin) and an angiotensin receptor blocker (irbesartan). This treatment targets the host response to Ebola virus infection, not the Ebola

virus. Only three inadequately treated patients are known to have died (attachment 4). This treatment reverses the endothelial dysfunction that is central to the host response to Ebola virus disease. It could probably also be used to treat pandemic influenza, MERS, SARS, and other life-threatening diseases in which endothelial dysfunction leads to an increased risk of multi-organ failure and death.

5. Research on treating the host response to influenza and Ebola has been ignored by scientists and government agencies in the US and elsewhere. It is not on WHO's agenda for pandemic preparedness (see attachment 2) or the Ebola response. I have not read the complete Gryphon Scientific report, but the article in attachment 3 is not mentioned in any footnote in its first 486 pages, and it appears not to have been discussed in the text.

6. Given our inability to predict the specific pathogen that will cause the next epidemic, pandemic or biosecurity crisis, the only sensible way to prepare for this event is to identify effective medical countermeasures that address the pathophysiological disturbances common to them all.

Discussion of the risks and benefits of GOF research should focus on practical measures that could be used to counteract this and any other threat to biosecurity. Thus far, the NSABB has not done this. The need for research on treating the host response to emerging biosecurity threats should be discussed by the NSABB. It should be placed on the agenda of the Second Symposium on GOF Research that the National Academies will convene on March 10-11, 2016.

I would be grateful if you would forward copies of my letter and the attachments to Drs. Stanley, Berns and Kanabrocki.

If you have questions about any of these issues, please do not hesitate to write.

With best regards,

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Attachments

1. **A "bottom up" treatment for Ebola that could have been used in West Africa**
2. **How Will Physicians Respond to the Next Influenza Pandemic? -- CID, 2014**
3. **The controversy over H5N1 transmissibility research: An opportunity to define a practical response to a global threat -- Hum. Vaccin. Immunother., 2013**

A "bottom up" treatment for Ebola that could have been used in West Africa

More than 11,000 people have died as a result of the Ebola outbreak in West Africa. Aside from conventional supportive care, no specific treatment has been available. In most treatment units, more than 50% of the patients have died. This needn't have happened.

Patients who die of Ebola have elevated plasma levels of pro-inflammatory cytokines. The same thing is seen in patients with sepsis, and in sepsis patients these findings are associated with endothelial dysfunction and the loss of endothelial barrier integrity [1-3]. Careful studies of foreign healthcare workers who were infected with Ebola virus and evacuated from West Africa for medical care showed they had developed massive fluid losses. These losses were due to a dramatic increase in vascular permeability, a direct effect of the loss of endothelial barrier integrity.

Cardiovascular scientists have known for many years that several common drugs, among them statins and angiotensin receptor blockers, have the ability to stabilize or restore endothelial barrier integrity. These drugs are safe when given to patients with acute critical illness, and clinical studies suggest they might improve survival in patients with sepsis, pneumonia and influenza [1, 3]. For these reasons, in November local physicians in Sierra Leone treated consecutively approximately 100 Ebola patients with a combination of atorvastatin (40 mg orally /day) and irbesartan (150 mg orally/day) [4-7]. Only three inadequately treated patients are known to have died. Unfortunately, apart from a private donation of $25,000, there was no financial or logistical support to conduct a proper clinical trial. Surprisingly, physicians and health officials in Sierra Leone have refused to release information on this treatment experience. Nonetheless, letters and memoranda they have exchanged provide good evidence that treatment brought about "remarkable improvement" in these patients.

Unlike experimental treatments (antiviral drugs, convalescent plasma) currently being tested in Ebola patients, atorvastatin and irbesartan target the host response to the infection, not the virus itself [3-7]. By stabilizing endothelial function and restoring normal fluid balance, combination treatment allows patients to live long enough to develop immune responses of their own and get rid of the virus.

All physicians who treat patients with cardiovascular diseases are familiar with atorvastatin and irbesartan, and most of them have used these drugs to treat their patients. They are widely available as inexpensive generics in West Africa. A 10-day course of treatment for an individual Ebola patient would cost only a few dollars.

Details on the Ebola patients who were treated need to be released, and these findings need to be externally reviewed and validated. Surprisingly, no one seems interested in doing this [8]. If cases of Ebola continue to occur, combination treatment should be tested in a proper clinical trial. In the meantime, physicians should consider the possibility that this combination might be used to treat patients with any form of acute infectious disease, including pandemic influenza [9], in which failure to overcome endothelial dysfunction often leads to multi-organ failure and death.

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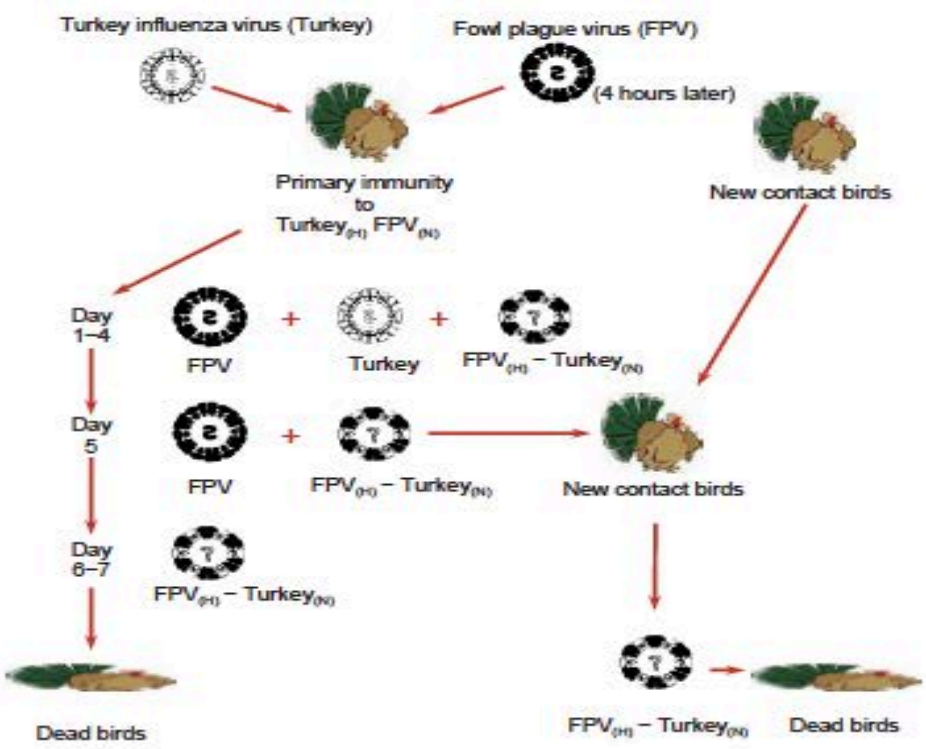

Figure. Genetic reassortment and genesis of a new pandemic influenza virus. This study was designed to determine whether the selection and transmission of a new reassortant influenza A virus could occur under experimental conditions in vivo that mimic what might occur in nature. Reassortment between 2 antigenically distinct influenza A viruses was studied in turkeys that had been previously immunized to induce low levels of antibodies to the hemagglutinin (H) of a nonlethal turkey influenza virus (Turkey), and to the neuraminidase (N) of a fowl plague virus (FPV), an avian virus that is highly pathogenic for chickens. Twenty-eight days after immunization, the immunized turkeys were sequentially infected, first with the Turkey virus and 4 h later with FPV. During the first few days, both parent viruses were isolated from the infected turkeys, but by day 4 a reassortant virus containing the FPV hemagglutinin and the Turkey neuraminidase ($FPV_{(H)}$–$Turkey_{(N)}$) was also isolated; within 2 days it became the dominant virus. All infected turkeys died, and only the $FPV_{(H)}$–$Turkey_{(N)}$ reassortant virus could be recovered. In a separate experiment, similarly immunized turkeys were again sequentially infected, but on day 5 a group of nonimmunized or selectively immunized turkeys ($Turkey_{(H)}$ $FPV_{(N)}$) were placed in the same room. All contact birds soon died of fulminant infection caused by the $FPV_{(H)}$–$Turkey_{(N)}$ reassortant virus. These experiments demonstrated that under conditions of selective primary immunity, a new virus could be generated through genetic reassortment in vivo and that this reassortant virus could be readily transmitted to contacts. The reassortant virus caused uniformly fatal disease in primary infected and contact birds. Thus, under the conditions of these experiments, genetic reassortment gave rise to a new influenza virus that led to a total population collapse. Adapted from Webster and Campbell (9).

Fedson DS. Meeting the challenge of pandemic preparedness in developing countries Emerg Infect Dis 2009; 15: 365-71. Adapted from Webster RG, Campbell CH. Studies on the origin of pandemic influenza. IV. Selection and transmission of 'new' influenza viruses in vivo. Virology 174: 62: 404-13.9

VIEW POINTS

How Will Physicians Respond to the Next Influenza Pandemic?

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The emergence of the H7N9 virus in China is another reminder of the threat of a global influenza pandemic. Many believe we could confront a pandemic by expanding our capacity to provide timely supplies of affordable pandemic vaccines and antiviral agents. Experience in 2009 demonstrated that this cannot and will not be done. Consequently, physicians may have little more to offer their patients than they had in the 1918 pandemic. Fortunately, several modern drugs (eg, statins, angiotensin II receptor blockers, angiotensin-converting enzyme inhibitors) can modify the host response to inflammatory illness, and laboratory and clinical studies suggest they might be used to treat pandemic patients. Unfortunately, little attention has been given to the research needed to support their use in patient care. There is no guarantee these drugs will work, but physicians will never know unless those responsible for pandemic preparedness recognize and act on the extraordinary possibility that they might save lives.

Keywords. pandemic influenza; statins; immunomodulatory agents; public health.

The recent emergence of the influenza A(H7N9) virus in China has led to a limited outbreak of disease that has been associated with an overall mortality of approximately 30% [1–3]. The impact has been especially severe among the elderly. It is widely known that influenza viruses can modify or exchange their genes, and these changes often yield new viruses with altered virulence and/or transmissibility. An experiment published in 1974 showed that infecting turkeys with 2 different influenza viruses generated a new reassortant virus that killed all of the infected birds and all of their contacts—a 100% population collapse [4]. The influenza pandemic of 1918 killed between 50–100 million people worldwide, and epidemiologists estimate that a similar pandemic today could kill 62 million people [5], almost twice the number that have ever died of AIDS. Since 1997 there has been deep concern about the high mortality (≥50%) seen in human infection with the avian influenza A(H5N1) virus, and recent controversy over H5N1 gain-of-function research has heightened this concern [6]. Billions of dollars have been spent preparing for an H5N1 pandemic. It is no wonder that scientists and health officials are worried about the H7N9 virus [7].

Several commentators writing in journals that target practicing physicians in the United States have expressed concern that the H7N9 virus could evolve to become easily transmissible and lead to a devastating global pandemic [8–10]. Many believe that the most effective way to respond to the next pandemic would be to greatly expand our capacity to rapidly produce influenza vaccines. They have been encouraged by new developments in influenza vaccinology, especially those based on antibodies and cytotoxic T lymphocytes that mediate heterotypic protection against influenza virus infection [11]. Targets for these new vaccines include the stem cell region of the hemagglutinin molecule and several internal proteins (eg, M2e, NP, M1, and NA). Many believe that research on these targets could lead to a universal influenza vaccine that would obviate the need for annual immunization and provide a foundation of protection against the next pandemic. Other developments in influenza vaccinology include (1) rapid

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preparation of seed strains for vaccine production using reverse genetics; (2) expanded cell culture vaccine production facilities; (3) recombinant glycoprotein HA antigens produced in pharmaceutical bioreactors; (4) antigen-sparing adjuvants that increase the number of vaccine doses that could be produced; and (5) monovalent live attenuated pandemic vaccines [12]. However, enthusiasm for these new developments in influenza vaccinology must be tempered by recognizing that they alone will not guarantee the success of pandemic vaccination.

If vaccination against a global pandemic is to succeed, other measures will be required [12]. New facilities for vaccine formulation and filling will be needed, experienced production technicians must be trained, supplies of syringes and needles for administering inactivated vaccines must be secured, clinical trials of candidate vaccines must be supported, procedures for rapid regulatory certification must be put in place, commercial arrangements between vaccine companies and patent holders must be worked out, advanced purchasing agreements and prices must be negotiated between companies and governments, the logistics of vaccine distribution must be set up, and a human infrastructure for vaccination programs must be established. In each country, the cumulative impact of these factors will directly affect the ability of vaccination programs to successfully confront the next pandemic [12].

The most important factor that will determine the global success of pandemic vaccination will be the level of expansion of seasonal influenza vaccination programs, especially in countries that currently use little vaccine [12]. This will require better understanding of the burden of influenza disease and the effectiveness of influenza vaccination. Remarkably, in recent years the global production capacity for seasonal influenza vaccines has increased to the point where it exceeds world demand, yet there is little evidence that demand will soon match production capacity [13]. In all likelihood, expansion of seasonal vaccination will depend on whether governments in low-use countries recommend and purchase influenza vaccines. In the absence of such decisions, implementing new advances in influenza vaccinology "will depend on company assessments of their individual scientific, technical and commercial advantages. These assessments will be viewed within the context of seasonal not pandemic vaccination" [12].

The global vaccination response to the influenza A(H1N1) pandemic in 2009 offers little encouragement that things will be much better for the next pandemic [14]. In the United States, because pandemic vaccines were not available in time, vaccination affected only 2%–4% of all pandemic cases, hospitalizations, and deaths (see Tables 3–5 of [15]). Consequently, health officials had to advise people to wash their hands and limit social contacts, a throwback to 19th-century public health "technologies." Although the vaccine and antiviral response in the United States was minimally effective, for most of the world it was a comprehensive failure: >90% of the world's people had no access to timely supplies of affordable pandemic vaccines [16].

The threat of another influenza pandemic, H7N9 or otherwise, is real [4–10]. If it is severe, hospitals and intensive care units will be swamped with patients. Extracorporeal membrane oxygenation treatment will help only a few. Even if excellent medical care (including antiviral agents) is available, experience with H7N9 and H5N1 influenza has shown that mortality rates could still be high. Wherever such care is not available, especially in low- and middle-income countries, the mortality impact of a global pandemic could be devastating. Although physicians in most countries will find themselves in healthcare settings much different from those in 1918, their experiences and those of their patients could be much the same [17]. Given this possibility, physicians everywhere need to ask whether agents they already know and use in the routine care of their patients might also be used to treat those who become seriously ill with pandemic influenza.

Until now, health officials have relied on influenza scientists—primarily virologists and epidemiologists—to guide pandemic preparedness efforts. Virologists who have adopted a systems approach to discovery have made important contributions to explaining influenza virus–host interactions and the consequences of these interactions for the pathogenesis of disease [18]. Nonetheless, they have yet to suggest agents that would be available to physicians who will be called upon to manage severely ill pandemic patients. Fortunately, investigators in other fields, especially cardiovascular and metabolic diseases, have developed several groups of drugs whose "pleiotropic" activities modify the innate and adaptive immune response to acute inflammatory illness. These drugs might be used for pandemic treatment and prophylaxis. Statins were the first group suggested [19], and since then angiotensin II receptor blockers (ARBs), angiotensin-converting enzyme (ACE) inhibitors, peroxisome proliferator-activated receptor (PPAR) γ and PPARα agonists (glitazones and fibrates, respectively), and adenosine monophosphate–activated kinase agonists (eg, metformin) have emerged as additional candidate agents. These developments have been comprehensively reviewed in a recent publication [16]. Laboratory studies of acute lung injury, sepsis, and other forms of acute systemic inflammation have shown that these drugs control damaging inflammation, promote its resolution, and improve survival [16, 20, 21]. The benefits of treatment may have little to do with the effects of these drugs on influenza virus–infected cells [16]. Instead, they might improve survival by maintaining or restoring pulmonary microvascular barrier integrity [22], accelerating the early return of mitochondrial biogenesis [23], and/or promoting beneficial changes in immunometabolism [24–26]. Laboratory and clinical research on these agents might help us understand why influenza mortality rates are lower in children

than in adults [16], and perhaps show that "disease tolerance" in children with influenza is a defense strategy that reflects the heritage of human evolution [16, 27–29].

Clinical studies support laboratory findings on the effectiveness of inpatient treatment with 3 groups of these agents (reviewed in [16]). For example, an observational study of 3043 patients hospitalized with laboratory-confirmed seasonal influenza showed that statin treatment was associated with a 41% reduction in 30-day mortality [30]. This reduction was in addition to any that might have been attributable to previous vaccination and antiviral treatment. Another observational study showed that inpatient treatment with ARBs, ACE inhibitors, and statins reduced 30-day pneumonia mortality by 53%, 42%, and 32%, respectively [31]. Importantly, a randomized controlled trial in 100 statin-naive patients (untreated for at least 2 weeks) who were hospitalized with sepsis showed that inpatient atorvastatin (40 mg per day) reduced progression to severe sepsis by 83% (24% in control patients vs 4% in treated patients; $P = .007$) [32].

Statins and other immunomodulatory agents that might benefit influenza patients are used by physicians every day to treat millions of patients with cardiovascular diseases and diabetes. For statins, long-term treatment is safe and effective in improving cardiovascular outcomes, and the benefits greatly outweigh the modestly increased risks of statin-associated diabetes, elevated liver enzymes, and myopathy [33], adverse events that are easily managed. Cases of severe liver injury or rhabdomyolysis are rare. For short-term inpatient treatment, cardiologists routinely initiate statin treatment in patients hospitalized with acute coronary syndrome (ACS), and such treatment has shown to be safe and effective in reducing hospital and 30-day ACS mortality (reviewed in [16]). This experience suggests that studies of treating influenza patients with statins and other immunomodulatory agents should focus on those with illness serious enough to require hospitalization, and an agenda for such research has recently been presented [16]. This research will allow physicians to carefully assess the clinical and immunological effects of treatment while monitoring patients for any signs of adverse events or drug–drug interactions. Special attention will have to be given to the safety of treating pregnant women and children.

Several small-scale studies of statin treatment in humans with experimental acute lung injury, sepsis, and pneumonia have been published (reviewed in [16]). Although these studies were too small to show evidence of clinical benefit, no adverse reactions were noted and several parameters associated with immune dysregulation showed improvement. If statins or other immunomodulatory agents could be shown to be safe and effective, treatment for most patients (especially those who are not older adults) would probably be limited to the duration of the hospital stay and would not need to be continued after hospital discharge. For hospitalized patients who have previously received outpatient treatment with any of these agents, continued treatment after hospital admission would probably be indicated, just as it is for ACS patients who have received outpatient statins [16].

All of the immunomodulatory agents discussed above are now produced as inexpensive generics in developing countries, and global supplies are huge [16]. If 1 or more of them were shown to be safe and clinically effective in treating severe influenza (or in the syndromic treatment of acute critical illness due to other causes such as pneumococcal pneumonia [34]), they would be immediately available to physicians in any country with a basic healthcare system. The cost of treating an individual patient would probably be less than $1.00 [16]. Nonetheless, the laboratory and clinical research needed to justify using these agents to treat influenza patients must be initiated and supported by governments and/or nongovernmental institutions; it cannot be left to pharmaceutical companies because the drugs are no longer of commercial interest.

In the United States, the Assistant Secretary for Preparedness and Response (ASPR), Department of Health and Human Services, joined by the directors of the Centers for Disease Control and Prevention (CDC) and the National Institutes of Health, recently published a set of key components for a research response to public health emergencies [35]. After listing the research failures during the influenza A(H1N1) pandemic in 2009, the authors called for several actions to be taken before the next emergency event. These actions include (1) identifying potential knowledge gaps and research questions; (2) developing and preapproving generic study protocols; (3) obtaining approval for these protocols from institutional review boards; (4) using prefunded research networks and preawarded just-in-time research contracts; and (5) developing an on-call "ready reserve" of clinicians, scientists, and other experts to undertake this research. The essential elements of ASPR's research response plan as they might apply to influenza pandemic preparedness were outlined in an article published in 2009 [36]. Unfortunately, none of ASPR's proposed actions has been implemented, and no plans have been made to study immunomodulatory agents (D.S. Fedson, unpublished observation).

The statins/influenza study mentioned earlier [30] was conducted by the CDC's Emerging Infections Program, but CDC's Influenza Division has not initiated studies to confirm or extend its findings (D.S. Fedson, unpublished observation). In September 2012, the Infectious Diseases Society of America (IDSA) published its US action plan for pandemic and seasonal influenza [10, 37]. The plan focuses on vaccines, antiviral agents, better diagnostics, improved surveillance, and more effective risk communication. The IDSA report briefly mentions

immunomodulatory treatment, but a careful reading indicates that research on these agents is not central to the IDSA's action plan. At the global level, the pandemic preparedness efforts of the World Health Organization (WHO) remain focused on vaccines and antiviral agents [38]. WHO has paid no attention to immunomodulatory treatment, and it was not discussed at the World Health Assembly meeting this past May [39].

George Orwell once wrote that "to see what is front of one's nose needs a constant struggle" [40]. Physicians inevitably will be called upon to care for patients in the next pandemic. They need to ask why influenza scientists and health officials who support their work have not undertaken pragmatically focused laboratory and clinical research to see if statins and other promising immunomodulatory agents could be used to reduce influenza-related mortality. There is no guarantee that any of these drugs will work, but physicians will never know unless those responsible for pandemic preparedness recognize and act on the extraordinary possibility that these agents might save lives.

Note

Potential conflicts of interest. The author has previously received honoraria and travel expenses from Sanofi Pasteur, Sanofi Pasteur MSD, and Merck, Inc, for speaking engagements on influenza and pneumococcal vaccination.

The author has submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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REVIEW

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The controversy over H5N1 transmissibility research

An opportunity to define a practical response to a global threat

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Keywords: influenza, transmissibility research, H5N1, immunomodulatory agents, statins

Since December 2011, influenza virologists and biosecurity experts have been engaged in a controversial debate over research on the transmissibility of H5N1 influenza viruses. Influenza virologists disagreed with the NSABB's recommendation not to publish experimental details of their findings, whereas biosecurity experts wanted the details to be withheld and future research restricted. The virologists initially declared a voluntary moratorium on their work, but later the NSABB allowed their articles to be published, and soon transmissibility research will resume. Throughout the debate, both sides have had understandable views, but both have overlooked the more important question of whether anything could be done if one of these experimentally derived viruses or a naturally occurring and highly virulent influenza virus should emerge and cause a global pandemic. This is a crucial question, because during the 2009 H1N1 influenza pandemic, more than 90% of the world's people had no access to timely supplies of affordable vaccines and antiviral agents. Observational studies suggest that inpatient statin treatment reduces mortality in patients with laboratory-confirmed seasonal influenza. Other immunomodulatory agents (glitazones, fibrates and AMPK agonists) improve survival in mice infected with influenza viruses. These agents are produced as inexpensive generics in developing countries. If they were shown to be effective, they could be used immediately to treat patients in any country with a basic health care system. For this reason alone, influenza virologists and biosecurity experts need to join with public health officials to develop an agenda for laboratory and clinical research on these agents. This is the only approach that could yield practical measures for a global response to the next influenza pandemic.

Introduction

In December 2011, the National Science Advisory Board for Biosecurity (NSABB) in the US recommended restricting publication of the experimental details of A/H5N1 influenza virus transmissibility research conducted by Ron Fouchier, Yoshi Kawaoka and their colleagues.[1,2] Fouchier had presented the results of his studies at a scientific meeting in September 2011 and his findings had received considerable attention among influenza virologists. However, following the announcement of the NSABB recommendation, there was widespread comment in major scientific journals and in the media, and the NSABB's decision quickly became controversial.[3]

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H5N1 Transmissibility Research and the NSABB

In response to the NSABB decision, Fouchier and Kawaoka reluctantly agreed to a voluntary moratorium on publishing their findings and continuing their research.[4] They and many other virologists were concerned that science was being censored.[1,2,5-9] In contrast, the NSABB[10,11] and others regarded as biosecurity experts[12-15] worried that a highly transmissible H5N1 virus could be released accidentally or deliberately among human populations. In February 2012, the World Health Organization (WHO) convened an international technical consultation that included the principal scientists involved in this controversy.[16] One month later, the NSABB received reassuring new data from Fouchier and Kawaoka. Moreover, intelligence officials had concluded that H5N1 transmissibility research did not present a biosecurity threat. Accordingly, the NSABB revised its earlier decision and unanimously recommended full publication of Kawaoka's findings,[17] which were subsequently published.[18] There was less than complete agreement on whether to publish Fouchier's findings, but after extensive revision his manuscript too was published.[19] The US Government also issued revised recommendations on its oversight of "dual use research of concern"; i.e., research that is considered scientifically useful but could also be used deliberately or accidentally to cause harm.[20]

Influenza virologists believe that publication of their findings will have several benefits. For example, Kawaoka has said, "The amino acid changes identified here will help individuals conducting surveillance in regions with circulating H5N1 viruses … to recognize key residues that predict the pandemic potential of

isolates. Rapid responses in a potential pandemic situation are essential in order to generate appropriate vaccines and initiate other public health measures to control infection. Furthermore, our findings are of critical importance to those making public health and policy decisions."[18] However, many influenza scientists doubt this research will yield any practical benefits for influenza virus surveillance or for developing vaccines and antiviral agents, at least in the foreseeable future.[21,22]

The ability of influenza viruses to mutate and yield new viruses that might be more virulent or more easily transmitted was earlier demonstrated in vivo for the 2009 pandemic A (H1N1) (pH1N1) virus in mice[23] and ferrets.[24-26] These reports appeared before the H5N1 studies of Fouchier and Kawaoka came to NSABB and public attention. A more recent study has reported the in vitro evolution of two mutant H5N1 viruses, one that was transmissible by direct contact and another that was partially transmissible by droplets in ferrets.[27] Fouchier and Kawaoka found that only 3 to 5 mutations were required to generate respiratory transmissible H5N1 viruses. Other investigators using mathematical models have concluded, "the remaining mutations could evolve within a single mammalian host, making the possibility of a respiratory droplet–transmissible A/H5N1 virus evolving in nature a potentially serious threat."[28]

The H5N1 transmissibility research controversy is slowly moving toward resolution. Eventually, new rules for this and other types of "dual use research of concern" will be formulated. In the meantime, it is worth asking whether this controversy has something else to teach us.[29]

Adequate Global Supplies of Vaccines and Antiviral Agents won't be Available for a Global Response to the Next Pandemic

The concerns expressed by influenza virologists and biosecurity experts about H5N1 transmissibility research are understandable. However, both groups have overlooked a far more important question: could an effective global response be mounted to confront a pandemic caused by a new highly transmissible and virulent influenza virus, regardless of whether it is a laboratory-generated H5N1 virus or (more likely) a naturally derived variant of the currently circulating H5N1 or seasonal influenza viruses? This question is critically important, for if a virus as virulent as the one that caused the pandemic in 1918 were to emerge today, it might kill 62 million people worldwide.[30]

The global response to the relatively mild H1N1 influenza pandemic in 2009 amply demonstrated that scientists, companies and public health officials working together lacked the capacity to rapidly develop,[31] produce[32] and distribute[33-35] affordable supplies of pandemic vaccines and antiviral agents in time to mitigate the pandemic's impact on more than 90% of the world's people. This is incontrovertible evidence that in the event of a new and more severe influenza pandemic, regardless of its provenance, it will be impossible to successfully implement an effective global public health response that targets only the virus.

Clinical and Epidemiologic Findings Suggest an Alternative Approach to a Pandemic

If vaccines and antiviral agents will be unavailable to most of the world's people when the next pandemic virus emerges, would it be possible to confront the pandemic using an alternative approach that targets the host response to the virus? A clue to the promise of this approach promise can be seen in the disparity in the case fatality rates of children and young adults in the 1918 influenza pandemic.[36] This pandemic caused exceptional mortality in young adults but not in children. Some scientists have ascribed the high mortality in young adults to secondary bacterial pneumonia,[37-39] but this explanation fails to account for the more frequent infection of children with the virus that killed young adults and the (almost certain) more frequent colonization of their nasopharyngeal passages with the same bacteria found in the lungs of young adults who died (**Fig. 1**).[36,40]

Influenza virologists recognize that children were not protected from infection, but "… for reasons that are as mysterious today as they were in 1918, they were able to cope with the disease much better than their adult counterparts."[41] Although these virologists have made extraordinary contributions to our understanding of the 1918, H5N1 and other influenza viruses, they have been unable to answer the question, "Why did young adults die." The more important question is "Why did children live?" The different case fatality rates in children and young adults in 1918 might have been due to characteristics specific to host responses of children and young adults that differentially affected their risks of dying.[36,40] Clinicians and epidemiologists have documented similar differences in the case fatality rates of children and adults in several other infectious and non-infectious conditions.[40] These differences might have arisen during the course of human evolution. Yet, influenza virologists, immunologists and evolutionary biologists appear to have given little attention to studying the mechanisms underlying these differences.

In older adults, mortality due to seasonal and pandemic influenza largely affects those with underlying high-risk conditions: cardiopulmonary diseases, diabetes and renal disease. In younger adults those with obesity, asthma and pregnancy are affected. In both young and old, these conditions share one feature in common: each is characterized by alterations in innate immunity that in many instances constitute a form of low-grade inflammation known to cardiovascular scientists as "metabolic syndrome."[42-46] Among children who die of influenza, most have known immune disorders. In those with fatal influenza and no recognized disturbance in immune function, it is possible that unrecognized antecedent events have induced cytokine dysregulation and increased their vulnerability to influenza-related complications and death. In all likelihood, all of these individuals are at increased risk because their "innate immune rheostats" have been set at different and more precarious levels, making them more vulnerable to a loss of innate immune homeostasis.[47]

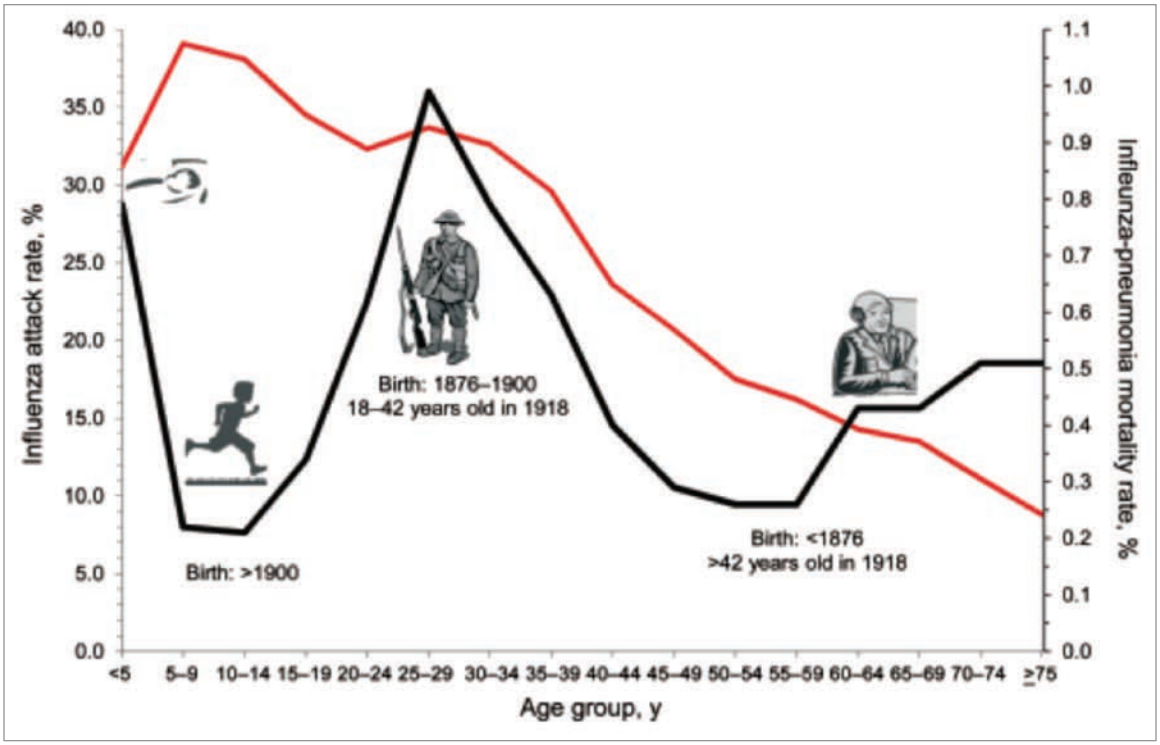

Figure 1. Discrepancy between clinical influenza attack rates and influenza pneumonia mortality rates in the 1918 influenza pandemic (adapted from ref. 38).

The Host Response to Influenza

Human influenza is associated with elevated levels of pro- and anti-inflammatory cytokines and chemokines, and the greater the degree of dysregulation, the greater the likelihood of severe or fatal illness.[48] Even in patients with mild illness, elevated cytokine levels distinguish between those who develop symptoms and those who have asymptomatic infection.[49] Few people with fatal influenza die during the first few days of illness when a pro-inflammatory response dominates. Instead, like patients with sepsis,[50] most die during the second week or later when an anti-inflammatory response and immunosuppression become dominant and virus replication has decreased.[36,40] These changes in the host response have been demonstrated in studies of H5N1 and non-H5N1 influenza viruses in mice,[51] ferrets[52] and non-human primates,[53] and interactions between virus and host factors that determine the course of illness have been discussed extensively by influenza virologists.[54-57]

Many influenza virologists are convinced that virus factors - infecting dose, extent of replication and degree of virulence - principally determine the outcome in influenza, hence their emphasis on controlling the disease with vaccines and antiviral agents.[57-59] No one would argue seriously that these factors are unimportant. Nonetheless, they cannot explain why an inactivated H5N1 virus can cause fatal acute lung injury in mice,[60] nor why survival in the acute lung injury seen in sepsis, pneumonia and influenza is determined by active resolution of inflammation,[61,62] the restoration of pulmonary endothelial barrier integrity,[63] mitochondrial biogenesis[64-66] and changes in energy metabolism.[67,68] Most of all, it is difficult to imagine how factors intrinsic to the virus could have been solely responsible for the different mortality rates seen in children and adults in the 1918 pandemic.[36,40]

A dysregulated host response appears to be the principal factor responsible for fatal influenza. Since timely and affordable supplies of vaccines and antiviral agents won't be available when the next pandemic virus emerges, the challenge to laboratory and clinical investigators is to identify existing agents that can reestablish the host's capacity for self-regulated homeostasis. An abundance of clinical and laboratory research indicates this can be done.

Targeting the Host Response to Pneumonia and Influenza with Immunomodulatory Agents

A growing body of evidence suggests it should be possible to modify the dysregulated host response of patients with community-acquired pneumonia and influenza and improve their survival.[36] For many years, physicians have used 3-hydroxymethyl-3-glutaryl coenzyme A (HMG-CoA) reductase inhibitors (statins), peroxisome proliferator activator receptor (PPAR)α and PPARγ agonists (fibrates and glitazones, respectively) and AMP kinase agonists (metformin) to treat the dysregulated host responses of patients with chronic heart diseases and diabetes mellitus. The clinical benefits and safety of these immunomodulatory agents are widely known. In addition to their effectiveness when given as long-term treatment, they have beneficial effects when given acutely; for example, when statins are given to patients within 24 h following hospitalization for acute myocardial infarction,

they significantly reduce hospital mortality.[69] These agents have also been shown to have overlapping anti-inflammatory and immunomodulatory (pleiotropic) activities in mouse models of systemic inflammation, both sterile [e.g., after endotoxin (LPS) treatment] and infection-induced [e.g., cecal ligation and puncture (CLP)] sepsis.[36]

Observational studies in humans have evaluated the effects of statins in patients with pneumonia (there are no studies of fibrates, glitazones or metformin). Most but not all of these studies have shown that outpatients taking statins (almost certainly for cardiovascular reasons) have reduced rates of pneumonia hospitalization and death.[70-75] Three observational studies have documented the effects of inpatient statin treatment on pneumonia mortality. In one study of 1985 patients, continued statin use in the hospital reduced hospital mortality by 27% [adjusted odds ratio (OR) 0.73; 95% confidence interval (CI) 0.47–1.13; p = 0.15].[76] In a second study of 121,254 inpatients, statin treatment reduced hospital mortality in those not admitted to intensive care by 21% (adjusted OR 0.79; 95% CI 0.71–0.87), but it had no effect on mortality in those who required intensive care (adjusted OR 0.93; 95% CI 0.81–1.06).[77] The third study reported the results of a propensity matched case-control study that used a Department of Veterans Affairs administrative database of patients ≥ 65 y of age hospitalized with pneumonia (11,498 cases and 11,498 controls).[78] Inpatient statin treatment was associated with a 32% reduction in 30-d mortality (adjusted OR 0.68; 95% CI 0.59–0.78). In addition, outpatient statins were associated with a 26% reduction in 30-d mortality (adjusted OR 0.74; 95% CI 0.68–0.82). Outpatient and inpatient use of angiotensin-converting enzyme (ACE) inhibitors and angiotensin II receptor blockers (ARBs) were also associated with significant reductions in 30-d mortality, but there was no analysis of combination treatment with a statin and either an ACE inhibitor or an ARB.[78]

No reports have been published of randomized controlled trials of statin treatment of patients with pneumonia. However, a single center clinical trial conducted in 100 patients hospitalized with sepsis has shown that atorvastatin (40 mg/day) significantly reduced progression to severe sepsis (4% in treated patients vs. 24% in controls; p = 0.007).[79]

Immunomodulatory Treatment of Pandemic Influenza

In 2004, it was suggested that statins might be useful in reducing mortality from pandemic influenza.[80] This idea was based on the well-established phenotypic benefits of acute statin treatment in patients with acute myocardial infarction, and the possibility that similar benefits might be seen in patients with severe influenza. Over the next few years, several influenza virologists failed to show that statins could reduce influenza mortality in mice, although none of their studies has been published (DS Fedson, unpublished observations).

Two recent studies failed to show that statins reduce mortality in mouse models of influenza. In one report, rosuvastatin (administered in the diet) failed to protect C57Bl/6 mice infected with H3N2 and WSN influenza viruses, but the infecting doses of virus were very high (LD_{100}) and there was clear evidence that after one or two days the mice stopped eating, and therefore were no longer being treated.[81] In a much larger study, several different statins were tested against several different influenza viruses in BALB/c mice.[82] No meaningful evidence of protection was shown, but again the infecting dose of virus was highly lethal. Moreover, treatment was given for only a few days, and it is well known that early cessation of statin treatment during an inflammatory illness in both mice and humans leads to a rebound hypercytokinemia and increased mortality.[83]

A limited number of laboratory studies have shown the effectiveness of other immunomodulatory agents in mouse models of influenza. Post-infection treatment with resveratrol (a plant polyphenol with immunomodulatory activities)[84] and gemfibrozil[85] significantly improved survival in influenza virus-infected mice, and similar improvements have been demonstrated for pre-infection treatment with pioglitazone[86] and pioglitazone combined with AICAR, a metformin-like drug.[87] In two studies that evaluated the effects of treatment on virus replication, pulmonary virus levels were either unchanged[86] or reduced.[84] A more recent study has shown that treatment of mice with the PPARγ agonist 15-deoxy-$\Delta^{12,14}$-prostaglandin J_2 (15d-PGJ_2), starting one day after infection, improved survival from 14% to 79% and markedly reduced.[88] Surprisingly, 15d-PGJ_2 treatment started on day 0 was not protective. Moreover, although protection by 15d-PGJ_2 could be reversed by a specific PPARγ antagonist, treatment with rosiglitazone (a clinical PPARγ agonist that also has non PPARγ activities) on day 0 or day 1 was not protective. In another study, a highly active glutathione derivative (glutathione is an important intracellular antioxidant) strongly inhibited PR8 influenza virus replication in vitro by blocking cytoplasmic maturation of the virus hemagglutinin, and treatment of influenza virus-infected mice reduced mortality 4-fold.[89] Statins, glitazones, fibrates and metformin all upregulate glutathione activity.[90] It is important to note that none of these experimental studies included co-treatment with a recognized antiviral agent.

Reports on the effects of immunomodulatory agents in human influenza are limited to statins. Two reports have appeared on the effects of statins on laboratory-confirmed human influenza. In an observational study of 1520 patients hospitalized in 2009 with pH1N1, preadmission statins were associated with a statistically nonsignificant 28% reduction in hospital mortality (adjusted OR 0.72; 95% CI 0.38–1.33).[91] Unfortunately, the investigators gathered no data on inpatient statin use. More important, an observational study has reported on statin treatment of 3043 older adults hospitalized in 2007–2008 with laboratory-confirmed seasonal influenza.[92] Statins were begun as outpatient treatment in 96% of patients and were either continued or started after hospital admission in 87%. Statin use was associated with a statistically significant 41% reduction in mortality within 30 d of a positive test for influenza virus (adjusted OR 0.59; 95% CI 0.29–0.92; deaths occurred either in the hospital or shortly after discharge). The results of this pivotal study provide compelling evidence to support the concept that immunomodulatory treatment of influenza should work.

Table 1. Cell signaling pathways that might be targeted by immunomodulatory treatment*

• Upregulate HO-1† and decrease TLR signaling by PAMPs and DAMPs
• Downregulate NF-kappaB and pro-inflammatory cytokines (e.g., TNFα, IL-1, IL-6)
• Upregulate anti-inflammatory cytokines (IL-10, TGFβ)
• Upregulate pro-resolution factors (lipoxin A4, resolvin E1)
• Downregulate HMGB1/RAGE and late mediators of inflammation
• Upregulate adipokines (adiponectin) that decrease inflammation
• Upregulate eNOS, downregulate iNOS, restore iNOS/eNOS balance and stabilize cardiovascular function
• Decrease formation of reactive oxygen species and reduce oxidative stress
• Decrease tissue factor and its associated pro-thrombotic state
• Attenuate the C5a-C5aR-related increase in vascular endothelial permeability
• Stabilize the actin cytoskeleton and adherens and tight junctions in endothelial cells, increase pulmonary barrier integrity and decrease vascular leak
• Attenuate acute disease-associated pulmonary hypertension
• Restore the balance between Th17 and Treg cells
• Differentially modify caspase activation and apoptosis in epithelial and endothelial cells, macrophages, neutrophils and lymphocytes in the lung and other organs
• Upregulate AMPK and PGC-1α, improve mitochondrial function and restore mitochondrial biogenesis and metabolic homeostasis

*Adapted from references 36 and 96 and DS Fedson, unpublished observations. †HO-1, heme oxygenase -1; TLR, Toll-like receptor; PAMP, pathogen-associated molecular pattern; DAMP, damage associated molecular pattern; NF-kappaB, nuclear factor kappaB; TNFα, tumor necrosis factor α; IL-1, Interleukin-1; TGFβ, transforming growth factor β; HMGB1, high molecular group box-1; RAGE, receptor for advanced glycation end products; eNOS, endothelial nitric oxide synthase; iNOS, inducible nitric oxide synthase; C5aR, C5a receptor; Treg, T regulatory; AMPK, adenosine monophosphate-activated protein kinase; PGC-1α, peroxisome-proliferator-activated receptor (PPAR)γ coactivator-1α.

Questions about the Effectiveness of Statins in Treating Influenza

The results of this pivotal study have been questioned because it is thought that patients who received statins were "healthy users."[93] The same reason has been used to claim that observational studies showing the effectiveness of influenza vaccination in reducing hospitalizations and deaths are similarly biased; in other words, vaccination appears to be effective (but is not) because relatively healthy older adults take better care of their health (and get more vaccines) than those who are less healthy, and thus they are more likely not to be hospitalized or die because they are healthier, not because they have been vaccinated.[94] The statins investigators responded to this criticism by listing the steps they took in their analysis to control for healthy user bias.[95] The critics failed to mention that the healthy user bias had already been accounted for by the investigators in their adjusted analysis: the 41% reduction in mortality with statin treatment was in addition to any reduction that might have been attributable to previous influenza vaccination and antiviral treatment.[92]

The results of most observational studies demonstrate the phenotypic effects of statin treatment in reducing pneumonia and influenza mortality. To date, no such studies have been reported on the effects of glitazones, fibrates or metformin, although observational studies of large groups of diabetic patients would be informative. Nonetheless, the known immunomodulatory effects of these agents in other conditions characterized by cytokine dysregulation (e.g., cardiovascular disease, metabolic syndrome, diabetes) as well as their effects in several experimental models of infection and inflammation have provided insights into some of their potential mechanisms of action (**Table 1**; refs. 36, 96, 97 and DS Fedson, unpublished data). Other immunomodulatory agents have been suggested as candidates for influenza treatment.[98] ACE inhibitors and ARBs are among the most promising agents,[78] but there are no studies of their use in experimental models of influenza. Among other agents that are licensed, (e.g., macrolides, cyclooxygenase-2 inhibitors), few data support their use. For other candidate agents (e.g., anti-TNF therapy, mesenchymal stem cells, angiopoeitin-1, high mobility group box-1 antagonists), limited supplies, high costs and/or their investigational status mean that many years will pass before any of them can be considered seriously for clinical trials in influenza patients.

We already have an indication that immunomodulatory treatment might reduce the higher influenza mortality rates of younger adults. In an experiment published in 2008, "children" and "young adult" mice were subjected to ischemia reperfusion injury of the liver.[99] (In "young adult" mice more so than in "children," this condition is highly inflammatory and often fatal). In this study, pre-treatment with rosiglitazone was able to "roll back" the harmful inflammatory response of young adults to the more benign response of children. This important experiment could have implications for patient care in an influenza pandemic. In a study comparing the effects of pH1N1 virus infection in newly weaned and adult ferrets, the immunological and pathological findings in newly weaned ferrets were less severe and the clinical illness was much milder.[100]

The four groups of the immunomodulatory agents mentioned above are now produced as inexpensive generics in developing countries. If these agents could be shown convincingly to reduce mortality in patients with severe influenza, they would be

available to treat patients in any country with a basic health care system on the first pandemic day. For each patient, the cost of this "bottom up" approach would be less than one dollar.[36]

Corticosteroid Treatment of Influenza: A Cautionary Note

Physicians often use corticosteroids to treat patients with sepsis, severe acute lung injury and acute respiratory distress syndrome in the hope that the anti-inflammatory effects of these agents will improve survival. Unfortunately, the evidence supporting their use is weak.[101,102] This includes observational studies in 6650 patients and ten randomized controlled trials involving 1090 patients hospitalized with pneumonia due to pandemic H1N1 virus infection.[102] Some of these studies have even shown that corticosteroids were harmful,[103,104] leading to a spirited discussion of the pros and cons of steroid treatment for viral pneumonia.[105,106]

A full discussion of corticosteroid treatment lies outside the bounds of this review. Nonetheless, it is worth noting the considerable overlap in their cell-signaling pathways and those for the immunomodulatory agents under discussion here (**Table 1** and ref. 106). There is also considerable molecular crosstalk between PPAR agonists and the glucocorticoid receptor.[107,108] Thus, despite encouraging results from the observational studies reviewed above, these similarities argue for caution regarding benefits that might be anticipated from treating influenza patients with statins and these other agents. That being said, fibrates and statins enhance the signaling effects of corticosteroids,[108,109] so combination treatment that includes a corticosteroid might be more beneficial than single agent treatment. In addition, a direct comparison of dexamethasone and pioglitazone treatment of smoke-exposed mice infected with H1N1 influenza A virus showed greater efficacy for pioglitazone.[110]

A Research Agenda for Immunomodulatory Treatment of Influenza Patients

Several years ago, a five-point research agenda was proposed for identifying one or more immunomodulatory agents that might be used to manage patients with pandemic influenza (**Table 2** and ref. 36). If immunomodulatory agents could be shown to be effective, they would be used primarily to treat pandemic patients with severe, life-threatening illness, although for special groups (e.g., health care workers or very high-risk patients) they might also be used for prophylaxis, especially when vaccines and antiviral agents are unavailable.

Since this agenda was first presented, there has been progress on several fronts. We now have good international information on the companies that produce statins, glitazones, fibrates and metformin. We also have information on quantities produced each year, distribution channels and wholesale prices for branded and generic products. For example, a few years ago it was estimated that in 2012, 48 billion doses of statins would be distributed throughout the world (DS Fedson, unpublished observation). Of these doses, 77% would be produced as generics, and the average price per generic dose would be $0.17. Almost 20 billion doses would be distributed in countries outside the United States, Canada and Western and Central Europe. If it were assumed that in a pandemic, 5% (350 million) of the world's 7.0 billion people would need to be treated for ten days (a deliberately exaggerated assumption), 3.5 billion doses would be required. This would account for approximately 7% of the annual consumption of statins worldwide. Information on statins and the other immunomodulatory agents mentioned above needs to be updated. Nonetheless, it is already evident that these drugs are currently available as generics wherever there are physicians who treat patients with cardiovascular diseases and diabetes. In most countries, expensive programs for stockpiling them would not be needed.

Soon after the H1N1 pandemic virus emerged in 2009, several groups of intensive care specialists tried unsuccessfully to initiate randomized controlled trials of statins in pH1N1-infected, ICU-admitted patients.[111,112] The focus on statins was based largely on encouraging findings from observational studies of statins use in patients with sepsis and pneumonia (no such information was available for the other agents). Nonetheless, there is broad agreement that randomized controlled trials will be needed to determine whether immunomodulatory treatments are efficacious. In anticipation of the next pandemic, clinical trials should be organized beforehand so they can be started immediately after the emergence of a new pandemic virus. In the meantime, similar trials conducted in patients with seasonal influenza should be undertaken. Investigators will have to decide whether the trials should be restricted to ICU-admitted patients, who might not benefit,[76,77,113] or include all hospitalized patients at risk of rapidly developing more serious illness.[79] Regardless of their design, the trials will be expensive, so animal studies comparing different immunomodulatory agents will be needed to guide the choice of which agent(s) to evaluate in clinical trials.

Animal Studies of Immunomodulatory Treatment of Influenza

Investigators will need to proceed with caution because the results of laboratory studies might be difficult to interpret.[81,82] For example, studies by several virologists have yet to show that statins are effective in mouse models of influenza, yet many human studies suggest that they are (see above). There is no ready explanation for these discordant results, but it is worth noting that although the molecular mechanisms for the inflammatory responses of humans and mice are in many ways similar, they are quantitatively very different. For example, a comparison of the response of human and mouse macrophages to LPS-induced inflammation showed that the human response was 10,000 times more sensitive to LPS than that of mice.[114]

In mouse models of immunomodulatory treatment, choosing a test virus that more clearly mimics human influenza virus infection could be important (**Table 2**). For example, the mouse-adapted PR8 virus is highly lethal for mice, but markedly less so for man, so a pH1N1 virus might be a better choice. Likewise, choosing an appropriate infecting dose is also

Table 2. Research to identify immunomodulatory agents that might be used to treat pandemic influenza patients*

• Test candidate agents in mice, ferrets and non-human primates to identify agents that might be used to manage patients
• Later study these agents in cell culture and animals to identify molecular mechanisms that explain their beneficial effects
• Document where these agents are produced as generics and determine quantities produced, surge capacities, patterns of distribution and costs to public programs
• Establish a process for managing their global stockpiling before a pandemic or distribution once a pandemic begins
• Plan randomized controlled trials of promising agents to begin immediately upon the emergence of a new pandemic virus

*Adapted from reference 36.

probably important; an illness caused by a dose that is 100% lethal in mice will probably not reflect the spectrum of human influenza because not all patients with severe illness die. The choice of mouse strain might also be critical. Influenza virologists usually use either inbred BALB/c or C57Bl/6 mice,[115] and these two strains have been used in all experimental studies of immunomodulatory agents.[84-89] These strains might not be optimal for determining which agent might best counteract the more intense inflammatory response in man. For example, in a study of host factors involved in the pathogenesis of pH1N1 virus influenza, BALB/c mice, which have a Th-2 bias, were shown to be less suitable than C57Bl/6 mice, which have a Th-1 bias.[116] Neither strain might be as suitable as DBA/2J mice, which have a more intense inflammatory response to influenza virus infection.[117-119] Investigators should also consider testing immunomodulatory agents in mice that have the same high-risk conditions as humans; e.g., pregnancy,[62] obesity[120] and cardiovascular disease.[121] Once the most promising immunomodulatory agent (or combination of agents) has been identified, it should then be studied in ferrets and, if necessary, in non-human primates. In all of these studies it will be important to compare responses in "children" and "adults."

The Broader Implications of Immunomodulatory Treatment for Global Health

Despite compelling arguments for undertaking the laboratory and clinical research needed to show definitively whether immunomodulatory agents would improve survival in severe influenza, virologists and public health officials, including those at the World Health Organization, remain focused on targeting the virus. Yet success with treating the host response to influenza might be extended to the management of several other diseases in which cytokine dysregulation and the loss of homeostatic defense mechanisms leads to poor outcomes; for example, pneumococcal pneumonia,[122] severe malaria,[123] dengue hemorrhagic fever[124] and critical illness associated with trauma[125,126] and burn injury.[127,128]

Almost a half-century ago, physicians and public health officials learned that syndromic treatment of the host response to severe acute diarrheal illness could be accomplished with an inexpensive and universally available oral rehydration solution (ORS).[129] Although vaccines that target a few of the pathogens responsible for diarrheal disease have been developed since then (e.g., cholera and rotavirus vaccines), it is syndromic treatment with ORS that has saved millions of lives. Had decisions been made long ago to ignore the possibility of simple and inexpensive treatment and instead focus only on developing vaccines, these millions would have died. Scientists and health officials responsible for developing a practical response to a global influenza pandemic should learn from this history.

Conclusion

The dysregulated host response seen in severe influenza (and many other conditions) might be treatable with safe, inexpensive generic immunomodulatory agents. Whether these agents will actually be effective in routine clinical care needs to be demonstrated in further laboratory and clinical research. Nonetheless, it should be clear to everyone that such treatment would be of immense practical importance to global public health. Until now, influenza virologists have been reluctant to undertake experiments to identify potentially useful and widely available agents that investigators could test in clinical trials and physicians could use to manage their patients. Until they do, public health officials will have no alternative but to recommend that most of the world's people confront the next global influenza pandemic with little more than hand washing and social distancing. These "technologies" represent the best of 19th Century public health practice. In the 21st Century, we can and should do much better.[36,130]

The debate about H5N1 transmissibility research should be about more than how to define its boundaries, important though this may be. The controversy presents influenza virologists, bio-security experts and public health officials with a new opportunity to jointly define a research agenda to identify existing immunomodulatory agents that could be used in a practical response to a global influenza pandemic. This opportunity must not be wasted.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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From: Stanley Plotkin [mailto:stanley.plotkin@vaxconsult.com]
Sent: Monday, December 21, 2015 12:26 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: gain of function

Dear Committee:

I have perused the document produced by Gryphon Scientific and chose to comment on the section titled "Benefits." My comments are contained in the attachment.

Yours truly,
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ALLEGED GoF BENEFITS

Excerpts from RBA Report	Plotkin comments
GoF approaches that alter host range and enhance virulence uniquely enable the development of animal model systems that recapitulate human disease pathogenesis	True for enhancement of animal virulence, but issue is increasing human virulence, which is not the same, and infectiousness is just as important, as shown by high virulence but low spread of avian strains.
GoF approaches that enhance virulence are also uniquely capable of showing that live attenuated vaccines (LAVs) do not recover virulence upon growth *in vivo*	LAVs are not made that way, they are made with RNA segments of attenuated virus and RNA segments of current virus that give immunogenicity. There is no example of LAV becoming more virulent in vivo
This particular type of experiment simply increases the human health risk of the attenuated strain to approach that of wild type strains	Not true if HA made hypervirulent.
GoF that lead to evasion of therapeutics are critical for the development and regulatory approval of new therapeutics	Nonsense. Resistance to neuraminidase inhibiitors has not heeded approval.
Of note, adaptation to a new host typically attenuates virulence in the original host (in the case of SARS and MERS-CoV, humans)	Don't understand this. Adaptation to humans of SARS resulted in more virulence for humans. MERS is more virulent for humans than camels.
GoF can enhance virus production	No relationship to enhancement of virulence
GoF approaches that enhance the infectivity, transmissibility and virulence of influenza viruses inform pandemic risk assessments of circulating influenza viruses	So far this is unproven.
These risk assessments facilitate more rapid initiation of response activities such as pre-pandemic vaccine	Only true if there is natural increase of virulence. In any case, avian flu has high mortality but has yet to become epidemic
GoF approaches also guide selection of viruses used as the basis of pre-pandemic vaccines	No truth to this. Antigenic match is more important than virulence match
GoF approaches that lead to evasion of vaccines are uniquely capable of determining whether viruses can acquire mutations to escape neutralization of candidate broad-spectrum or universal influenza vaccines, a critical aspect of testing the potential field efficacy of vaccines in development	This is tautology. This is the unproven argument for GoF. We do not know if causing evasion in the lab predicts what will happen in nature.
No increase in human health risk is posed by strains that can overcome the protection afforded by universal vaccines because the latter are not available.	Don't understand logic. If a strain evades future vaccines it is perforce a threat to health if it escapes.
GoF approaches that lead to evasion of existing natural or induced immunity have potential to improve the efficacy of seasonal influenza vaccines	I suppose there is that potential, but no proof as yet and danger of escape.

From: Inglesby, Thomas [mailto:tinglesby@upmc.edu]
Sent: Tuesday, December 22, 2015 10:29 AM
To: Viggiani, Christopher (NIH/OD) [E]
Subject: RE: agenda and guidance

Chris – Thanks for sending the attachments.

Attached is the paper I referred to in the call. It would be great if you could circulate it with the members of the NSABB.

Much appreciated,
Tom

Thomas V. Inglesby, M.D.
Published online: December 18, 2015

December 22, 2015

EMBO reports

Science & Society

How likely is it that biological agents will be used deliberately to cause widespread harm?

Policymakers and scientists need to take seriously the possibility that potential pandemic pathogens will be misused

Thomas V Inglesby[1,2] & David A Relman[3]

During the past few years, there has been substantial debate concerning the risks and benefits of certain experiments with pathogens—initially motivated by two publications in 2012 that described laboratory efforts to enhance the mammalian transmissibility of the avian H5N1 influenza virus. One of these two reports was particularly noteworthy because the experiments were designed to yield new viruses with a set of properties that together might confer pandemic potential, such as high transmissibility, high pathogenicity, and resistance to commonly available countermeasures. Not all research on pathogens generates such concerns; in fact, it is only a rare experiment that might lead to the creation of a novel pathogen with pandemic potential (PPP). The term "gain-of-function" has also been used to describe this realm of research, but it refers to a much broader range of widely accepted non-controversial research techniques and goals. For that reason, we think it should not be used in this discussion and refer to this work with the more precise term of PPP.

> *"To say that no one would now or ever use PPP for deliberate misuse is grossly irresponsible guesswork"*

Proponents of such research argue that it is necessary to understand the evolution of pathogens and mechanisms of pathogenesis and transmission and that this knowledge can help public health authorities, vaccine manufacturers, and governments prepare for potential epidemics. Those concerned about PPP argue that this work is not critical for vaccine development or disease surveillance and that the accidental release of PPP—owing to insufficient biosafety or biosecurity or to laboratory accidents—could cause major outbreaks or even a pandemic [1,2]. Much less has been said or written, however, about the danger that such pathogens or their genome sequences could be deliberately misused to cause harm.

While the reporting of accidents and the collection and sharing of this safety information could (and should) be improved, it is possible to calculate a baseline probability of accidental releases from laboratories that perform PPP-related research with data based on existing records and statistics about biosafety and laboratory accidents in the USA and elsewhere [1]. Such calculations for example suggest at least a 0.2% chance of a laboratory-acquired infection per BSL3 laboratory year. A similarly quantitative risk assessment of the intentional misuse of PPP, however, is not possible. Such a calculation would require reliable, quantitative data on a variety of probability assessments: the probability that a person, group, or country intends to release PPP; that a person, group, or country has the means to obtain the pathogen or has the capacity to generate one from published data; and, that a person, group, or country has the means of distributing a PPP in a way that would start an epidemic. Those kinds of data are not presently available, nor will they be in the foreseeable future. However, other kinds of assessments could and should be made, including the human and political motivations that might lead to the misuse of PPPs, the weaknesses of security systems, the global distribution and quality of research capacity, and the availability of published research information. All of these could provide insight related to the risk posed by the deliberate misuse of PPP.

> *"The fact that a technology has not been misused is an unreliable predictor of its potential future misuse..."*

How can we therefore assess the risk that individuals, groups, or countries will start a pandemic with a PPP either now or in the future? Some involved in this debate have argued that since there have been no known attempts to use pathogens to start pandemics in recent times, there is little risk of it occurring in the future. Throughout history, however, there are examples of periods in which the potential of a new technology to be used for harm was not seen, or was denied up until the moment it was used as a weapon. Such moments often occur during periods of political, economic, or social upheaval, especially as the technology proliferates and disseminates. Tanks,

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DOI 10.15252/embr.201541674

for example, were initially seen as having very specific limited uses in battle, until the purpose and technologies related to tank warfare changed substantially in light of the trench warfare in World War I. Chlorine and its derivatives were first used to bleach textiles and anesthetize patients—until combatants introduced the large-scale use of chemical weapons on the battlefield during World War I, beginning with the deadly use of chlorine gas. Commercial airplanes were a boon for international travel and commerce, and hijackings were uncommon—until the late 1960s when scores of hijackings occurred. Modern terrorists, it was broadly stated, only wanted to frighten people, not kill large numbers of them—until terrorists hijacked airplanes and flew them into buildings, or blew them up in mid-air. Today, some extremist groups seek to kill as many of their enemy as possible—the attacks of 9/11 and many since have shown that clearly. The fact that a technology has not been misused is an unreliable predictor of its potential future misuse. Similarly, past actions of a particular terrorist group do not dictate what it will do in the future.

> *"If a terrorist group or country were to place a high enough value on obtaining a PPP strain, there would be a successful theft"*

Some might say that because experts cannot agree on the likelihood of a PPP being used in a terrorist attack, but that it might be small, the reasonable path to follow would be to assume that it will not happen. The counter-argument, which is the one we would support, is that the lack of agreement is a sign of the great complexity and uncertainty surrounding these issues. Given the potential consequences, we should err on the side of caution. There is a frequent presumption that other people, institutions, and countries will act as we do. This idea is called the Rational Actor Model for behavior, and it had serious potential consequences when it affected decisions made during the Second World War and the Cuban Missile Crisis [3]. We must avoid this pitfall when assessing the risks posed by the deliberate use of PPP.

There are other possibilities as to why people might deliberately use PPP to cause harm. Scientists could conceivably be co-opted to do things against their will because of extraordinary pressure or threat brought to bear. Alternatively, scientists could be convinced or seduced unwittingly to do things that aid and abet someone else whose ultimate purpose they did not appreciate or support.

If the potential consequences of PPP were not so serious, then speculation about the motivations of various actors around the world would be less important, as the penalty for being wrong would not be so great. But given the potential consequences of the misuse of PPP, it is critical to admit how much we do not and cannot know. The world is a huge, heterogeneous, complicated mix of cultures, motivations, drivers, and decisions. To say that no one would now or ever use PPP for deliberate misuse is grossly irresponsible guesswork.

Under what conditions might a person or group choose to start a pandemic with a PPP? The Islamic State and its affiliates use apocalyptic rhetoric and have seemingly few limits to their brutality, as the recent, horrific attacks in Paris demonstrated. The Islamic State has also sought to recruit scientists to help meet its ends. Paris is only the latest in a long list of recent examples of mass killings. Suicide bombers working for religious extremist groups have targeted places of worship, markets, and schools throughout the world. A lone, suicidal airplane pilot killed hundreds of people as collateral to his own suicide. As a thought exercise, would you give The Islamic State, or suicidal or homicidal people access to guns? Would you give them access to a virus that was both lethal and transmissible? You probably answered no to both because you think it at least conceivable that people in these situations could make terrible decisions that seem inconceivable to most of the world.

Are there any conditions under which a country might choose to start a pandemic with a PPP? It would seem improbable given that the consequences could devastate that country itself as the pandemic spreads. However, there are reasons why a country might consider it. Countries that wish to have an insurance policy against invasion might threaten use of a PPP in retaliation, as some do now with nuclear weapons. Countries could also use the prospect of PPP to compel other countries to act in certain ways, or to levy demands or extort concessions, as some countries that possess nuclear weapons now do. If a country were to develop a vaccine that was effective against a particular PPP and so could protect its own population, then it might have a lower threshold for using PPP for harm. Countries could even plan to use PPP in the case of defeat, as the former Soviet Union planned to do during the Cold War [4]. As a thought experiment, do you think it would be prudent to disseminate PPP laboratory strains to every nation in the world for their own national research programs? You probably do not; perhaps not even those nations you might be inclined to trust. Part of your reasoning might be that you have concerns about laboratory safety. But you also probably have other concerns and uncertainties about the possible fate of those PPP strains. And yet because of reverse genetics, publishing PPP genome sequences in the public domain is in some ways the same as distribution of the virus itself.

> *"If the number of laboratories doing this work grows, the opportunity to divert and obtain PPP strains will grow too in addition to the risks associated with potential laboratory accidents"*

Another consideration is that the line between countries and terrorist groups is not always distinct. It is clear that some terrorist groups are supported by nation-states and vice versa. And it is evident that some terrorist groups act as proxies for nation-states. In addition, leading scientists working within a country might not be under the control of national authorities, as was the case in the history of nuclear weapons proliferation (www.fas.org/sgp/crs/nuke/RL34248.pdf). What might seem implausible when considering the intentions of one specific entity might become plausible when considering the connections and relationships between individuals, terrorist groups, and countries.

Would a person, group, or country intending to start a pandemic be able to obtain a PPP? Some laboratories working on pandemic strains have

sophisticated security measures to prevent theft, which present a considerable challenge for anyone intent on stealing them. But if an entity with the means were committed to obtaining those strains, would those security plans be insurmountable? Could anyone working in those laboratories be convinced through some means—bribery, extortion, or disgruntlement, for example—to steal strains from the laboratory? Humans design and operate security systems, which means these systems have vulnerabilities. Items with high value—money, art, weapons designs, new technologies, financial information—are stolen all the time. If a terrorist group or country were to place a high enough value on obtaining a PPP strain, there would be a successful theft. In the past, when countries wanted access to new weapons or business technologies to give them an edge, they used insiders at appropriate locations to obtain that information. If PPP strains were seen as similarly valuable for their potential to do harm, then similar efforts would be likely. These efforts would be all the more successful if they targeted laboratories with lesser degrees of physical and operational security.

These risks will increase if PPP research continues and expands. For now, research on PPP strains is conducted in only a few laboratories. But given the attention and high impact publications that have followed this work, other laboratories will want to initiate similar research programs. Some will have capabilities in viral reverse genetics to re-create viral PPP starting with only the genomic sequence data. Currently, there are no global standards to say who will or will not be allowed to do this kind of work. Calls to try to limit PPP experiments have already been rejected as a misguided effort to control new technologies. If the number of laboratories doing this work grows, the opportunity to divert and obtain PPP strains will grow too in addition to the risks associated with potential laboratory accidents.

Clearly, the vast majority of life scientists are dedicated to the search for new knowledge that might benefit the planet, or for cures and vaccines, as examples. But this is not universally true. Some scientists may have a morbid curiosity to learn whether an alleged finding or a claim holds water. Some have infected others with pathogens from their own laboratories [5]. Some have cheated and misled their colleagues with false data [6]. Various countries have employed scientists to create weapons from pathogens, in some cases at large scales [7]. Scientists have joined terrorist groups, as was the case of Yazid Sufaat working for Al Qaeda (http://www.weeklystandard.com/al-qaedas-anthrax-scientist/ article/16989). Our planning to cope with the risks of PPP—as well as for other potential future challenges in the life sciences—needs to acknowledge this.

Even if interested parties were not able to obtain PPP directly, there is still a risk that a person, group, or country with the intention of starting a pandemic could create such agents based on publicly available information. One of the fundamental building blocks of scientific research is its reproducibility: if an experiment cannot be reproduced, the results will be called into question. Scientists publishing their work in peer-reviewed scientific journals are therefore required to describe their methods and experiments in sufficient detail so as to enable their colleagues to repeat it. Unless this requirement were changed in the special case of PPP research—and there are no indications that this will occur at this point—any existing and future publications on PPP will contain sufficient information for recreating novel strains of pathogens that are potentially lethal and transmissible in humans.

> *"Countries around the world have a right to know where this work is being done given the risks it poses to their populations"*

Only a small number of laboratories can perform such experimental work under appropriate biosafety conditions; but if safety were no longer a major concern, then the work could be carried out in a broader variety of laboratory settings. There are thousands of academic, government, and private science laboratories around the world. The 100 leading universities in the world for microbiology, based on their research record and reputations, are located in 20 different countries on five continents (http://www.usnews.com/education/best-global-universities/microbiology?page=10). Participants and winners in the International Genetically Engineered Machines competition (IGEM) come from all over the world. More than three-dozen BL4 laboratories existed or were being constructed as of 2011, located in 18 countries (http://fas.org/programs/bio/biosafetylevels.html). More than 1,300 registered BL3 laboratories existed in the USA alone as of 2007 (http://www.gao.gov/new.items/d08108t.pdf). There is not just an abundance of laboratories that have the necessary equipment and setup to conduct PPP research, but there is also no shortage of expertise and workforce. Employment in the US life sciences industry alone totaled 1.62 million in more than 73,000 companies in 2012 (http://www.nature.com/nbt/journal/v33/n1/full/nbt.3116.html). Another report noted that there were at least 500,000 life scientists in the EU (https://ec.europa.eu/research/infrastructures/pdf/enabling-science.pdf).

Some have commented that any unsanctioned work to create PPP will not go unnoticed and will eventually draw the attention of laboratory members or superiors or outsiders. But it is not always straightforward to know what kind of work is going on in a given laboratory, even from within the same institution. From a distance, it will be all the more challenging. The former Soviet Union had a massive bioweapons program for decades that was a complete mystery to the rest of the world.

Another critical factor here is that unlike the pathogens themselves, which may be limited to a single location or even destroyed at some point, their genome sequences and the information on how to genetically manipulate them will be publicly available from the moment it is published in perpetuity. We not only have to consider risks for the present, but possible risks for the future. It is important not only to acknowledge the limits of our own ability to make predictions, but also to acknowledge that we are often wrong about these predictions.

Given these considerations, the only reasonable and safe approach for continuing PPP research is to have two planning assumptions: There may be people, groups, and/or countries that are motivated to obtain PPP and either threaten, or in fact use them to start a pandemic, and there will be the means available to obtain PPP strains if they are created or to re-create them based on published information.

What should be done about this? Given these risks—and the risks involved in potential laboratory accidents discussed elsewhere—deliberate efforts to create new PPP should not be pursued unless a compelling case can be made that the benefits of a particular experiment outweigh the risks including the risks of deliberate misuse. We do not see that compelling argument for PPP, but it is possible that such a case could emerge.

If a decision is made nonetheless to proceed with PPP research, then a range of steps should be taken to reduce the risk of misuse. First, the risk of deliberate misuse should be taken more seriously. There has been little debate on the risks of deliberate misuse since the discussions about GOF, DURC, and PPP started a few years ago. This risk has often been dismissed with facile mischaracterizations such as "people in caves can't do this work". The discussion clearly requires a far more insightful analysis than that, and any entity funding or authorizing PPP work should have the best possible expert assessment on these issues before proceeding. An expert assessment—far beyond the considerations raised in this commentary—would have to include a determination of the level of scientific training that would be necessary to re-create these strains based on published information. This kind of assessment would necessarily include scientists who understand how this work was conducted in the original setting, as well as whether and how it could be conducted in a variety of other distinct settings.

A full assessment of the risk would also include a serious analysis of the conditions under which people, groups, and countries might consider the deliberate use of PPP. That kind of assessment would logically include social scientists, political scientists, and historians who have studied how technologies to do harm have evolved, dispersed, and been used. It would also draw on the talents of those who study the psychological elements of modern terrorism.

There are very few pathogens in the world with the potential to generate large-scale human-to-human transmissible epidemics. Laboratories working with those pathogens, particularly with PPP strains, should undergo exceptional external evaluations of safety and security. Countries around the world have a right to know where this work is being done given the risks it poses to their populations. They should not just learn about the work when it is published in a scientific journal. They should know about the work before it is started. International norms guiding research that could result in a pandemic do not now exist, but should be pursued (http://www.upmchealthsecurity.org/our-work/publications/synopsis-of-biological-safety-and-security-arrangements).

We are hopeful that the US government review process that is now underway—and others that follow it elsewhere in the world—will include a thorough assessment of the prospect of the deliberate misuse of PPP. We are also hopeful that US policymakers and policymakers around the world will start to take the possibility of deliberate misuse more seriously as part of an overall calculation of the risks and benefits of this narrow but highly consequential area of work. As such, we hope they will consider whether, given the risks, it is defensible to continue supporting such research.

Conflict of interest

The authors declare that they have no conflict of interest.

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From: D Gold
Sent: Wednesday, December 30, 2015 6:08 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Comments on GOF Risk Benefit Report

Dear Dr. Viggiani,

Attached are my comments on the Gryphon Scientific risk-benefit analysis. I am very concerned about the short time-frame provided for public comment. I believe this important issue deserves a thorough review, not only by the scientific community immediately involved in the issue, but by a lot of other interested people, such as myself, who do not have the resources to review a 1000 page document, plus additional material, in less than 30 days.

Thank you for your consideration.

Deborah Gold, MPH, CIH

Deborah Gold

December 30, 2015

Christopher Viggiani, Ph.D.
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Dear Dr. Viggiani and Members of the Board:

I am writing in regards to the recently published draft report by Gryphon Scientific, *Risk and Benefit Analysis of Gain of Function Research* (Report). My comments today are based on my 21 years of experience with Cal/OSHA, which began as an industrial hygienist in the Enforcement unit, and ended as Deputy Chief for Health, from which I retired in December 2014.

I think the less than 30 day period provided between the publication of the Report and the January 7-8 meeting is completely inadequate for a thorough review. For that reason I strongly suggest that you allow a public comment period of no less than 90 days, which would be more typical for such a significant project that has occupational as well as local, regional and world-wide public health implications. The Report has a number of significant gaps and unsubstantiated assertions, which will require time and research to address. The document doesn't address a number of risks, such as occupational risks to many categories of workers (which I will briefly explain below). It also does a poor job of explaining any true benefits to be achieved from this research. In this letter, I am addressing only the issues of biosafety as they apply to occupational exposures both immediate and distant from the laboratory. I will not try to restate the excellent discussion by the Cambridge Working Group (CWG), and encourage you to address the issues that they raise.

As a person who has been involved in public health as an advocate and as an occupational safety and health professional for decades, I am particularly appalled that in 2015, a government agency would consider basing a decision on a report that discounts the global risk from intentional development of drug resistant viruses with the following statement:

> *"The creation of an antiviral resistant strain could increase the consequences of a global outbreak, but only in more economically developed countries where caches of these antivirals could be handed out to a significant fraction of the infected population. A strain of seasonal influenza that can overcome protective vaccination could also increase the consequences of an outbreak in high income countries, which has the resources to vaccinate their population quickly."* (Executive Summary, page 2)

This is an extremely cynical statement, particularly in the light of the recent experience with Ebola Virus Disease (EVD), in which it became abundantly clear that countries with more resources MUST find ways to make care and treatment available for infectious diseases throughout the world, if for no other reason than their own self-interest. One would hope that should a pandemic influenza strain emerge in lower income countries, the US, in particular, would make sure that all relevant treatments were made available to reduce loss of life and improve outcomes in impacted countries.

Biosafety Risks

I believe that the Report fails to take seriously the biosafety hazards that currently exist in research laboratories. It states that the *"state of knowledge of the rates and consequences of human errors in life science laboratories is too poor to develop robust predictions of the absolute frequency with which laboratory accidents will lead to laboratory acquired infections."* This is an understatement regarding the lack of information, which is due both to lack of recognition of laboratory acquired infections (LAIs) as well as under-reporting. There is no public means of tracking other losses of containment, although there is apparently some tracking under the select agents program, which does not include all pathogens under consideration. A Report by the National Research Council (Review of Risk Assessment Work Plan for the Medical Countermeasures Test and Evaluation Facility at Fort Detrick: A Letter Report) cited unpublished 2010 CDC data, which found 395 reports of potential release events of select agents from 2003 to 2009.

The scientific and popular literature describe a plethora of laboratory incidents. For example, in 2012, an employee at the San Francisco Veterans Administration laboratory conducting research to develop a meningitis vaccine contracted meningitis and died. The joint investigations conducted by Cal/OSHA, OSHA, and the California Department of Public Health, found numerous problems in biosafety protocols, including unverified biosafety cabinets, during the investigation. In 2004, workers at the Children's Hospital Oakland Research Institute had to undergo chemoprophylaxis to prevent development of anthrax after it was determined that a shipment of purportedly deactivated *B. anthracis* had caused the death of some laboratory animals injected with the material. In 2014, CDC workers were exposed to live anthrax, and in 2015, the US Department of Defense was initially reported to have sent live (instead of deactivated) anthrax spores to labs in 9 states; this estimate was later revised to include labs in all 50 states and 9 countries. Mistaken shipments of pandemic or other virulent influenza strains have also been documented.

High containment laboratories, particularly BSL 3 laboratories, have proliferated in the past two decades, and on several occasions the US General Accounting Office has warned of the hazards associated with the lack of centralized regulation. Nancy Kingsburg, speaking on behalf of the GAO at a 2014 Congressional hearing following the anthrax exposures at the CDC explained some of their findings:

> *"The number of biosafety level (BSL)-3 and BSL-4 laboratories (high-containment laboratories) began to rise in the late 1990s, accelerating after the anthrax attacks throughout the United States. The laboratories expanded across federal, state,*

academic, and private sectors. Information about their number, location, activities, and ownership is available for high-containment laboratories registered with CDC's Division of Select Agent and Toxins (DSAT) or the U.S. Department of Agriculture's (USDA) Animal and Plant Health Inspection Service (APHIS) as part of the Federal Select Agent Program. These entities register laboratories that work with select agents that have specific potential human, animal, or plant health risks…

"According to most experts that we have spoken to in the course of our work, a baseline risk is associated with any high-containment laboratory. Although technology and improved scientific practice guidance have reduced the risk in high-containment laboratories, the risk is not zero (as illustrated by the recent incidents and others during the past decade). According to CDC officials, the risks from accidental exposure or release can never be completely eliminated and even laboratories within sophisticated biological research programs—including those most extensively regulated—has and will continue to have safety failures. Many experts agree that as the number of high-containment laboratories has increased, so the overall risk of an accidental or deliberate release of a dangerous pathogen will also increase. We recommended that CDC and APHIS work with the internal inspectors for Department of Defense and Department of Homeland Security to coordinate inspections and ensure the application of consistent inspection standards." (Testimony of Nancy Kingsbury, July 16, 2014, available at: http://gao.gov/assets/670/664799.pdf)

Occupational Risk

The Report appears to consider that any risk below that of a pandemic has been addressed through other biosafety guidance, such as the 2013 CDC Biosafety Recommendations for Work with Influenza Viruses Containing a Hemagglutinin from the A/goose/Guangdong/1/96 Lineage (MMWR June 28, 2013 / 62(RR06);1-7). However, the Report fails to consider how workers will be affected by enhanced (GOF) pathogens.

The immediate risk is to laboratory workers, who are the only workers addressed in the 2013 CDC Recommendations. If pathogens are successfully engineered to be more virulent, then exposed laboratory employees are at risk of more serious disease, including permanent sequelae or death. If those pathogens are engineered to be more resistant to anti-viral drugs, then employees who contract LAIs are also at greater risk of serious illness. Similarly, infections which might have been prevented through vaccination of employees will occur if employees have unprotected exposures.

California is unique among the states in adopting regulations to address biological risks to laboratory workers (beyond the requirements of the Bloodborne Pathogens standards). In 1994, the California Occupational Safety and Health Standards Board adopted a standard requiring employers to maintain biosafety cabinets in accordance with CDC recommendations, and adopted a laboratory biosafety section as part of the Aerosol Transmissible Diseases Standard in 2009. (This regulation can be found at: http://www.dir.ca.gov/Title8/5199.html.) During the relatively few inspections Cal/OSHA has conducted in chemical, biochemical, biomedical and microbiological laboratories the agency has found significant problems in maintenance of

containment equipment, training, personal protective equipment, ventilation, and other control measures.

Although laboratory employees are at the greatest risk of exposure and may be aware of their risk, they are only one category of employees who may be at risk. It is often the case that specific research projects in a lab, particularly research that may have defense implications, is unknown to other occupants of the building or outside of the specific lab. Although BSL 3 and BSL 4 labs are required to have secondary containment, the minimal level of acceptable negative pressure, and more importantly, the minimal maintenance provided in some facilities, may expose workers outside of the lab to the enhanced pathogens. Other routes of exposure include contact with waste or equipment that has been inadequately decontaminated, contact with co-workers who either have been inadequately decontaminated, are infectious but asymptomatic, or have symptoms that they and others attribute to seasonal influenza, particularly when the pathogen has been enhanced to be more transmissible between people. First responders, such as firefighters, police and paramedics may also be exposed to these pathogens in responding to incidents at these facilities. Those non-GOF workers may be unaware that they have been exposed to an enhanced pathogen, and therefore will not provide that information to medical providers, or even seek prompt medical attention, because they assume they have contracted a wild-type, self-limiting infection.

Nor does the occupational risk stop there. Unless a health care facility is specifically informed about the nature of the enhanced pathogen, health care workers would treat a symptomatic patient as they would any similar patient, unaware that they are being exposed to an enhanced pathogen that may not be susceptible to anti-viral drugs, etc. An influenza patient is not typically housed in airborne infection isolation, for example. Clinical laboratories conduct analyses for various pathogens and do not have BSL3 capacity. (This contributed to decisions to handle EVD samples at state or federal labs). If a pathogen such as SARS or MERS is not currently circulating in the US, absent a positive history such as travel to outbreak areas, it is unlikely that health care providers would suspect that infection. While a laboratory may instruct its employees to contact a specific health care provider if they become ill, when the employee is ill they may not be able to direct their medical care. It is unlikely that employees with secondary or inadvertent exposures as described above will be able to provide information to health care providers. We have seen with SARS in Asia and Canada, with MERS in Saudi Arabia and Korea, and with Ebola, that health care workers are at significantly increased risk from diseases borne by patients. All of these occupational risks would also apply if there were an intentional breach of the type identified in the biosecurity section.

Although these local infections may never rise to the level of an epidemic or pandemic, the risks to workers and their families and other contacts must be addressed in conducting this research. The risk to the community from laboratory exposures is illustrated by the nine cases of SARS in 2004 in Beijing resulting from exposure of two graduate students at China's National Institute of Virology Laboratory. In addition to the two graduate students who became ill, the mother of one student contracted the disease and died, and a nurse who treated the student also became ill. Five other SARS patients were linked to contact with the nurse. The 1978 Sverdlovsk anthrax leak, in which an estimated 100 people died due to the release of anthrax spores from a military facility, is another example of how laboratory incidents may impact the surrounding community.

I do not believe that this Report provides a basis for reinstituting NIH funding for GOF research on influenza or coronaviruses. Given the current state of control measures in “high containment” laboratories, the risks to employees and the community from GOF, such as enhanced virulence, transmissibility, drug resistance and evasion of immunity, are serious enough to warrant continuation of the moratorium. The benefits identified in the report are speculative, and in most cases can be achieved through other, less dangerous means. I refer you to comments by Dr. Raina MacIntyre and the CWG for more thorough discussion of the Report.

I hope that the NSABB decides to extend the period for public comment, as I look forward to providing additional comments on the full document and associated working papers. I also believe that the discussion must go beyond the interested scientific community to reach out to unions and other employee representatives, and members of the public. Thank you for your consideration.

Sincerely,

Deborah Gold, MPH, CIH

From: Lynn Klotz [mailto:lynnklotz@live.com]
Sent: Thursday, December 31, 2015 4:35 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Comments on the Gryphon risk-benefit assessment

Dear NSABB,

Attached are my comments on the Gryphon risk-benefit assessment in advance of the January 7 meeting.

Lynn Klotz
Senior Science Fellow
Center for Arms control and Non-proliferation

Attachment

A Commentary and Analysis of Chapter 6 in Gryphon Scientific's Report: Risk and Benefit Analysis of Gain of Function Research

A Commentary and Analysis of Chapter 6 in Gryphon Scientific's Report: Risk and Benefit Analysis of Gain of Function Research

By: Lynn C. Klotz, Ph.D.
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Date: December 30, 2015

With less than a month to analyze and comment on the thousand-page report before the December 31 "soft" deadline for the NABCC January meeting, it would be nearly impossible for anyone to follow in detail Gryphon's analysis and comment on all the chapters. I chose, therefore, to limit my comments and analysis to only Chapter 6, the Biosafety Risk Assessment.

Summary

Based largely on Gryphon's numbers, I estimated the likelihood-weighted fatalities for a pandemic seeded by a laboratory-acquired infection (LAI) from an mtHPAI (a mammal-adapted airborne-transmissible highly pathogenic avian influenza virus). Along the way, comments on aspects of Gryphon's Chapter-6 analysis will be made.

Generally, likelihood-weighted pandemic risk equals probability of a pandemic times consequences of the pandemic. The probability of a pandemic from a lab escape through an LAI for ten labs conducting research on mtHPAI strains for ten years was found to be 1.8×10^{-05} using Gryphon's numbers that an LAI lab escape leads to a pandemic. Ten labs for ten years is my estimate of the "research enterprise" that already is or will be conducting research with these strains.

In my analysis, consequences were restricted to fatalities. The case-fatality rate was chosen to be 5%, which is twelve-fold less than the World Health Organization's accepted case-fatality rate of 60%. For a pandemic infecting 25% of the world's population, the number of fatalities would be 90 million. With these numbers, the Likelihood-weighted fatalities for the research enterprise are

$$\text{likelihood-weighted fatalities} = (1.8 \times 10^{-05}) \times (90 \times 10^{6}) = 1{,}640$$

For a single lab for a single year, the likelihood-weighted fatalities are 10x10-fold less or 16.4, which I call "the fatality burden" for the lab. To put this fatality burden in perspective, no Institutional Review

Board tasked with assessing human subject research would approve a proposed research project with an expected 16.4 fatalities per year.

This 5% case fatality rate is much higher than the small fraction of 1% claimed by Morens and Taubenberger. But airborne-transmissible mtHPAI, a key focus of the NIH deliberative process, are not wild type viruses. They infect lung to lung via the airborne route. We do not know the case-fatality rate for these strains. It could be quite high, perhaps over 60%. Arguments over case fatality rate for wild-type HPAI are likely moot. Since we don't know, and the potential consequences in morbidity and mortality are so high, caution dictates instituting a ban on making and researching live airborne-transmissible mtHPAI. This will be discussed a bit more at the end of my Commentary.

The Gryphon report seems to dismiss gain-of-function studies in SARS and MERS, by assuming that mitigation measures such as quarantine should prevent a large outbreak. SARS has about an eight-day incubation period before an infected person can transmit infection, a fair amount of time to quarantine those exposed to an infected person. Timely and strong mitigation measures may be possible in developed nations, but we need only to look at the Ebola epidemic in the poor and war-torn African nations to understand the potential for large outbreaks. GOF studies in SARS and MERS should be looked at very carefully and perhaps many banned as well.

Details of and rationale for my analysis

In describing my analysis and the rationale for the numbers and estimates used, I will rely on quotes and data from the Gryphon risk assessment. Also, I will reproduce here relevant tables and graphs from the Gryphon RA as a convenience to you.

The three steps to a pandemic are illustrated in Gryphon's Figure 6.2 for seasonal influenza.

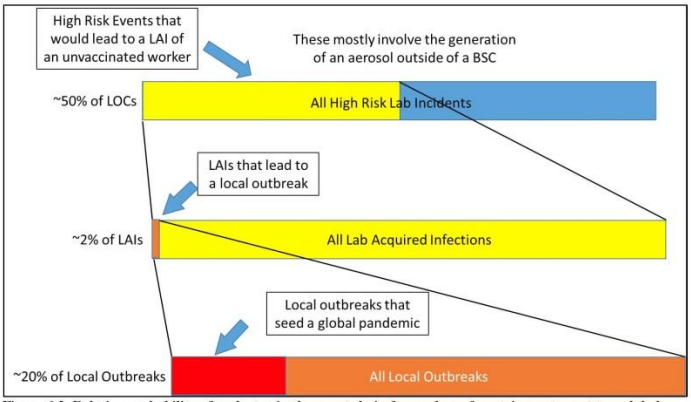

Figure 6.2. Relative probability of each step in the event chain from a loss of containment event to a global pandemic for a loss of containment event involving seasonal influenza.

The absolute probability of an escape from a single lab in a single year though an LAI or other routes, p_1, is not shown in the graph. Making a reasonable guess for this absolute probability is the subject of Section 6-8 of the Gryphon report. For the second and third steps, the probability that the LAI will lead to a local outbreak is estimated by Gryphon to be about 2% for seasonal influenza, and the percentage of local outbreaks that will lead to a pandemic is about 20%. The probability that a single lab in a single year seeds a pandemic, pan_1, is then

$$pan_1 = p_1 \times 0.02 \times 0.2 = 0.004\ p_1 \qquad (1)$$

The 0.004 or 0.4% figure is quoted many times throughout Chapter 6 (sometimes Gryphon uses 0.5%). It is the result of their analysis of risk using branching theory and the HHS-BARDA Interactive Influenza Model.

Gryphon's dividing the path from a lab escape leading to a pandemic into two steps--(1) the escape causes a local outbreak and (2) the local outbreak then causes a pandemic--is not necessary. A single infected researcher can seed a pandemic directly. From Figure 4 in the Lipsitch *et al.* (2003) paper, the probability that the single infected researcher can seed a pandemic is 10% to 30% (for R_0=1.3 and smaller k values). Thus, the 0.4% value is likely 1/0.02 = 50-times higher due to eliminating this intermediate local-outbreak step.

These are two well-established methods; and given Gryphon's high-level mathematical and analytic skills, I will use the 0.4% Gryphon number to stay closer to their analysis. In Gryphon's words,

> "Sufficient biomedical and epidemiological evidence exists to develop robust models of the initiation of an outbreak from the primary to the secondary cases and the expansion of this outbreak within a community to eventually spark a global pandemic."

For a "research enterprise" of 10 x 10 = 100 lab years, the probability that some lab in some year will seed a pandemic is approximately 100 x pan_1 or 100 x 0.004 p_1 = 0.4 p_1. Clearly, p_1 is the key probability to carry out the analysis with high confidence. The two parts of Gryphon's and my analysis that are uncertain are values for the probabilities p_1 and for the case fatality rate.

To obtain an absolute probability for p_1, in Section 6.8 Gryphon basically guesses. In Gryphon's words,

> "…absolute risk estimates are desired. For this reason, the historical rate of laboratory acquired infections could be used to predict a reasonable upper bound for the frequency with which these incidents occur. However, the research team is unaware of any laboratory acquired infections in laboratories that study influenza or coronaviruses and so an absolute risk analysis will have at its foundation a weak estimate of the frequency at which laboratory acquired infections occur. That being said, this historical rate of laboratory infections can then be combined with calculated rates of laboratory acquired infections leading to secondary infections, local outbreaks and global pandemics from this assessment to produce an estimate of absolute risk."

The remarkable observation here is that in 100 mostly seasonal influenza BSL2 research labs over 20 years of research, Gryphon was unable to find any reported LAIs. Gryphon offers the following explanation:

> "The project team knows of no laboratory acquired infections involving any one of these laboratories. This lack of a laboratory acquired infection could be due to the fact that none have occurred in that time frame or that some have occurred but the project team does not have access to the reports or data."

The report neglects additional possible reasons: asymptomatic or subclinical infections, or misattribution of LAI to the community. If a researcher contracts seasonal influenza, it might not be detected, as a high proportion of seasonal influenza is subclinical particularly among individuals with considerable levels of natural immunity or immunity from vaccinations. If it were detected clinically, it would likely be attributed to a community infection, not from the lab. In any case, reporting it as possibly an LAI would lead to time-consuming follow up. It could be unspoken policy in seasonal influenza research labs to not report infections of uncertain origin given that the infected person will be better in few days. I find it difficult to believe that there have been no LAIs in 100 mostly BSL2 labs in 20 years. That would be inconsistent with rates of LAI in other BSL2 labs, even in settings where underreporting is known to be a problem.[1]

In any event, where Gryphon expected to find statistically-useful real data on LAIs in seasonal influenza labs, it found none. I suspect Gryphon then resorted to historical data from other labs researching other pathogens to obtain its range of zero to ten LAIs. Gryphon raises a valid and important point on using accident data from other pathogens and laboratories.

> "very little data exists on human reliability in life science laboratories, which drives the probability that laboratory acquired infections occur in the first place. Fortunately, the accidents that humans cause (or contribute to) in the laboratory are the same regardless of the pathogen manipulated. That is, workers may overfill a centrifuge tube with the same frequency regardless of the pathogen in the tube, or will slip while working with scissors during a necropsy with the same frequency regardless of the pathogen studied. Because the absolute rate at which these accidents happen and cause infections is not supported by robust data, absolute estimates of the rate of laboratory acquired infections cannot be made using the method described in this report."

Lacking real data, Gryphon makes an educated guess that perhaps three LAIs did occur in the hundred mostly seasonal influenza labs over the twenty years. Gryphon calculates

> "Across all 100 laboratories…if the assumption is made that three LAIs have surreptitiously occurred, then …a global pandemic could be triggered once every 750-5,000 years."

Gryphon chooses to report its findings as "return periods" in years, not probabilities. Return periods are the reciprocal of probabilities per year. My problem with return periods is that they can fool you into thinking something is safe when it is not when consequences are considered. It is necessary to stick to the more fundamental probabilities for calculations.

For seasonal influenza, with Gryphon's guess of 3 LAIs in 20 x 100 = 2,000 lab years, the probability of an LAI (escape) per lab per year is $p_1 = 3/2{,}000 = 1.5 \times 10^{-3}$. (Three LAIs in over 2,000 lab years seems conservative to me, there were likely more.) Thus, the return period for one lab in one year is $1/p_1 = 667$ years for an LAI to occur. This may seem like the experiments are safe, as they will be completed in

[1] Marc Lipsitch contributed to this paragraph

perhaps 10 years, well short of the return period. But looked at another way, in 20 years this means that there are three LAIs, where each one has a not-insignificant chance of causing a seasonal influenza pandemic. I would not accept those odds.

What is the probability, $p_{1,HPAI}$, for research on mtHPAI? I assume that research on mtHPAI is conducted in BSL3 labs using the level of biosafety for research on SARS, as SARS has a case-fatality rate of around 10% considerable caution is warranted. Gryphon lists relative probabilities compared to work with seasonal influenza in their Table 6.2, reproduced here.

Table 6.2. Relative probability of a laboratory acquired infection for the various pathogens considered in this study as compared to work with seasonal influenza.

Pathogen	Biosafety Level	Relative Probability of an LAI*
Seasonal influenza virus	BSL2	1 (defined)
Pandemic influenza virus	BSL3	0.10 (0.07-0.15)
Avian influenza virus	BSL3	0.43 (0.21-0.90) (mostly of birds)
SARS-CoV	BSL3	0.03 (0.02-0.04)
MERS-CoV	BSL3	0.01 (0.006-0.02)

These data are generated by comparing the sums of the frequency of infection from all loss of containment pathways for each pathogen. In this case, we use the term laboratory acquired infection to include an infection of wild birds to capture the comparative risk of working with avian influenza viruses. The numbers in the parenthesis are the results from the p5 and p95 outputs of the Monte Carlo analysis.

Before using data from Table 6.2, this is a good place to state what I view as a major shortcoming in the Gryphon report. Sources of data and calculations to obtain it are not referenced throughout Chapter 6. Are the sources not referenced in the Supplementary material? In the published literature? In spreadsheets available from Gryphon? In Table 6.2, for instance, the caption could have provided references. Thus, we don't know how solid or significant various pieces of data are, unless Gryphon chooses to discuss it. I suspect that Gryphon could have used much more time in preparing its report.

Furthermore, Gryphon ignores the frequency of accidents over the years in labs researching Select Agents compiled by the CDC in 2013. Gryphon's analysis also ignores the highly publicized recent accidents in the CDC lab. While none of these accidents involved seasonal influenza, somewhere in Chapter 6 they should have been acknowledged and incorporated into their analysis. It is unclear why guesses well below the empiric rate of LAI should be used for a risk analysis. Nonetheless, in what follows, Gryphon's numbers are accepted for the sake of argument.

From Table 6.2, the probability of an LAI in a SARS lab is a factor about 0.03 times that of seasonal influenza. Specifically, $p_{1,HPAI} = 0.03 \times 1.5 \times 10^{-3} = 4.50 \times 10^{-5}$ for a SARS or mtHPAI lab where $p_{1,HPAI}$ is the probability of an LAI for a single lab for a single year. The probability of a pandemic from a single lab in a single year, pan_1, is

$$pan_1 = 0.004 \times p_{1,HPAI} = 0.004 \times 4.50 \times 10^{-5} = 1.8 \times 10^{-7}$$

As an illustration, I conservatively estimate 10 labs conducting mtHPAI research for 10 years (100 lab years),[2] each with the laboratory safety of a SARS lab. The probability that the research enterprise will seed a pandemic, RE, is approximately

$$RE = 100 \times pan_1 = 1.8 \times 10^{-5}$$

The return period, 1/RE, is 55.6 thousand years, which would seem to make the research very safe if it were not for the potential consequences of millions of fatalities.

The likelihood-weighted pandemic risk, LWR, is given by

LWR = (Probability of a Pandemic) x (Consequences of a Pandemic)

Consequences are restricted to fatalities in this analysis. The case fatality rate was chosen to be 5%, which is twelve-fold less than the World Health Organization's accepted case fatality rate of 60%. For a pandemic infecting 25% of the world's population of 7.3 billion, the number of fatalities, F, would be

$$F = 7.3 \text{ billion} \times 0.25 \times 0.05 = 90 \text{ million.}$$

With these numbers, the likelihood-weighted fatalities, LWF, for the research enterprise is

$$LWF = RE \times F = (1.8 \times 10^{-05}) \times (90 \times 10^{6}) = 1{,}640.$$

The Likelihood-weighted fatalities for a single lab in a single year is 1,640/100 = 16.4, which I call the "fatality burden" for the single lab in a year. As pointed out earlier this fatality burden is likely 1/.02 or 50 times higher. To put this fatality burden in perspective, no Institutional Review Board tasked with assessing human subject research would approve a proposed research project with an expected 16.4 fatalities per year (or 50 x 16.4 = 820 fatalities per year, accounting for the 50-fold error discussed above). There are research approaches not involving live mtHPAI for elucidating the molecular virology of airborne transmission. Such safe research approaches ought to be employed, and research with lab-made, airborne-transmissible, live mtHPAI be banned.

One point still needs to be discussed, case fatality rate. The 5% case fatality rate used in this analysis is much higher than the small fraction of 1% claimed by Morens and Taubenberger. There are well-documented studies (for instance, here and here) that claim the case fatality rate is not low but close to the 60% often quoted for wild type H5N1 HPAI. But the airborne-transmissible mtHPAI, a key focus of the NIH deliberative process, are not wild type viruses. They infect lung to lung via the airborne route. We do not know the case-fatality rate for these strains. It could be quite high, perhaps over 60%. So, arguments over case fatality rate for wild-type HPAI are likely moot. Because the potential consequences in morbidity and mortality are potentially high, caution dictates instituting a ban on making and researching live airborne-transmissible mtHPAI.

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[2] Gryphon estimates "approximately 40 research groups in the US because these groups have been performing, or have the capacity to perform, certain types of GOF experiments involving influenza, MERS, and SARS viruses. This maximum number is supported by the case studies examined which showed that a new discovery in virology may proliferate to as few as one and as many as 70 new groups around the world within 10-15 years."

The Gryphon report seems to dismiss gain-of-function studies in SARS and MERS, by assuming that mitigation measures such as quarantine should prevent a large outbreak. SARS has about an eight-day incubation period before an infected person can transmit infection. Timely and strong mitigation measures may be possible in developed nations, but we need only to look at the Ebola epidemic in the poor and war-torn African nations to understand the potential for large outbreaks. GOF studies in SARS and MERS should be looked at very carefully and many perhaps banned as well.

December 31, 2015

Christopher Viggiani, Ph.D.
Executive Director, NSABB
NIH Office of Science Policy
6705 Rockledge Drive, Suite 750
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nsabb@od.nih.gov

RE: Draft Report By Gryphon Scientific, *Risk and Benefit Analysis of Gain of Function Research*

Dear Dr. Viggiani and Members of NSABB:

Below are my comments on the draft GOF report prepared by Gryphon Scientific. I have spent nearly 35 years as an industrial hygienist on the staff of labor organizations, most recently with the Safety and Health Department of the AFL-CIO (from which I retired in October 2013). During these years, I devoted my efforts at protecting workers from exposures to hazardous substances and infectious agents, some of which are the subject of the referenced report. At the AFL-CIO, I held major responsibility for representing the AFL-CIO at OSHA rulemaking proceedings and public hearings on proposed safety and health standards. I also served on various policy and scientific committees addressing occupational safety and health issues, including two Institute of Medicine committees dealing with respiratory protection for healthcare worker exposed to H1N1 influenza and personal protective equipment for healthcare workers exposed to pandemic influenza and other viral respiratory diseases.

I would like to make the following points on the draft document:

(1) When OSHA issues any major proposed new or revised safety and health standard, the agency typically provides for a minimum 90 day period for submitting written comments and documents to the record, followed by public hearings and a post-hearing opportunity to submit additional comments and documents to the record. In the case of the draft GOF Report, a period of less than 30 days was established for receiving public comments. Additionally, this shortened timeframe included the holiday and New Year period. A comment period of less than 30 days for a report of such importance and magnitude as the draft GOF is absurd. It does not provide for an adequate time period in which to digest, analyze, and respond to the many critical issues raised in the report. Instead, this woefully shortened comment period has all the appearance of nothing more than a superficial attempt at giving the public an opportunity for

comment while the real underlying objective is to move the process to a rapid conclusion. In my view, the comment period needs to be extended considerably.

(2) I have serious concerns that the report fails to address the consequences of the release of highly virulent and drug resistant viruses in the laboratory as well as the general environment. The infection risk posed to laboratory workers who are exposed to these newly designed agents, via whatever protective measure is breached, is hugely problematic. With no effective drugs or vaccine available, a highly virulent virus is likely to cause serious, if not fatal, adverse health effects in an infected lab worker. And healthcare workers who provide care to infected lab workers are even more vulnerable, given the absence of preparedness by our healthcare facilities to protect its workforce from patients infected with dangerous viral agents – one only needs to examine the problems uncovered in Dallas, TX for healthcare workers during the Ebola outbreak. This problem is further magnified enormously by the fact that 49 of the 50 states in the United States do not have a mandatory OSHA standard that requires employers to protect workers from infectious disease (California being the exception). Instead, CDC guidelines are merely recommendations that state and local health departments and healthcare facilities can ignore with impunity (this was rampant during the H1N1 pandemic).

(3) I'm not convinced that the benefits of this research path outweigh the risks. Once the genie is out of the bottle, it will be difficult to put it back in. Developing the technologies to create highly virulent, drug resistant infectious agents represents a security risk that is too dangerous to undertake. For when the technologies are developed in the US, essentially all governments and other forces will at some point be able to utilize the technologies, be they friendly government's or not. In the "wrong hands", we would then have a huge problem to address.

Thank you for the opportunity to submit these comments,

Sincerely,

Bill Kojola

Bill Kojola

Silver Spring, MD 20902

Written comments for NSABB meeting Jan 7-8, 2016

Marc Lipsitch, DPhil

Harvard T.H. Chan School of Public Health

Cofounder, Cambridge Working Group

Contains original written comments submitted December 31, 2015 plus additional comments (on benefits) submitted January 3, 2016. Additional comments added to this version concern the Benefit Assessment and are in dark red font.

Dear Chairman Stanley and Members of the NSABB:

I am pleased to have the opportunity to offer written comments pertinent to the upcoming meeting of the Board, specifically concerning the Risk-Benefit Assessment provided by Gryphon Scientific and the Working Paper Draft dated December 23, 2015 by the NSABB in response to the RBA. I consider these in order and conclude with some comments on the process. My comments are in no sense a complete evaluation of any of these documents, given their enormous length and the short time available. I may choose to submit additional comments at a later date. These are simply my comments on the most important issues I have noticed in the time available.

In these comments I make reference to written comments submitted by other members of the public. I will not reiterate the details of their arguments, but I register my agreement with them in particular cases.

I. Comments on the NSABB working paper (WP)

Comment I.1. Overall, the working paper accurately identifies that the research involving a reasonably anticipated creation of a strain combining high virulence and high transmissibility is the central "Gain of Function of concern" research that should be the focus of scrutiny. That has been apparent since the start of this process, and it was the NSABB that broadened the charge of Gryphon to include many less-risky experiments. The NSABB has now appropriately narrowed the focus to GOF of concern.

Comment I.2. The scope of GOF of concern identified by the NSABB, however, is unduly narrow. It includes as a condition for GOFoc, not only combined virulence and transmissibility, but also the ability to evade countermeasures. This is inappropriate because countermeasure availability for a transmissible, virulent strain produced by GOF is not guaranteed even to the US, and timely countermeasures will be unavailable for the vast majority of the world. Thus even a strain susceptible to antivirals and to immunity produced by a hypothetical vaccine could do tremendous damage. **Resistance to countermeasures should be deleted from the requirements for GoFoc.**

Comment I.3. The WP fundamentally fails to answer the question posed in the NSABB's own Principle 9 to determine "whether there are certain studies that should not be conducted under any circumstances, and if so, articulate the critical characteristics of such studies." Instead, it states "There are life sciences research studies that should not be conducted on ethical or public health grounds if the potential risks associated with the study are not justified by the potential benefits" (p. 4). **This is an abdication of responsibility given that the Working Paper is a response to a 1000-page RBA.**

Comment I.4. Given the findings of the RBA, the most important of which is that a single year of BSL3 work on mammalian-transmissible high-path avian influenza has an expected fatality toll of some 50+ lives, **creating mammalian-transmissible avian influenza is GOF of the highest concern and should not be undertaken**. Similarly, creation of novel coronaviruses with transmissibility similar to SARS have, by Gryphon's reasoning, an expected toll of >10 lives per laboratory-year. This also is research that should not be

undertaken, by Gryphon's own reasoning (here I rely heavily on the Public Comments submitted by Lynn Klotz). As noted by Klotz, no Institutional Review Board would approve a research plan with an expected fatality toll in this range. The fact that the expected fatality toll is in this case a low probability of a catastrophic death toll should, if anything, be an even stronger bar to such activities.

Comment I.5. Recommendation 2, that "In general, oversight mechanisms for GOF studies of concern should be incorporated into existing policy frameworks" should be modified or replaced. **There is strong evidence that existing policy frameworks are *inadequate* to regulate GOF of concern.** That evidence includes the following:

- Prior to the Funding Pause in October 2014, HHS had put in place a Framework for review of H5N1 GOF research [1] and later for H7N9 GOF research[2]. These frameworks were inadequate in that (i) no formal risk or benefit assessment (ie nothing quantitative) was done when HHS considered these studies [this I have heard from a participant in the review]; (ii) the review was done in private with no public input; (iii) the same day that the H7N9 framework was published [2], Fouchier and colleagues published a paper describing HHS-sponsored GOF research on H7N9 (see http://comments.sciencemag.org/content/10.1126/science.1244158). This is prima facie evidence of the inadequacy of the Frameworks.
- During the funding pause, Baric and colleagues published a paper [3] describing NIH-funded experiments that by any standard met the terms of the funding pause: "may be reasonably anticipated to confer attributes to influenza, MERS, or SARS viruses such that the virus would have enhanced pathogenicity and/or transmissibility in mammals via the respiratory route." While the circumstances surrounding this work (in particular why it was permitted under the funding pause) have not been publicly described, this is clear evidence that even enhanced scrutiny may be circumvented by NIH as funder and/or an investigator.
- These instances, along with common sense, indicate that placing NIH or CDC (both direct funders and in the case of CDC, performers of GOF of concern research) as the judges of what may and may not be performed is a direct conflict of interest and is not a way to arrive at impartial judgments.

Given these considerations, an interagency task force that receives input from HHS but is independent of it seems much preferable to existing mechanisms[4]. Expansion of the Select Agent rule to prohibit GOF of concern without the specific consent of such a board would be a possible policy solution.

Comment I.6. **The suggestion to use existing regulatory approaches for regulating GOF of concern requires that institutional oversight have the capacity to deal with this topic, making fine distinctions that have not yet been defined, much less codified in ways that can be applied at the institutional level.** There is no reason to think that Institutional Biosafety Committees have the requisite expertise to perform risk-benefit evaluations on this scale. As an example, the minutes of the University of Wisconsin IBC obtained by *Nature* for GOF work by Prof. Kawaoka (http://www.nature.com/polopoly_fs/7.18249!/file/WISC_Review.pdf) contain no numerical estimates of risk (that is to say, do not perform risk assessment, although they assert on p. 1 that it includes a risk benefit assessment) and accept uncritically all assertions of the investigator about benefits of the proposed work, including false statements ("The proposed research will determine the likelihood of an influenza virus similar to the 1918 pandemic strain of [sic] emerging naturally." The research has been published, and that likelihood has not been determined. Thus the benefit assessment cannot be considered adequate either. *This further demonstrates the inadequacy of existing regulatory mechanisms to deal with GOF of concern.*

II. Comments on the Gryphon Risk-Benefit Assessment (RBA)

Comments on Biosafety Risk.

Comment II.1. There is a presumption in the RBA, starting with the Executive Summary, that experiments with the pandemic H1N1 strain of 1918 constitute an acceptable level of risk against which other experiments should be compared. Moreover, it is stated (section 1.1) that " No GoF experiment is likely to create a strain riskier than work with wild-type 1918 H1N1." **Both the assumption that this level of risk is acceptable, and the claim that no GOF experiment is likely to create a strain riskier than work with wt 1918 H1N1, are unjustified.** The source of either claim is unclear, and in particular the claim that no more dangerous strain exists is based on a misreading of the literature on H1N1 case-fatality risk (see comment below). The quoted statement also directly contradicts the statement (RBA p. 78-9): "In short, a strain of influenza virus that is as transmissible (or to which the population has as little minimal immunity) as newly emerged pandemic strains WHILE leading to a case fatality rate of more than 5%, would pose more of a risk of a global pandemic than any wild type strain heretofore identified. No experiments that are likely to be conducted under the rubric of GoF research will drive risk more than this combination of traits or significantly increase the risk of a laboratory acquired infection."

Comment II.2. The RBA appropriately identifies creation of novel viruses combining mammalian virulence with mammalian transmissibility as the most risk-enhancing experiments (Figure 6.1). Notably, it does *not* add "resistance to countermeasures" to this category, although it does note that resistance to countermeasures would further enhance the risk of such experiments in the developed world, where countermeasures might be available. **I recommend that the NSABB adhere to this classification, without requiring resistance to countermeasures, when defining GOF of concern.**

Comment II.3. Notwithstanding the serious flaws in the analysis that lead to an underestimate of the risk of such experiments, I draw the NSABB's attention to the fact that: Using Gryphon's own numbers, the expected fatality toll from a lab-year of coronavirus experimentation with enhanced transmissibility in BSL3 is approximately 16 fatalities

(Written comments of Lynn Klotz to the NSABB, December 2015). A corresponding calculation for mammalian-transmissible avian influenza would be around 50 fatalities. **Absent an exceptionally compelling prospect of life-saving, justly distributed benefits, this conclusion from the RBA merits the immediate discontinuation of experiments meeting the definition of GOF of Concern proposed by the NSABB, with the modification suggested above to remove the requirement for escaping countermeasures.**

Comment II.4. The RBA contains a number of erroneous parameter assumptions that lower the estimate of risk of various experiments relative to appropriate estimates. These are shown in a table below.

Table 1: Errors in the Risk Assessment Leading to Underestimate of Risk

Assumption	Source of Error and corrected assumption	Impact on risk estimates
CFR of 1918 influenza is 10-20% of infected persons (Table S7 in supplement http://www.gryphonscientific.com/wp-content/uploads/2015/12/Supplemental-info-disease-course-of-influenza.pdf)	Misreading of a graph in the reference cited, ref 82. Actual values are mainly in the range of 0.5%-3% of those with clinical disease (except for extremes of age). This is therefore a 6-20x overestimate, not accounting for medical improvements and larger denominator of asymptomatic cases)	Allegedly acceptable risk of experiments with 1918 pandemic flu are significantly overstated, raising the bar for what should be permitted to a much higher level and seemingly justifying false statements like that noted in Comment II.1.
CFR of influenza is 0.0001%-0.00043% of those infected (Table S7 in supplement http://www.gryphonscientific.com/wp-content/uploads/2015/12/Supplemental-info-disease-course-of-influenza.pdf)	Error source unclear. Actual estimate from authoritative systematic review [5] is 0.001%-0.010%. Thus this is more than a 10x error.	Suggests manipulations of seasonal influenza have smaller risk than they do.
R0 of SARS is 1.5, may go as low as <1 (http://www.gryphonscientific.com/wp-content/uploads/2015/12/Supplemental-information-R0-of-CoV.pdf).	This seems to result from a combination of not understanding what R0 is (it does not incorporate the later stages of the epidemic or the impact of control measures), especially as used in a branching process. Averaging over different phases of the epidemic is completely inappropriate. Two of the three authoritative estimates of R0 are not cited; with Riley (cited) they all estimated approximately 3.0 [6-8]	Significantly underestimates severity of SARS outbreaks
Control measures (community mitigation) will be effective	There is no evidence of this in modern influenza pandemics	Underestimates severity and probability of pandemic from

		modified influenza strains
Assumes that all event trees for LAI happen in the source lab at the specified biosecurity level	Errors with a probability of leading to a LAI have repeatedly, consistently occurred outside the source lab, usually at a lower BSL. For example, 2014 CDC anthrax exposure occurred in BSL2 after inadequate decontamination; 2014 CDC HPAI exposure occurred outside source lab (though fortunately at BSL3) due to contamination of sample; 2014 CDC Ebola exposure occurred at BSL2 due to falsely assumed decontamination and removal to lower BSL; 2015 DOD anthrax exposures occurred in conditions designed for inactivated anthrax because of lack of proper inactivation.	This leads to neglect of a fault tree that routinely occurs in top US government labs, in which the probability of LAI is higher, the likelihood of its going undetected is higher, the likelihood of having prophylactic measures in place for laboratorians is lower, and thus the risk of outbreak and escaping local control is higher. For more details, see [9].
Probability that a single LAI with a pandemic-capable influenza triggers a pandemic is 0.4%.	Other branching process models, which account for negative-binomial overdispersion, find estimates of 5-60%[6, 10, 11]	Vastly underestimates by 1-2 orders of magnitude all risks.

Comments on biosecurity

These may be supplied at a later date when time allows.

Comments on benefits of GOF

Comment II. 5. A very good feature of the BA is the consideration of alternatives to GOF experiments to either answer the same scientific questions or achieve similar public health benefits in a different way. Had appropriate skepticism been applied to the claims of those performing and sponsoring GOF research, these alternatives would have proven far more

compelling than the Benefit Assessment suggests**. The extreme skew of the experts consulted for the Benefit Assessment (see Section III below), combined with a surprisingly credulous evaluation of their claims, leaves the BA with a number of statements that do not stand up to scrutiny.**

Comment II.6. The vast majority of the public health benefits asserted for GOF experiments are for the development of costly countermeasures, including vaccines and antiviral drugs**. These benefits will be limited to the wealthiest populations, which have access to the newest drugs and vaccines.** This problem is recognized in the BA, for example with respect to antiviral development in the statement (p. 438): "In sum, although U.S. policy supports the donation of influenza antivirals in the event of a pandemic, the relatively small number of doses donated in comparison to the global need in the event of a pandemic means that developing countries would face shortages, which would in turn exacerbate poor usage in-country." In the case of pandemic preparedness benefits, similar statements are made (pp. 442 and 444) In contrast, the risks of GOF research, which are distributed globally and if anything will fall harder on lower-resource populations, [12], As recently as 2009, developing countries had little access to antivirals or vaccines until long after the peak of pandemic risk. **In this sense, GOF experiments unjustly require unconsenting populations to bear pandemic risk while promising them no realistic prospect of benefit. This is a serious and independent ethical objection to such research, which is not adequately addressed in the separated ethical analysis commissioned by NSABB.**

Comment II.7. At multiple points in the BA and in the corresponding section of the Executive Summary (1.4), there are statements that particular types of experiments involving the evasion of novel therapeutics or vaccines involve no human health risk because the countermeasures are not yet extant. This statement is false unless one assumes that the immunity produced by novel vaccines, and the protection by novel treatments, is unrelated to that produced by existing natural exposure or vaccines (for immunity) or antivirals (for resistance). Vaccine-related immunity and natural immunity may involve the same epitopes (especially as vaccine development is often based on observations of naturally acquired immunity), and cross-resistance between novel and existing antivirals within a class is expected, just as cross-resistance within existing classes (eg zanamavir and oseltamivir, or

rimantadine and amantadine) can occur with the same mutation. **In summary, these statements -- that GOF to evade countermeasures not yet available has no human health risk -- are unjustified and tend to underestimate the risk of corresponding GOF experiments.**

Comment II.8. Virtually all of the benefits of GOF experiments described in the Benefit Assessment are characterized as *not* unique to GOF (Table 9.1, 3rd column). This is extremely important, as it means that the Benefit Assessment characterizes nearly all of the claimed benefits as being achievable by alternative means. While some of these alternative means involve localized risk of infection of a few laboratory personnel, these risks are minimal in comparison to pandemic risk. Thus **the BA implies that nearly all of the benefits of GOF (especially of GOF of concern) could be achieved with alternatives that avoid the vast majority of GOF risk. This finding creates a strong presumption in favor of alternative approaches [13]. Indeed, under such circumstances, I would argue it is unethical to perform GOF of concern experiments[14].**

Comment II.9. It is stated (Section 1.4, p. 6) that "GoF approaches that enhance virulence represent the most efficient and effective strategy for discovering novel virulence factors, which may be good targets for new therapeutics." This does not make sense. If the virulence factors found are not present in naturally circulating strains, then finding changes that could result in increased virulence could only facilitate the development of therapeutics for strains that do not exist. **Development of therapeutics for nonexistent strains would be a highly speculative activity with little likelihood of being supported in the absence of a foreseeable market.**

Comment II.10. The most important unique benefit asserted for GOF of concern (enhancement of mammalian transmissibility of avian influenza) is informing pandemic risk assessment and prioritization of countermeasures. The BA asserts these are of particular importance in rapid risk assessment and prioritization: "GoF data play an important role in rapid risk assessments when novel flu viruses first emerge in human populations due to the early availability of sequence data. These risk assessments facilitate more rapid initiation of response activities such as pre-pandemic vaccine development" (p. 244).

The assertion of these unique benefits represents an uncritical acceptance of the assertions of GOF proponents that is contrary to the evidence. The assertion has four **fatal flaws**:

1. **Every mutation cited by GOF proponents as having been discovered in GOF experiments and used to prioritize pandemic response [15, 16] has been found (in most cases prior to the GOF studies) in a non-dangerous, non-GOF study and identified as a predictor of pandemic risk.** Thus the claim of uniqueness is unjustified (see Table below). Alt-GOF can, and indeed have, identified mutations and phenotypes of concern.
2. While it is true that GOF-identified mutations have been used to inform surveillance and preparedness strategies, **there is no evidence that the use of such findings has improved the accuracy of these strategies.** Using information is different from using it productively. There is no case in which a pandemic has been anticipated using GOF-derived data. The evidence that decisions are improved is weakened even further by the fact that many GOF mutations have highly context-dependent effects, so that they may or may not be predictive in actual wildtype strains [17, 18].
3. **GOF data may be misleading, resulting in worse not better decisions.** In the one case when a pandemic has emerged during the era of widespread virus sequencing (2009) it lacked the mutation PB2 E627K[17], which has been identified as perhaps the most important single GOF mutation for mammalian adaptation [19]. Surveillance did not identify this virus in swine before it became pandemic, but had it been identified, use of GOF data would have incorrectly classified it as low risk. Ruling out one of the four strains that caused a pandemic in a century as low risk would be a remarkably large error. Incidentally, this story also highlights the uselessness of any genetic information when surveillance does not catch a strain before it emerges. No pandemic strain has ever been discovered in animals before it caused a pandemic.
4. The accuracy of ferrets in predicting human transmissibility is imperfect, though they are the best available model [20]. Indeed, **several GOF researchers and proponents have said in public meetings that they expect the strains isolated from ferret transmission experiments would not be readily transmissible in**

humans. This uncertainty nullifies or even negates the benefit for pandemic preparedness, because mutations identified in these studies, which are being used as *positive predictors of human pandemic potential*, are in fact uncertain predictors and may not indicate human transmissibility. This could mean that strains with little human pandemic potential are tagged for special prevention efforts, and/or that strains with different genetic profiles that are actually high-risk are identified as low-risk and deprioritized. Notably, this uncertainty makes the use of such mutations highly impractical for decision-making, yet it does not nullify the risk presented by these strains. It negates or nullifies the benefit, and yet only reduces the risk, because the statement that the GOF strains would not be pandemic-capable in humans are informed guesses, which may be wrong.

Table 2: Non-uniqueness of benefits for GOF of concern studies for pandemic response

Mutation claimed to be significant based on GOF by Davis [15]or Schultz-Cherry [16]	Prior studies not involving PPP creation that identified these mutations	Counterexamples
H5 &H7N9 HA Q222L HA	[21-23] [18, 24-26]	CONTEXT DEPENDENCE: Changes do not quantitatively shift receptor binding in related H5 strains [18]
H5N1 HA S133A S135N S123P S155N	[23, 27]	
H7N9 HA T156A, Q222L	[28, 29]	
PB2 E627K, D701N	[30]	MISLEADING INFERENCE: Both absent in 2009pdm [17]. Would have led to its misclassification as low risk

Comment II.11. I endorse the critiques submitted as comments to the NSABB by Dr. Stanley Plotkin of the asserted benefits of GOF experiments. These represent further examples of the widespread exaggeration of benefits and downplaying of alt-GOF in the Benefit Assessment. I will not recapitulate these here but simply incorporate them by reference to his remarks.

III. Comments on the NSABB process

On the whole, I would characterize the process of the RBA development as distinctly unwelcoming of public participation, and as heavily weighted in favor of those who do and fund GOF of concern research. Major shortcomings include the following:

- At all in-person meetings of the NSABB including the upcoming one, public comment has been possible only in writing or in person, but not in real time by any electronic medium. This excludes many persons who may wish to comment in real time on the proceedings but do not have the ability to attend in person.
- The development of the RBA included site visits and conversations with many investigators in 14 labs, most of which do GOF research. The benefit assessment in particular received more than 80 percent of its input from scientists who do PPP research or representatives of agencies that fund it (RBA Fig. 9.3). In contrast, only about 10 (12%) of those interviewed for the benefit assessment were persons who have expressed reservations about RBA research.
- The timeline for public comment was extremely short, with the NSABB waiting apparently 2 weeks from the time it saw Gryphon's RBA until it posted it publicly, and then only 1 month (including Christmas and New Year's) before its meeting. There were only 8 days including Christmas from the release of NSABB's draft working paper to the deadline for public comments to be submitted and seen by the NSABB members.
- The unbalanced representation of GOF researchers/funders versus those who have raised concerns is continued in the agenda for the January 7-8 meeting. 3 outspoken critics are on the panels, plus one additional member of the Cambridge Working Group; 9-10 funders or researchers of GOF studies are speaking. This imbalance was raised in plenty of time to the NSABB leadership, which chose not to address the problem.

Overall, it is difficult to see this process as having been designed to maximize public input or to achieve balance between proponents and critics of GOF, or indeed to address the

inherent conflicts of interest of those whose research or funding portfolios are at issue in the discussion.

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January 3, 2016

National Science Advisory Board for Biosecurity
Office of Science Policy, OD
Rockledge 1, Suite 750
6705 Rockledge Drive
Bethesda, MD 20817

Dear Chairman Stanley and Members of the NSABB:

I am writing to express my support for the comments submitted by Marc Lipsitch, Stanley Plotkin, and Lynn Klotz. I am deeply concerned by the potential fatalities that could result from accidental laboratory infections that might occur in a laboratory conducting gain-of-function research on influenza and other infectious diseases. The number of accidental releases of potentially fatal pathogens in recent years has demonstrated unequivocally that human error is inevitable and impossible to completely eliminate from experiments with deadly pathogens. Specifically, I agree with Dr. Lipsitch that resistance to countermeasures should be deleted from the requirements for Gain of Function of concern research. I concur that the benefits of this research are overestimated, and that the risks are being borne by non-consenting members of the public and disproportionately by those in developing nations that would not be able to implement countermeasures.

Thank you for taking these concerns seriously and including the voices of concerned scientists in your deliberations on how to address the potential dangers to the public from GOF research.

Sincerely yours

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From: Nariyoshi Shinomiya [mailto:shinomi@ndmc.ac.jp]
Sent: Monday, January 04, 2016 3:43 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>; Viggiani, Christopher (NIH/OD) [E] <christopher.viggiani@nih.gov>
Cc: 'Husbands, Jo' <JHusband@nas.edu>
Subject: Written comment to the NSABB meeting
Importance: High

Christopher Viggiani, Ph.D.
Executive Director, NSABB
NIH Office of Science Policy

Dear Dr. Christopher Viggiani,
(CC to Dr. Jo Husbands)

I am a person who were invited by Dr. Amy Patterson to the 2012's workshop on "Gain-of-Function (GOF) Research on Highly Pathogenic Avian Influenza (HPAI) H5N1 Viruses" as a panelist. Since then I have been having a strong interest in this topic. This time I got the information about the NSABB meeting from Dr. Jo Husbands. Unfortunately, I cannot attend the meeting because of my tight schedule. She suggested me to make some comment to the meeting. Here I send my comment about the issue of GOF studies. I hope it is taken up in the session of Public Comment Period or so.

I hope my comment reaches you in time.

Best regards,
Nariyoshi Shinomiya

****
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****

National Defense Medical College

A comment from the viewpoint of balance between scientific advancement and risks to the society

by Nariyoshi Shinomiya
Professor, Department of Integrative Physiology and Bio-Nano Medicine
National Defense Medical College, Japan

Mr. Chairman, distinguished representatives of the NSABB, and participants in the symposium,

It's my pleasure if I could have a chance to make a comment in such an important meeting about gain-of-function (GOF) studies.

In collaboration with the group of the University of Bradford, UK, our research group has developed a biosecurity education module for scientists which is translated into may languages and used worldwide. In my school the biosecurity education is very successful; the educational programs for undergraduates as well as graduate course students are now dealt with a regular subject and supported by the faculty members. I lead a symposium related to "dual use research of concern (DURC) issues" in the Japan Association for Bioethics every year from 2011, in which many participants have an interest in this issue and join the discussion.

As many of you may know, after we introduced the discussion of this issue several years ago, the Science Council of Japan revised a code of conduct for scientists in which an article has been added as one of the most important standards that the scientists should think about. The article says "Dual use concern of scientific research: The scientists should recognize that their research results might be used for malign destructive purposes against their will, so when they perform research activities and make their results public, they select appropriate measures and methods which are acceptable to the society (*the original sentence is written in Japanese*)." Also, the Center of Research and Development Strategy, Japan Science & Technology Agency released a book for strategic proposal entitled "Preparedness Framework and Its Governance of Dual Use Research of Concern for Promising Progress of Life Sciences". However, those efforts just showed a general instruction and a framework. So a precise explanation and a scenario setting in each case of DURC should be added.

Here, I would like to make a comment about GOF studies from the viewpoint of balance between scientific advancement and risks to the society.

I believe the freedom of research activities should be guaranteed to the maximum within professional ethics, yet the following points should be considered.

1. In the risk-benefit analysis, a way of thinking or a condition that the benefit exceeds the risk should be explained in plain words to lay persons. Sometimes the concept, recognition, or perception of risks is quite different among people, and may change depending on the situation. The same thing can be said about the benefits. So, not abstraction but specific idea in each case should be provided.
2. What are real risks in each GOF study? Possible scenarios should be provided, and the influence of the risks needs to be analyzed with accuracy. Are the risks acceptable to the society? If the benefits are considered to exceed the risks, the researchers should ask the society about their research idea and need to get people's consent.
3. It is important for mass media to inform the society about the facts of GOF studies because mass media is the main source for people to get information of this sort. Some mass media may have their own opinions and off course the freedom of speech should be considered, yet information without a bias/arbitrary expression is a priority matter.
4. Similar to nuclear or chemical weapons there is no going back once we get a thing in our hands. So, before making new infectious agents we should deliberate upon the GOF studies. Not only the control of a new infectious agent itself but also the regulation of the information how to make it should be considered as the subject of this issue.

I hope these points are extensively discussed, and clear conclusions are provided in the NSABB meeting.

Thank you, Mr. Chairman, distinguished representatives of the NSABB, and participants in the symposium.

From: Steven [mailto:steven.salzberg@gmail.com] **On Behalf Of** Steven Salzberg
Sent: Tuesday, January 05, 2016 3:26 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Cc: Steven Salzberg <salzberg@jhu.edu>
Subject: comments on risks and benefits of gain-of-function research in the life sciences

Dear NSABB,

Please accept the attached letter as my comments on the risk-benefit assessment provided by Gryphon Scientific and the Working Paper Draft of Dec 23 by the NSABB.

My comments are very brief, but given the time constraints I didn't have time to write more. Nonetheless I feel this is such a critical issue that I wanted to at least register my grave concerns about the continuing efforts by a small number of scientists to create highly virulent viruses in their laboratories.

Sincerely,
Steven Salzberg
--

Steven L. Salzberg, Ph.D.
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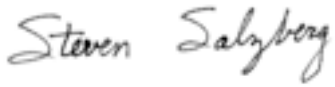
January 5, 2015

Dear Chairman Stanley and Members of the NSABB:
I'm writing to express my strong support for the comments submitted by Dr. Mark Lipitsch, which I have read closely and with which I agree in almost every detail. I am very concerned that the continuing gain-of-function research on influenza viruses, and more recently on other viruses, presents extremely serious risks to the public health. As a former influenza researcher myself, I also concur with Dr. Lipitsch and others that the benefits of gain-of-function research are minimal at best. These minimal benefits could easily and far more safely be obtained through other avenues of research.

In addition to my primary research at Hopkins, I also write a popular science blog at Forbes magazine, where I expressed grave concerns about this topic in August 2013, in an article that had over 50,000 hits (see http://www.forbes.com/sites/stevensalzberg/2013/08/08/scientists-will-create-a-deadly-new-flu-strain-just-to-prove-they-can/). As I wrote then, it seems clear that some of the scientists leading the GOF research on influenza are doing it primarily for the publicity and acclaim (including publication in high-profile journals), while downplaying the risks. Their primary justification for their work–that lab-created influenza strains will teach us how to avoid or treat future pandemics–has no evidence to support it.

I am pleased that the U.S. government has called for a pause in this research, and I strongly urge you to recommend that this pause become permanent. Continuing research that is intended to make influenza or other viruses more infectious, or more deadly, carries great risks and almost no practical benefits.

Sincerely,



Steven Salzberg, Ph.D.
Bloomberg Distinguished Professor of Biomedical Engineering, Computer Science, and Biostatistics
Director, Center for Computational Biology
McKusick-Nathans Institute of Genetic Medicine
Johns Hopkins School of Medicine

From: Charles Stack [mailto:cstack3@uic.edu]
Sent: Wednesday, January 06, 2016 10:04 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: NSABB Public Comment regarding Gain of Function safety
Importance: High

I am a Public Health Advisor to the FBI through the Chicago "Infragard" Chapter and have this comment regarding your upcoming NSABB meeting.

I have reviewed the "Risk and Benefit Analysis of Gain of Function Research" Draft Final Report, December 2015 and am VERY concerned that the largest, deadliest incident of domestic breach of biosafety, namely the "Amerithrax" incident involving the late Bruce Ivins PhD, was only mentioned once in 1006 pages.

The incident of Dr. Ivins is very troubling because he had a high-level US Government security clearance, worked within the government's secure bioterrorism research infrastructure, had privileged access to dangerous infectious materials, and was able to single-handedly conduct an attack upon the American public that resulted in five deaths and other injuries. Ivin's actions put scores of US government workers, including law enforcement, politicians postal service and others at risk, and this event cost untold millions in remediation and lost business.

Gain of Function research entails a similar risk to the public. I consider the likelihood of a researcher releasing potentially pandemic agents much higher than an armed assault upon university laboratories by terrorists or criminals, but this scenario is downplayed. Motivations could include mental illness, coercion by a foreign power, or self-aggrandizement as seemed to be the case for Ivins.

Thank you for your consideration of my comments for your meeting.

Sincerely,
Charles R. Stack, MPH
DrPH Candidate
Estelle Goldstein Memorial Scholar
UIC School of Public Health

Deputy Sector Chief, Healthcare and Public Health
FBI Infragard

From: Simon.Warne@hse.gsi.gov.uk [mailto:Simon.Warne@hse.gsi.gov.uk]
Sent: Wednesday, January 06, 2016 12:10 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Cc: m.skinner@imperial.ac.uk; Andrew.Cottam@hse.gsi.gov.uk; Michael.Paton@hse.gsi.gov.uk
Subject: FW: Risk and Benefit Analysis of Gain of Function Research undertaken by Gryphon Scientific

This is a brief response to the public consultation on the above document. I am a Specialist in Biosafety working in the UK for the Health and Safety Executive (HSE). I replying as the Secretary of the UK Scientific Advisory Committee on Genetic Modification (SACGM). Ideally I would have liked to put together a response to reflect the consolidated views of SACGM and other parts of the UK regulatory structure covering genetic modification. However, this has not been possible in the limited time available. I, therefore, hope that there will be a further opportunity to have an input as this Risk and Benefit Analysis covers an important area of science policy and the consequences of 'getting in wrong' are clearly very significant.

In the time available I have not been able to go into all the detail within the Risk and Benefit Analysis. My attention has been primarily focused on section 6 covering 'Risk Assessment of Laboratory Accidents and Natural Disasters'. In my analysis to date there one statement that has particularly caught my attention. On page 164 it is stated that 'a global pandemic caused by research on pandemic influenza viruses is expected every 560-13000 years'. I believe that as part of this exercise it is crucial that this figure is placed in some kind of context. As part of this I would draw your attention to the HSE document 'The Tolerability of Risk from Nuclear Power Stations' that is available at the following link http://www.onr.org.uk/documents/tolerability.pdf . This HSE document identifies what is seen as an acceptable risk for a major accident at a nuclear or chemical plant causing roughly 1500 casualties (see pages 31-33).

I would like to put down this e-mail as a marker that I would be interested in being informed about any further consultation on this issue. As I have said above, it is unfortunate the current consultation period of less than a month (including the Christmas break) has not provided time to prepare a more substantial response. If we were given sufficient time I would hope that the UK would be able to put together a consolidated response to represent the views of the various regulatory and policy making bodies.

Simon Warne PhD
Biotechnology Portfolio Holder / HSE Biological Agents Unit / Desk 41 5S.2 Redgrave Court / Bootle / L20 7HS / United Kingdom

Telephone +44 (0) 151 951 3335

From: Andrew Kilianski
Sent: Thursday, January 07, 2016 1:42 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Tangible translational products from GOF research

Some members of the board have asked for clarification and specific examples of basic-to-clinical research products generated from GOF research. The attached article and link below can clarify some of these questions. It was published during the RBA and might not have been available to everyone. Thanks!

Andy

Attachments

1. **Gain-of-Function Research and the Relevance to Clinical Practice -- J Infect Dis. 2015**
   http://jid.oxfordjournals.org/content/early/2015/10/27/infdis.jiv473

2. **When gain-of-function research is not "gain-of-function" research -- EMBO Rep., 2015**
   http://embor.embopress.org/content/early/2015/11/04/embr.201541617

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REVIEW ARTICLE

Gain-of-Function Research and the Relevance to Clinical Practice

Andy Kilianski,[1] Jennifer B. Nuzzo,[2] and Kayvon Modjarrad[3]

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The ongoing moratorium on gain-of-function (GOF) research with highly pathogenic avian influenza virus, severe acute respiratory syndrome coronavirus, and Middle East respiratory syndrome coronavirus has drawn attention to the current debate on these research practices and the potential benefits and risks they present. While much of the discussion has been steered by members of the microbiology and policy communities, additional input from medical practitioners will be highly valuable toward developing a broadly inclusive policy that considers the relative value and harm of GOF research. This review attempts to serve as a primer on the topic for the clinical community by providing a historical context for GOF research, summarizing concerns about its risks, and surveying the medical products that it has yielded.

Keywords. gain of function; potential pandemic pathogens; coronavirus; influenza; science policy; health policy.

Gain-of-function (GOF) research typically involves mutations that confer altered functionality of a protein or other molecule. These types of mutations have been used as powerful tools to understand basic bacterial and viral biology and pathogen-host interactions. Despite the recency of a public debate, GOF research has constituted a common, long-standing practice in the discipline of microbiology. In recent years, a public discussion has surfaced, centering on the application of GOF research to highly pathogenic and potentially lethal viruses [1]. Despite the emergence of this public dialogue, much of it has been steered by members of the microbiology and policy communities. There remains room for additional input from clinical and public health practitioners, who are often the end users of the products GOF research yields. As the results from GOF research are salient to both the improved understanding of disease pathogenesis and the development of medical countermeasures to infectious diseases, the debate over its safety and value is of direct relevance to medical and public health practitioners. This review article will provide a historical context for the current debate, describe the potential risks and benefits of this type of experimental study, and present some examples of how GOF research translates into tangible products of use to practicing clinicians.

GOF: AN HISTORICAL PERSPECTIVE

Genetic mutations can be classified in many ways, one of which is by their impact on protein function. In the simplest terms, mutations can result in a protein's loss of function or GOF. The distinction between the 2 phenotypes is not always clear. GOF research, in this context, usually results in the introduction of changes to biological agents that might increase their ability to infect a host and cause disease by enhancing their transmissibility or pathogenicity [2]. In recent years, this class of research has provoked controversy, particularly in the setting of dual use research of concern (DURC). DURC is a subset of microbiological research that, as defined by the US government, "can be reasonably anticipated to provide knowledge, information, products, or technologies that could be directly misapplied to pose a significant threat with broad potential consequences to public health and safety, agricultural crops and other plants, animals, the environment, materiel,

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or national security" [3, p. 1]. Some of the potential consequences of DURC that have been cited include the manipulation of pathogens for use as biological weapons and the development of mechanisms by which pathogens can evade countermeasures. DURC currently pertains to the select agents and toxins defined by the US Centers for Disease Prevention and Control and the US Department of Agriculture [4]. Among these pathogens, highly pathogenic avian influenza virus (HPAI) is of high concern to both public health and agriculture authorities.

Public discourse on the controversies of influenza virus research is about a decade old, beginning in 2005 with the reconstitution of the 1918 influenza A(H1N1) [5–7]. The more recent debates over the safety and merits of GOF research first surfaced in 2010, in the context of studies on the transmission dynamics of HPAI A(H5N1) (Figure 1). Laboratories at the University of Wisconsin (Madison) and Erasmus University Medical Center (EMC; Rotterdam, the Netherlands) performed a series of experiments [8, 9] that involved the mutation of 2 influenza A (H5N1) strains through multiple passaging. The two laboratories identified specific amino acid changes that enhanced airborne transmissibility of the virus between ferrets—a standard animal influenza model that exhibits a natural history and pathology similar to what is observed in humans. The potential translation from ferrets to humans raised concerns among funders (ie, the National Institutes of Health [NIH]) and the broader biosecurity policy community that the research could be used for intentionally harmful purposes or result in an accidental release of pathogens from the laboratory into the general population.

In 2011, the Department of Health and Human Services (DHHS) convened the National Science Advisory Board for Biosecurity (NSABB)—an independent federal advisory committee chartered to provide advice on the biosecurity oversight of dual use research. The NSABB was asked to weigh in on whether the GOF studies should be published in the public domain. After initial review of 2 manuscripts, one submitted to *Science* (by investigators at EMC) and the other to *Nature* (by investigators at the University of Wisconsin), the NSABB requested that study authors and the journals withhold from publication the details about the study methods [10]. Consequently, the influenza research community voluntarily implemented a year-long moratorium on GOF research. In March 2012, the NSABB recommended publication of both studies, with some minor changes to the EMC manuscript [11]. These deliberations led to the creation of a US framework for DURC studies [3, 12] and further stimulated a debate on GOF research within the scientific community [13].

Recently, influenza virus researchers laid out a rationale for GOF experiments in the context of influenza A(H7N9) [14, 15]. These arguments were met with some criticism [16–18], especially with respect to the risks of accidental or intentional release of this HPAI. Given the growing concern over this and other HPAI subtypes, the White House Office of Science and Technology Policy and the DHHS announced a moratorium, on 17 October 2014, on all new funding for GOF research on all influenza viruses, severe acute respiratory syndrome coronavirus (SARS-CoV), and Middle East respiratory syndrome coronavirus (MERS-CoV) [2]. Additionally, the US government called for a voluntary moratorium on all such research, irrespective of funding source, while the risks and benefits of such experiments could be assessed. On 15 and 16 December 2014, the National Academy of Sciences, National Research Council, and Institute of Medicine convened experts from the disciplines of infectious diseases, research ethics, and science policy to discuss the potential risks and benefits of GOF research in a public forum to help inform the federal government on how best to proceed in regulating GOF research on potentially dangerous biological agents [19]. Shortly after the meeting, the NIH notified a subset of researchers affected by the research pause that their work could resume [20]. Specifically, 5 research projects on MERS-CoV animal model development and 2 on HPAI were cleared to continue.

The discussion on the merits and risks of GOF research has not been limited to the United States, as the Dutch Court of Appeals recently handed down a verdict concerning EMC's objection to export license rules regarding the publication of HPAI GOF research [21]. Export licenses in the European Union are in place to prevent the proliferation of weapons of mass destruction and, thus, apply to specific biological agents, chemical agents, and technologies. In 2012, the Dutch government ruled that EMC had to apply for an export license to publish their GOF work, which they did to expedite publication. However, EMC later filed an objection, maintaining that GOF research in this context was for "basic scientific research." The Dutch Court of Appeals ruled that EMC had no legal standing to contest the export license regulations but did not address the legality of the export license itself, leaving the issue open for continued debate. Currently, all GOF research within the European Union requires export licenses for publication.

A deliberative review process, headed by the NSABB, is currently underway [22] to evaluate the potential impacts of GOF research and to set criteria for what types of research can be conducted and made available in the public domain. A large part of the risk analysis will likely involve the potential for these pathogens to be misused either intentionally or accidentally. Attempts have been made to anticipate the likelihood of the latter scenario, resulting in wide-ranging estimates [1, 19, 23]. The recent safety lapses at the Centers for Disease Control and Prevention and the NIH that could have resulted in exposure to anthrax and smallpox, respectively, have diminished public confidence in the ability of even high-containment laboratories to mitigate the risk of accidental release of pathogens of potential harm. Though the actual risk of accidental release of highly pathogenic viruses may be low, public tolerance of that

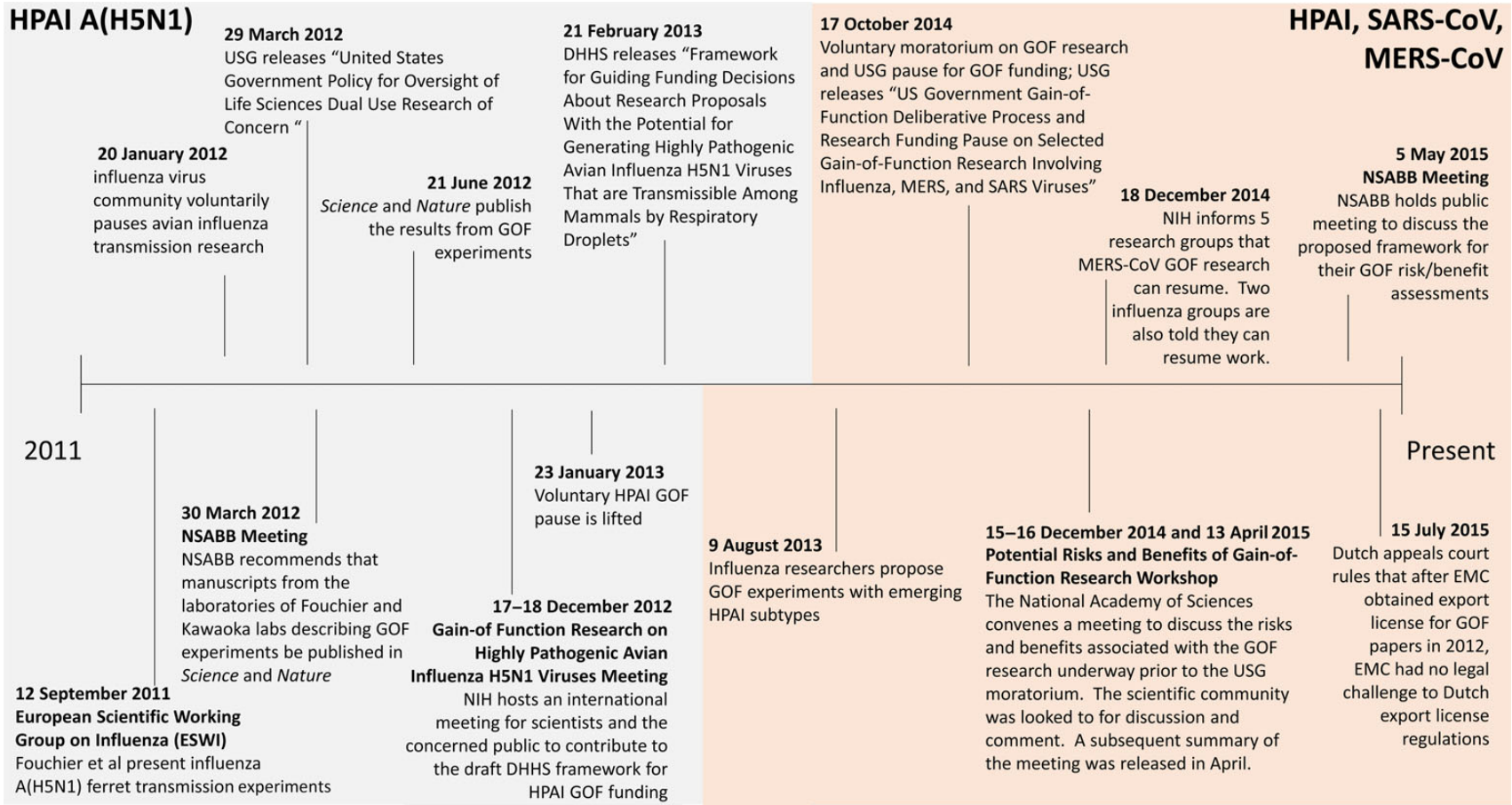

Figure 1. Historical perspective on recent debates associated with gain-of-function (GOF) research. Abbreviations: DHHS, Department of Health and Human Services; EMC, Erasmus University Medical Center; HPAI, highly pathogenic avian influenza virus; MERS-CoV, Middle East respiratory syndrome coronavirus; NIH, National Institutes of Health; NSABB, National Science Advisory Board for Biosecurity; SARS-CoV, severe acute respiratory syndrome coronavirus; USG, US government.

risk may be the ultimate determinant of what types of research are allowed to proceed.

Increasing attention has been brought to the use of alternative methods of investigation in areas that have historically been studied through GOF research. Some of the alternatives that have been proposed rely heavily on in silico technologies, such as computational modeling and disease forecasting [24–26]. The relevance of these other methods is an important consideration for the scientific community, medical practitioners, and the general public, as the risks and benefits of each approach and the tangible outcomes they yield will vary according to the interests and needs of each sector. All of these factors are being considered by the NSABB, which will decide how to proceed with the current moratorium and the future of GOF research. As the GOF debate has transpired to date, the ramifications of this research for the practicing clinician have not been made clear.

CLINICAL APPLICATIONS OF GOF RESEARCH

Animal Models

The development of novel prophylactic and therapeutic interventions invariably requires evaluation in animal models that, at least partially, recapitulate the disease in infected humans. Many emerging and reemerging zoonotic diseases lack relevant animal models that closely recapitulate human disease [27]. In these instances, GOF experiments are often needed to adapt virus isolates from humans to different, sometimes unnatural, mammalian hosts. Adaptation to a new host inherently involves the alteration of pathogens through mutation. As the development of appropriate animal models can be a rate-limiting step in the evaluation of prophylactic and therapeutic interventions, GOF modifications to viral strains can be an important tool toward accelerating the product development pipeline.

Coronaviruses such as SARS-CoV and MERS-CoV require meaningful small-animal models that elucidate viral pathogenesis and immunity. The human isolates are manipulated either through natural evolution, targeted mutation, or repeated exposure to human factors in nonhuman hosts. One of the more reliable SARS-CoV murine models was developed by modifying a human isolate through 15 serial passages, after which it was lethal to young mice [28]. This mouse-adapted virus strain contained 6 coding mutations that conferred increased virulence, approximating many features of SARS-CoV disease in humans and thus providing a robust and reproducible challenge model for testing vaccines, antivirals, and other interventions [29]. The development of an appropriate animal model for MERS-CoV, on the other hand, provides unique challenges because the viral receptor used for cell entry is radically different in mice. Models thus far have included transient transfection [30] and transgenic mice [31], although it is still unclear whether these models accurately recapitulate human infection. Approximating human disease in these small-animal models might require further passaging in the presence of a humanized receptor, thus creating a potential for the development of GOF phenotypes.

Vaccines

Many live-attenuated vaccines, including some of the most successful vaccines ever developed, have been generated through GOF research. From polio to smallpox to influenza, live-attenuated vaccines elicit immunity against authentic epitopes on whole pathogens without causing disease. The live-attenuated measles vaccine was created by passaging the virus until mutations arose that altered virus tropism—a technique that could be considered, by current definitions, GOF research [32]. New research on highly pathogenic viruses has emphasized the different ways GOF mutations can generate even-more-effective live-attenuated vaccines. Mutations within RNA virus polymerases, for example, modify replication fidelity to generate higher or lower mutation rates during viral replication. These fidelity mutants could potentially alter viral tropism, modify key antigens, and increase resistance against novel therapeutic interventions or antibody responses, but they could also lead to a virus that is less fit [33, 34]. These particular types of experiments have been carried out on a range of viruses, including alphaviruses [35, 36] and picornaviruses [37]. The introduction of GOF mutations not only attenuates the virus but also provides improved understanding of the mechanics of viral replication, thus potentially uncovering new strategies in the development of vaccines against emerging pathogens.

Therapeutic Interventions

The generation of escape mutants in the presence of an investigational agent is common practice for the evaluation of antibiotics, antivirals, and other monoclonal antibodies. GOF experiments with HPAIs and highly pathogenic human influenza viruses, for example, have identified specific mutations that can confer multidrug resistance [38, 39]. GOF experiments are necessary in this context because naturally occurring resistant strains may not yet exist or the complex background of naturally occurring mutations may preclude identification of the amino acid residues that are critical to resistance [40]. These GOF studies are equally important in research on antivirals and antibiotics and can help inform the development of combination therapies. Passive immunotherapy, which often includes a combination of products, is particularly dependent on GOF experiments for evaluating efficacy [41–43], as seen in the current Ebola outbreak that has prompted a robust program to evaluate combination monoclonal antibody therapies [44, 45].

Disease Surveillance

In the past half-century, GOF research has contributed to an improved understanding of the epidemiology of emerging pathogens and has informed efforts to conduct surveillance for

future outbreaks. In the context of influenza, data, derived from GOF research, on the relative transmissibility of hemagglutinin mutations has aided in the interpretation of molecular surveillance data [46]. Specifically, the initial influenza A(H5N1) [8, 9] and later influenza A(H7N9) experiments identified amino acid changes in influenza virus hemagglutinin or RNA polymerase through viral passaging or site-directed mutagenesis. This research elucidated mechanisms by which naturally occurring influenza virus strains might evolve to replicate more efficiently and transmit more easily within mammalian hosts [47, 48]. The results of these experiments can be used to cross-reference traits found among circulating strains and help predict transmission patterns and pathogenicity [49]. As the field of disease surveillance evolves to accommodate a growing repository of viral sequences, GOF research will also play an important role in assessing the public health significance of genotypic variation. Though current understanding of the relationship between genotypic data and phenotypic expression is suboptimal, the increasing reliance by the clinical community on molecular diagnostic tools may help to reduce that uncertainty. As costs of whole-genome sequencing continue to decrease, data from these techniques are likely to become more central to disease surveillance programs. The results of GOF experimentation can also help inform decisions about countermeasure selection and stockpiling, particularly in the context of influenza surveillance programs [50]. The improved understanding of how HPAIs evolve to transmit more efficiently has also factored into decisions about the creation of prepandemic vaccine stockpiles.

THE ROLE OF CLINICIANS IN THE GOF RESEARCH DEBATE

The world has been witness to a number of emerging infectious disease pandemics over the past several decades. Each time, clinical and public health practitioners were on the front lines, providing care and treatment and finding ways to interrupt transmission, and were ultimately responsible for containing the outbreak. Healthcare providers require effective medical countermeasures and epidemiologic information to assess risk and support decisions about treatment and prevention. Recent outbreaks of infection due to Ebola virus, MERS-CoV, and pandemic influenza virus, however, continue to demonstrate that medical and public health readiness for emerging infections is not always optimal and could benefit from more research and development. As outlined above, GOF research plays a significant role in ensuring that clinicians have the tools they need to respond to infectious disease outbreaks. Therefore, the clinical community is directly affected by policy decisions on what types of research are and are not is allowed to continue. There are also risks associated with GOF research, of which the clinical community will have to be acutely aware. As recent lapses at high-profile laboratories have illustrated, there remains the potential that bacterial and viral strains can escape even the most secure environments. Should a pathogen escape, whether it is naturally occurring or the product of GOF research, the clinical community will have an important role in detecting and responding to such incidents. Because of their unique role as both beneficiaries of the products of GOF research and mitigators of its risks, clinicians have a vital stake in the public debate on how GOF research should proceed.

Notes

Acknowledgment. Information in this report is cleared for public release, and distribution is unlimited.

Disclaimer. The conclusions and opinions presented here are those of the authors and are not the official policy of the National Research Council, the Defense Threat Reduction Agency (DTRA), the US Army, the Edgewood Chemical Biological Center, or the US government. The authors declare no conflicts of interest.

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Potential conflicts of interest. All authors: No reported conflicts.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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Andy Kilianski, Ph.D.
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EMBO reports

Opinion

When gain-of-function research is not "gain-of-function" research

Andy Kilianski[1] & Randall S Murch[2,3,4]

There is ongoing discussion among the scientific and biosecurity communities over how to address concerns about "gain-of-function" (GOF) research using highly pathogenic agents [1–3]. The discussion has mainly centered on previous work by Yoshihiro Kawaoka's group at the University of Madison-Wisconsin in the USA [4] and Ron Fouchier's group at Rotterdam University in the Netherlands [5]. Both groups introduced mutations into highly pathogenic H5N1 avian influenza (HPAI) that could potentially increase human-to-human transmission of the virus. These mutations are classified as GOF because they increase airborne transmissibility in ferrets—a good model for human transmission. Some in the research and biosecurity communities are concerned that these experiments could result in accidental or intentional releases of the mutated pathogen, or that the now publicly available information about how to increase the human-to-human transmissibility of H5N1 influenza could be abused for developing biological weapons [6,7].

Earlier this year, Kawaoka's group again published the results of GOF research on the PR8 influenza backbone in which they created a high-yield vaccine strain capable of hosting multiple HA/NA antigenic combinations [8]. The high-yield phenotype was observed in diverse host cells in addition to chicken embryos, which are used for influenza vaccine production. This is a potentially major breakthrough for vaccine development and production, as it would greatly reduce the time and cost of rapidly producing influenza vaccines in response to disease surveillance and prediction, as well as to emergent pandemic strains. Nonetheless, and despite the obvious scientific and commercial value of this research, the decision whether to publish GOF-related research such as this, especially in human pathogens like influenza, is not straightforward.

The research performed by the Kawaoka group—which was finished before the current moratorium on GOF research in the USA came into place—resulted in a GOF phenotype. This work would have fallen under the current moratorium [9], but should not be classified as GOF research in our view. It is unlikely that the release of these high-yield strains from the laboratory would have any negative effect on human health because these are vaccine strains of influenza. Neither is this a case of dual-use research of concern (DURC) because the information in the paper has little potential to be applied to pathogenic strains of influenza. The mutations described are unlikely to be broadly applicable to other influenza subtypes or strains: growth-enhancing mutations from other influenza backbones did not necessarily confer a high-yield phenotype in the PR8 backbone. The decision to categorize this work as GOF—meaning that it falls under the current moratorium that has halted such research in the USA—was because of the previous experiments to increase transmissibility of avian H5N1 and HPAI's designation as a "Pathogen with Pandemic Potential (PPP)".

This example illustrates why we need a more appropriately structured classification system of GOF research with sufficient fidelity to consider individual pathogen strains and their features, instead of merely the pathogen being used. As demonstrated by the lack of HPAI human pandemics—and the emergence of other known and unknown pathogens causing severe disease—singling out pathogens as having "pandemic potential" without sufficient supporting evidence is scientifically problematic. Furthermore, determining the "pandemic potential" of pathogens is sometimes only possible with GOF research. For the infectious disease community, the only way to proactively prepare for the next pandemic is to clearly define what constitutes a GOF and/or DURC in a way that is not wholly defined just by the pathogen. While the NIH and National Science Advisory Board for Biosecurity (NSABB) are reviewing the risks and benefits of GOF research, a clearer and more effective definition of what constitutes GOF research—one which circumscribes all infectious disease agents and not just a select list—should be established. The community needs to build this consensus to be able to safely continue GOF research and responsibly keep these experiments in the traditional antibiotic, antiviral, and vaccine development methodology.

The scientific community has always had a great interest in openly and accurately disseminating knowledge, which is now becoming possible with the advent of open access publications and other web-based tools; the research to increase the yield of the PR8 influenza backbone was in fact published in an open access journal. The proliferation of open access journals, preprint servers, and posting of scientific research on the internet is inherently good for science as a whole. However, it provides multiple challenges for DURC and GOF

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DOI 10.15252/embr.201541617

research to prevent their dissemination without proper review and management. It is clearly not sufficient to simply perform DURC reviews at the editorial level prior to publication in peer-reviewed journals because, in today's publication landscape, it is possible to publish work without review on pre-print servers or open-review journals. To better evaluate DURC and GOF research as a whole, a more comprehensive "systems" construct is needed. The review process should be initiated earlier, at the proposal step at the funding agency. In addition, it may require regular monitoring after the initial review to avoid "surprises", as occurred with Kawaoka's and Fouchier's original papers.

As the NIH and NSABB determine a course forward how "gain-of-function" research should be evaluated in the USA in the future, it needs to flesh out guidelines that list which pathogens and experiments require review and that standardize the review process itself. We suggest that the review and reporting should encompass the most critical phases of research from the proposal to the publications stage. Draft guidelines should be made available for public comment with meaningful responses considered for incorporation, published, and then formally reviewed on a regular basis and modified if required. These reviewing and reporting structures should be exercised prior to the formal requirement, with participation from outside actors and full transparency.

US government-funded research proposals should require a consistent, comprehensive DURC review prior to funding and to the initiation of the research, and not only at the level of the institution (which has recently been recently enacted [10]) and the publication stage. This review process should be consistent across agencies. A common set of standards and guidelines should guide the review procedures of US public funding entities to determine whether research proposals present GOF and DURC concerns. Such a process will ensure that the research being funded has been cleared of these issues, and any potential dissemination of this work has been vetted. Similar to the definition of GOF research, the NIH and NSABB should establish how this work is to be reviewed, not simply whether the work has tangible merits.

The international scientific community, governments, private funders, overseers, regulators, publishers, and stakeholders should consider designing, testing, implementing, and embracing a consistent end-to-end protocol which promotes safe and valuable research while minimizing uncertainties and risks, including the misuse of science. We recognize that this is not an easy achievement to attain, but we believe that it will be worth the investment and effort and will help to prevent future funding moratoriums being placed on the GOF and DURC research communities.

Conflict of interest

R.S.M. was a former member of the NSABB from December 2009 to April 2012. The conclusions and opinions presented here are those of the authors and are not the official policy of the National Research Council, DTRA, the US Army, ECBC, or the US Government. Information in this report is cleared for public release, and distribution is unlimited.

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From: David Wolinsky []
Sent: Sunday, January 17, 2016 12:21 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: no pain, no gain of function

Please: NO gain of function research or development on pathogens. In practice, "defensive" capacities will remain limited, while offensive uses will appear as they always do. Meanwhile the risks of accident should be clearly -- for theses cases--unacceptable.

--

David Wolinsky

Frederick, MD

From: Francy Williams []
Sent: Tuesday, January 19, 2016 10:03 PM
To: Viggiani, Christopher (NIH/OD) [E] <christopher.viggiani@nih.gov>
Cc: Beth Willis
Subject: GOF Research - concern comment

Comment: Frances Williams RN MS (retired and living in Frederick MD - the location of BSL-3 labs), private citizen and member of the Religious Society of Friends (Quakers)

I am writing in response to a request for public comment regarding the upcoming NAS symposium March 10-11 2016.

With the story of Flint Michigan's polluted water (an unintended consequence of the city's attempt to save money) in the headlines,

I reflect on the possibility of unintended consequences from Gain of Function research gone awry in our community here in Frederick.

My prayer is that science be conducted for the highest good and that we not fall prey to events that occur as the result of conflicts of interest, or as the result of nefarious intentions.

I hold the vision that someday non-violence will become the American Way and resources will no longer be used to support tools designed to destroy life.

Gain of Function (gain of function of microrganisms for the purpose of eliminating humans). Conducting research on pathogens to make them more virulent, transmissible, and resistent to treatment, in my opinion should be illegal.

I endorse the commments made by Beth Willis at the workshop held at NIH on Jan. 7-8 2016.

I send hope that good minds and hearts will develop measures to assure safety for all.

Respectfully,

Frances Williams

(**Note:** The comments delivered by Beth Willis, a panelist at the Jan. 7-8 NSABB meeting, referenced above were copied in the original email but omitted when comments were compiled. Ms. Willis' comments were previously conveyed to the NSABB and can be found as part of the Session IV presentations archived on the Jan 7-8 NSABB meeting webpage.)

From: ROLAN.CLARK@comcast.net []
Sent: Saturday, January 23, 2016 9:05 AM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>; GOF <GOF@nas.edu>; Willis, Beth
Subject: comments nsabb gof

23 January 2016

To: NSABB (National Science Advisory Board for Biosecurity)
From: Rolan O. Clark
Subject: Bio Labs, Gain of Function comments

Dear NSABB and to whom it may concern.

Experience

U.S. Navy 1957-63, 4 years active, 2 years inactive reserve. 6 months ETA Radar school, Treasure Island California, 2 years, 9 months U.S.S. Estes AGC-12 radar tech

15 years calibrate and repair of electronic test equipment to Bureau of Standards specs, now NIST, then 10 years two way radio systems repair, 20 years R&D space tubes/technology and space battery testing and writing computer programs to control data collection equipment, collect and display test data and help write reports.

My lack of formal higher education has not changed the laws of physics nor the meaning of words.

Testing Philosophy and Data Collection

Science is first philosophical, then anecdotal, then data collection and knowing the limits, application, specifications and accuracy of data collection equipment/procedures , the first steps in collecting believable data.

Next, presentation of data in proper reference to properties of test under consideration, such as what is listed/tabled/plotted/displayed against what. There are lies, damn lies and data. Science is only as good as the next peer review.

Gain of Function

Definition. A type of mutation in which the altered gene product
possesses a new molecular function or a new pattern of gene expression.
Gain-of-function mutations are almost always Dominant or Semidominant.
See also: Amorphic Mutation.

The concept of Gain of Function as I try to understand it, by watching the archived video of the NIH meetings, downloading and reading 5 plus web sites dealing with Gain of Function and reading linked comments by scientists, leads me to come to the conclusion that trying to apply Gain of Function to Bio Labs research with its inherent lack of means to detect non recognized/unanticipated

mutations/variations, along with expected results, is not a reliable research methodology while at the same time recognizing at times the possible need for research into to the unknown but possibly Gain of Function as it is used in Bio Labs research should not be considered as an accepted research tool, rather a definition.

I don't know if Gain of Function is supposed to be a guide to amplify or exaggerate disease transmission/reaction characteristics and then see what will mitigate the result but if some method is found to mitigate these amplified or exaggerated results that in itself may not be an indicator that the original disease will react favorably to what may have been a favorable reaction to amplified or exaggerated conditions. But what I referenced in this paragraph may not be a purpose of Gain of Function.

Why would one try to develop more efficient methods of transmitting diseases or make diseases more virulent when there may be no way to mitigate or detect all variations of these 'developments' of such dangerous measures and how can these new mutations be considered typical or representative of diseases being researched.

http://www.livescience.com/53410-stephen-hawking-warns-of-planetary-doom.html?cmpid=NL_LS_weekly_2016-1-19 , it is interesting to note the concern by Mr. Hawking.

The below, between the ********** are from the above URL
****
Stephen Hawking has once again warned that humanity could wipe
itself out before it has a chance to establish far-flung space colonies.
At a recent talk in England, the famed physicist singled out nuclear war,
genetically engineered viruses and global warming as likely culprits.

****

Ferrets

Googling why ferrets were the animal of choice for some studies on spread of diseases I found that ferrets sneeze about as often as humans and putting 'infected' ferrets up stream in a controlled air flow environment simulated studying disease spread by the sneeze route down the air stream. The comments in the article weren't too exciting about the controls of this type of test but I got my ferret use question answered.

Programming, Computers, DNA/RNA

Re:
http://www.ncbi.nlm.nih.gov/books/NBK26887/
I don't understand the info in the immediate above URL.

If a computer program does not 'work' one has to know the source code and programming and the operating system to analyze the problem if a resolution to the problem is not found using other methods.

Gain of Function, or research, as it relates to the topic of diseases requires knowledge of how DNA/RNA 'source code' signals/triggers molecular changes in the disease and other molecules probably not

possible at the present level of research therefore one is relegated to observing results of tests and drawing conclusions based on how data and data collecting procedures function all the while possibly blinded to other DNA /RNA reactions/instructions and results because other detection mechanism for other mutations present in bio mutations work may not be available unlike other spectrum identifying devices such as spectrum analyzers for rf energy and mass spectrometers for molecular activity identification. I believe at present there is no DNA/RNA 'spectrum' identifier to detect unwanted or unanticipated results.
I assume DNA RNA react upon contact with other molecules and the molecular/chemical reactions simulate 'instructions' as per the chemicals in the DNA , RNA and molecules, whether from 'normal' cells or pathogens, bacteria or viruses or anything in the body. I would assume blood flow is the distribution method for these bio entities to make contact with each other.

Patterns

It is my belief that patterning is a very useful tool, if data follows a pattern it denotes some consistency. I believe patterns can be a very useful security tool if used with a computer and possibly this example: use a computer to sample and log all air pressures in labs and entrances, hall way pressure, transition room pressure, lab pressure along with possibly iris and fingerprint info and clock times.

If all data mentioned above is plotted over time then any deviation from 'normal' should immediately signal an alarm. This may also require input from behavioral experts to develop sign in routines to enhance security. As perfection is the enemy of progress too much routine in sign in procedures leads to laxness in security and this pattern can also be put in a computer display to detect any changes in patterns.

Regulations

I have written to our local government entities that the only thing worse than regulations/laws/codes in a democracy is no regulations/laws/codes. Single Source Federal oversight consisting of Regulations/laws/codes are needed for all the biolabs for a one voice oversight function regardless of the inconveniences regulations/laws/codes may bring. We have seen recent failures in governments oversight function as in the Flint Michigan water issue but all proper oversight functions in a properly functioning government rests solely on the integrity of the persons responsible for administrating the rules assuming the guidelines are in place and correct.

Bio Labs not in Residential Areas

Bio Labs should not be in residential areas for at least 2 major reasons:
1. Terrorist's thrive on publicity and a terrorist attack on a biolab in a residential/inhabited would be more desirable to a terrorists goal as compared to a biolab apart from residential/inhabited areas. areas.
2. If a bio lab was 5 miles from a residential/inhabited area and an aerosol type escape of test pathogens occurred there would be dispersion plus the time to react. Distance is time in an aerosol environment plus determining the time the escape happened may be very difficult to determine and time is important. There is also the possibility that the escaped pathogen may be rendered moot in the environment of open air and sunlight plus the concentration would probably continuously decrease, say particles per unit volume.

Single Source Bio Labs Oversight Entity

The last 2 or 3 years I have written my U.S. Senators and U.S. Representative and our local State Delegation about the need for a Single Source Bio Labs oversight entity consisting of a single Federal Department for oversight of for ALL biolevel labs and the need and right for the public to know where these labs are.

Not only would there be one voice but there would be defined word/words to describe each issue. Words are important. There would be the advantage that when any issue needs to be addressed all labs could be notified at the same time by the internet, for example, using words defined and understood by all entities.

Communications

Communications and speed of communications is extremely important in emergency conditions. A single source method to distribute and communicate using accepted and approved procedures/wording would be very beneficial.

Conclusions

I believe there should be one Federal oversight entity for all Bio labs with accepted procedures, wording and communications paths.

Gain of Function seems to be a definition instead of a research procedure. I believe that Gain of Function is a very narrow definition, though references a very complicated process, of a type of research.

There is nothing wrong with not knowing, there is something wrong with not asking.

Respectfully submitted as my concerns and what I think I understand, however limited, about a very complicated process.

Rolan O. Clark

From: Beth Willis []
Sent: Tuesday, January 26, 2016 6:21 PM
To: Viggiani, Christopher (NIH/OD) [E] <christopher.viggiani@nih.gov>
Subject: Concepts to inform decision-making about risk: Setting Specfic Safety Goals

Hi Chris,

I hope you've survived the last week safe and warm. As have I.

A colleague asked that I pass the following on to the NSABB and the NAS.

As you may know, DOE and the NRC have established specific public safety goals and mechanisms to determine what public risk is considered acceptable. The concepts in these materials might help to inform a similar effort for GOF Research of Concern, DURC, Select Agent and other risky biological research. I don't believe such safety goals currently exist.

DOE Nuclear Policy Safety and Goals: http://energy.gov/sites/prod/files/2013/12/f5/DOE_P420-1_Final_2-8-11.pdf

And

http://energy.gov/sites/prod/files/2013/12/f5/Technical_Basis_for_DOE_P_420-1.pdf

The Nuclear Regulatory Commission has a similar set of goals:

http://nuclearsafety.gc.ca/eng/pdfs/Presentations/Guest-Speakers/2014/2014-01-13-Safety-Goals-and-Risk-Informed-Regulation-at-the-US-NRC.pdf

with best regards,

Beth Willis
Frederick Citizens for Bio-lab Safety

From: Kim Loll []
Sent: Saturday, February 06, 2016 12:37 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Comments from the Containment Laboratories Community Advisory Committee (CLCAC) Frederick, Maryland

I am writing on behalf of the Containment Laboratories Community Advisory Committee (CLCAC) of Frederick, Maryland. The CLCAC was formed as joint committee sponsored by both the City of Frederick, as well as Frederick County, MD.

The purpose of the Committee is to:

- Foster two-way communication between the public and the operators of the high containment laboratories operating at Fort Detrick and elsewhere in Frederick County.
- Seek information about public concerns and ways to address those concerns.
- Advise and make recommendations on behalf of the public to government, containment laboratory and Fort Detrick officials regarding opportunities to improve any laboratory-related operational matters that may potentially impact public safety and health.

The CLCAC has been following the many issues related to the current discussion on Gain of Function research over the last several years. Several members of CLCAC attended the January 7/8, 2016 NSABB Meeting or observed the webcast, and the past Chair of CLCAC, Ms. Beth Willis, was a panel member on the Workshop. The CLCAC would like to take this opportunity to endorse the following papers and presentations provided at the NSABB Meeting:

- Presentation and written comments provided by Ms. Beth Willis
- Presentation and written comments provided by Dr. Marc Lipsitch

Thank you for the opportunity to participate in this important deliberative process. We look forward to future opportunities to provide additional public perspective on biosafety and biosecurity policy issues as they relate to public health concerns.

Local newspaper coverage of the January 13, 2016 CLCAC meeting following the NSABB Meeting can be found at:

http://www.fredericknewspost.com/news/health/treatment_and_diseases/frederick-committee-addresses-pathogen-research-debate/article_10e694b8-2a98-59ba-85dd-674337980152.html

Additional information about the CLCAC and its activities can be found at:

https://www.cityoffrederick.com/index.aspx?nid=127

Sincerely,

Kim R. Loll, Vice-Chair
Containment Laboratories Community Advisory Committee

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February 23, 2016

[Submitted electronically to nsabb@od.nih.gov]

Samuel L. Stanley, MD
Chairman of the NSABB
Office of Science Policy
National Institutes of Health

IDSA Comments to the NSABB Working Paper on Evaluating the Risks and Benefits of Gain-of-Function Studies to Formulate Policy Recommendations

Dear Dr. Stanley,

The Infectious Diseases Society of America (IDSA) has closely followed the National Science Advisory Board for Biosecurity (NSABB) as it develops formal recommendations on how to assess the risks and benefits of gain-of-function (GOF) research of concern on pathogens with pandemic potential. IDSA members will be among the first responders to care for affected individuals in any disease outbreak, and will also lead research efforts to counter these disease threats. Accordingly, they are well positioned to understand the risks and benefits of these potentially dangerous experiments. Last summer, our society submitted recommendations for the NSABB as it worked with its contractor, Gryphon Scientific, to undertake a risk-benefit assessment (RBA) of the paused GOF research projects of concern, and then release its initial findings and recommendations.

IDSA has limited our comments today to those that apply to the NSABB's working paper, as it will shape the U.S. Government (USG) policy on the oversight of GOF research of concern. We applaud the NSABB's efforts to address IDSA's recommendations in the working paper, including its focus back to only the research of highest concern and its exclusion of seasonal influenza vaccine manufacturing and development. On the other hand, we are unified in our conclusion that the NSABB's draft findings and recommendations will not provide the appropriate guidance needed to develop a streamlined mechanism that provides appropriate oversight of the risk and benefits of GOF research of concern.

Below, IDSA offers specific recommendations to improve the areas of the working paper of greatest concern:

1. **Remove resistance to public health control measures as an attribute of GOF studies of concern**

IDSA strongly supports the NSABB's "key finding 1," that only a small subset of GOF research has risk that warrants an additional level of oversight." As IDSA stated in its earlier comments, a narrow focus only on GOF research of concern will

avoid an inadvertent regulatory capture of low risk research, which was not mentioned in the original White House description of research to be included in this deliberative process.

Consequently, IDSA believes the NSABB's proposed scope of GOF of concern, research that generates a pathogen that is highly transmissible, highly virulent, and resistant to public health control measures, may be unduly narrow. The limitations set forth on research in the NSABB document may fail to identify any GOF research for review and regulatory oversight, notably the types of experiments that sparked our current deliberation over the risk of GOF of research on pathogens with pandemic potential. Moreover both Gryphon Scientific and a number of panelist speakers at the January NSABB meeting concluded that public health control measures would have little ability to control a widespread outbreak of a highly virulent and transmissible pathogen. As stated in our earlier comments, IDSA again recommends that the NSABB focus oversight on GOF research that would be anticipated to combine both high pathogenicity and transmissibility in a pathogen; while escape from medical countermeasures is a concern, it is secondary to the above characteristics. This definition would capture the GOF experiments of greatest concern, and ensure that they are reviewed appropriately to assess their risk and benefits.

2. Exempt routine, responsible vaccine manufacturing from GOF oversight

The NSABB explicitly identifies the development and manufacture of seasonal influenza vaccines as not GOF research of concern. IDSA strongly agrees with this conclusion, understanding the critical importance of adapting and manipulating wild type influenza virus for improved growth in eggs and mammalian cell lines for vaccine manufacturing. However, our society believes that this explicit exclusion can be expanded to include all routine, responsible vaccine manufacturing activities. For example, the development of pre-pandemic and pandemic influenza vaccines uses standard methods and safety procedures that are widespread in the field. IDSA affirms that these routine activities pose little risk to the public, and play a critical role in public health preparedness.

3. Institute an independent standing board to review GOF of concern

The NSABB working paper concludes that "the U.S. government has effective policy frameworks in place for managing risks associated with life sciences research." IDSA strongly disagrees that the current policy frameworks, the USG Policy for Federal Oversight of DURC and the Department of Health and Human Services (HHS) GOF framework for H7N9 and H5N1 influenza, are sufficient to oversee GOF research of concern. For example, the USG DURC policy requires institutions to provide initial oversight of a GOF research project. As raised on several occasions by panelists at the January NSABB meeting, institutional biosafety committees (IBCs) vary widely in their expertise on assessing GOF research and lack transparent, easily accessible guidance to aid in these efforts. Often GOF research may reach a final line of review during submission for publication, where journal editors must take on the task of assessing the risk of publishing the findings; again they lack accessible guidance to ensure they provide appropriate review. In addition, the multiple frameworks of oversight for DURC, select agent research, recombinant DNA research, research that poses biosafety risks to human health or agriculture, research activities involving the shipment or export of infectious agents, and GOF research of concern create an often confusing regulatory environment that can impede scientific research, public health responses, and product development that are in the public interest.

Instead of building upon current oversight efforts, IDSA recommends the NSABB examine the formation of a standing advisory board for GOF research of concern. This board should be independent of GOF funding bodies and of those units within the government that may perform GOF research of concern, and could review GOF research of concern while also providing advice to investigators, IBCs, and journal editors. IDSA believes this board should include stakeholders with expertise in biosecurity, public health, and other relevant perspectives, and also have full access to the security information needed to appropriately assess GOF research. Given the security risks of the GOF research reviewed, it is likely that much of this board's activities may not be made publically available. Therefore, it is critical that the review process itself be as transparent as possible, with aspects that do not involve biosecurity being open to the public. While IDSA proposes that this board initially focus only on GOF research of concern, we do believe it could provide the template -or be expanded in scope-to replace current oversight frameworks in providing a streamlined and appropriate oversight of all DURC.

4. Develop recommendations to address biosecurity information risks

IDSA has noted that the NSABB working paper largely accepts Gryphon Scientific's conclusion that the information risk of GOF research of concern was minimal, stating that "most of the information of interest is already published, or non-GOF information relating to pathogens that are more attractive agents of harm is already available." IDSA asserts that while current GOF research information is already publically available, it is almost certain new research approaches, sequence information, and other data will be generated in the future that would pose novel, additional biosecurity information risks. IDSA strongly recommends that the NSABB reassess these risks, and either develop new recommendations that appropriately address them, and/or request input from other external science advisory groups that currently serve the Intelligence Community, with expertise in the life sciences and access to relevant classified information.

5. Strengthen working relationships with international GOF stakeholders

While the NSABB working report discusses the importance of global engagement and how U.S. policy will likely impact other global efforts, it does not make any specific recommendations on how to better engage international GOF stakeholders. IDSA understands that GOF research is proceeding in a relatively unimpeded manner in many countries outside of the US, but strongly believes that any USG activity would likely play a key role in the establishment of any international consensus on GOF oversight. We urge the NSABB to consider recommendations on how the USG can build strong working relationships with the international GOF stakeholder community. A robust global dialogue would allow the USG to observe the effectiveness of other GOF oversight efforts to better inform domestic USG policy; these stronger relationships will also be critical in making any progress towards international GOF oversight.

IDSA remains committed to ensuring that the broader scientific and science policy communities participates in efforts to guide GOF research appropriately. We hope the March National Academies of Science meeting on the NSABB's draft recommendations will include the perspectives of scientists, healthcare workers, policy-makers, ethicists, and representatives from the public that our society believes are critical in developing an appropriate oversight of GOF research of concern.

4: *IDSA comments on the NSABB draft GOF recommendations*

IDSA thanks the NSABB for this opportunity to comment, and looks forward to continuing to work with the U.S. Government and those who advise it to clarify the decision-making process on how and whether to undertake high-risk life science experiments. Should you have any questions or concerns about these comments, please feel free to contact Greg Frank, PhD, IDSA Program Officer for Science and Research Policy, at gfrank@idsociety.org or 703-299-1216.

Sincerely,

Johan S. Bakken, MD, PhD, FIDSA
IDSA President

About IDSA

IDSA represents over 10,000 infectious diseases physicians and scientists devoted to patient care, disease prevention, public health, education, and research in the area of infectious diseases. Our members care for patients of all ages with serious infections, including meningitis, pneumonia, tuberculosis, HIV/AIDS, antibiotic-resistant bacterial infections such as those caused by methicillin-resistant *Staphylococcus aureus* (MRSA) vancomycin-resistant enterococci (VRE), and Gram-negative bacterial infections such as *Acinetobacter baumannii, Klebsiella pneumonia*e, and *Pseudomonas aeruginosa*, and, finally, emerging infectious syndromes such as Ebola virus fever, enterovirus D68 infection, Zika virus disease, Middle East Respiratory Syndrome Coronavirus (MERS-CoV), and infections caused by bacteria containing the New Delhi metallo-beta-lactamase (NDM) enzyme that makes them resistant to a broad range of antibacterial drugs.

From: Lynn Klotz [mailto:lynnklotz@live.com]
Sent: Tuesday, February 23, 2016 6:45 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>; GOF@nas.edu
Subject: Commentary on Gryphon RBA to the NSABB and NAS

Dear NSABB and NAS,

The attached Commentary shows that an absolute probability of escape from a lab of an mammalian transmissible HPAI may be calculated, contrary to Gryphon's claim. I also do the calculation.

Lynn Klotz, PhD
Senior Science Fellow
Center for Arms Control and Non-proliferation

>>

To: National Science Advisory Board for Biosecurity (nsabb@od.nih.gov)
National Academy of Sciences (GOF@nas.edu)

From: Lynn C. Klotz, Ph.D.
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Date: February 23, 2016

Commentary for the March NAS meeting on GOF: Toward absolute probabilities for escape from a laboratory

Summary and conclusion

This Commentary presents a calculation of "direct" or "absolute" probability[1] of escape from a laboratory of a potential pandemic pathogen, specifically mammalian-airborne-transmissible, highly-pathogenic avian influenza viruses (matHPAI). Absolute probabilities are necessary to calculate the probability of a laboratory escape and subsequently the likelihood of a pandemic from an escape, a key goal of Gryphon Scientific's risk-benefit (RBA) analysis.

Gryphon employed a relative probability approach that in the end failed to arrive at an absolute probability of an escape. Thus, this key part of their analysis ended up where it started, not accomplishing its goal of estimating the risk of the research (risk = likelihood x consequence). Gryphon acknowledges this failure.

Here, I will argue that Gryphon went down a wrong path by pursuing a relative probability approach. I will further show that it is possible to estimate absolute probability of escape by actually carrying out the calculation using laboratory incident data reported under the NIH reporting guidelines for BSL3 or BSL4 laboratories. Since all steps of my analysis are explicit and transparent to the reader, it provides a basis for focused discussion and assessment of each step.

In comparison, Gryphon's analysis does not explicitly provide the exact data employed or direct references to it, and Gryphon often provides little detail of the steps in its various analyses. This lack of transparency makes it difficult to verify Gryphon's conclusions. Furthermore, Gryphon fails to define the meanings of or labels for various variables. For instance, if they report a value for a lab-related accident probability, they fail to say if the probability represents one lab for one year, one lab for many years, etc. This failure to define precisely key variables adds to the lack of transparency and the ability to assess their RBA.

My analysis concludes that the probability of escape and likelihood of a potential pandemic is much too high, with an expected "fatality burden" of 512 fatalities per year for each lab conducting this research. To put this fatality burden in perspective, no Institutional Review Board tasked with assessing human subject research would approve a proposed research project with an expected 512 fatalities per year.

Dr. Marc Lipsitch, in his presentation at the January 2016 National Science Advisory Board for Biosecurity (NSABB) meeting, described published research to understand how HPAI may become airborne transmissible in humans that does not require live matHPAI viruses. Many mutations that contribute to airborne transmission have already been identified by this research without employing live virus. Thus, there is little to be lost by banning the live virus research.

I conclude that NIH should not fund this specific matHPAI research and should also not fund any other research with comparable risk. Since the NSABB mandate is very narrow, only whether NIH should fund the research, the NSABB should strongly recommend that the U.S. ban the research regardless of funding source, and recommend that the State Department make a serious effort at an international agreement to ban the research.

Two approaches for estimating absolute probabilities of a lab escape and subsequent pandemic

To estimate the likelihood (probability) of a pandemic beginning with a laboratory escape of a matHPAI, there are two general approaches:

(1) A "bottom-up" approach where probabilities are obtained for significant mechanical/equipment failures or for human error that can lead to laboratory acquired infections (LAIs) and other escape paths into the community. Then, add them all up. This appears to be Gryphon's approach. The approach here is bottom-up as well, but it starts with laboratory incident data reported under the NIH reporting guidelines for BSL3 or BSL4 laboratories, a starting point and path forward different from Gryphon's.

(2) The "top-down" or "real-data" approach. A number of us have been arguing that Gryphon should have taken into account real data as well (for instance, the probability of escape into the community of undetected or unreported LAIs calculated from the 2013 CDC report). Gryphon's valid criticism of the CDC data is that the LAIs were for bacterial pathogens, and certainly not for matHPAI viruses.

Gryphon could have carried out a "control" calculation to demonstrate that its approach can produce probabilities of escape through LAIs comparable to those calculated from the 2013 CDC data. If the two calculations end up with greater than one or two orders-of-magnitude difference, there is a problem with their data used in the bottom-up approach. In a conversation with Gryphon's Managing Director, Rocco Casagrande, he pointed out the data they have collected is not relevant to bacterial select agents, so the control calculation could not be done. But they could and should have collected the missing data as part of their risk-benefit analysis (RBA) to gain confidence in their bottom-up approach data.

In its RBA, Gryphon notes that human error far exceeds mechanical failure. This is borne out by NIH reported incident data (see below) and by the highly publicized recent incidents of human errors leading to escapes into the community.

It is a hypothesis of this Commentary that likelihood of human error will be similar in laboratories researching matHPAI and in laboratories researching other less dangerous select agents. A further hypothesis is that absolute probabilities of escape can be estimated from data already publically available and can be supplemented by data gathered easily. This is a more useful and different approach from Gryphon's approach that employs relative probabilities.

Toward absolute probabilities: A flow chart analysis of paths for escape from a laboratory

To determine the absolute probability of escape for a matHPAI virus from a BSL3 laboratory, a number of events must occur, beginning with an incident that can involve mechanical or equipment failure or human error. The flow chart in Figure 1 describes the events and connections among events, and it lists symbols for probabilities[2] that would eventually lead to an escape. For a matHPAI virus, an escape could lead to a pandemic.

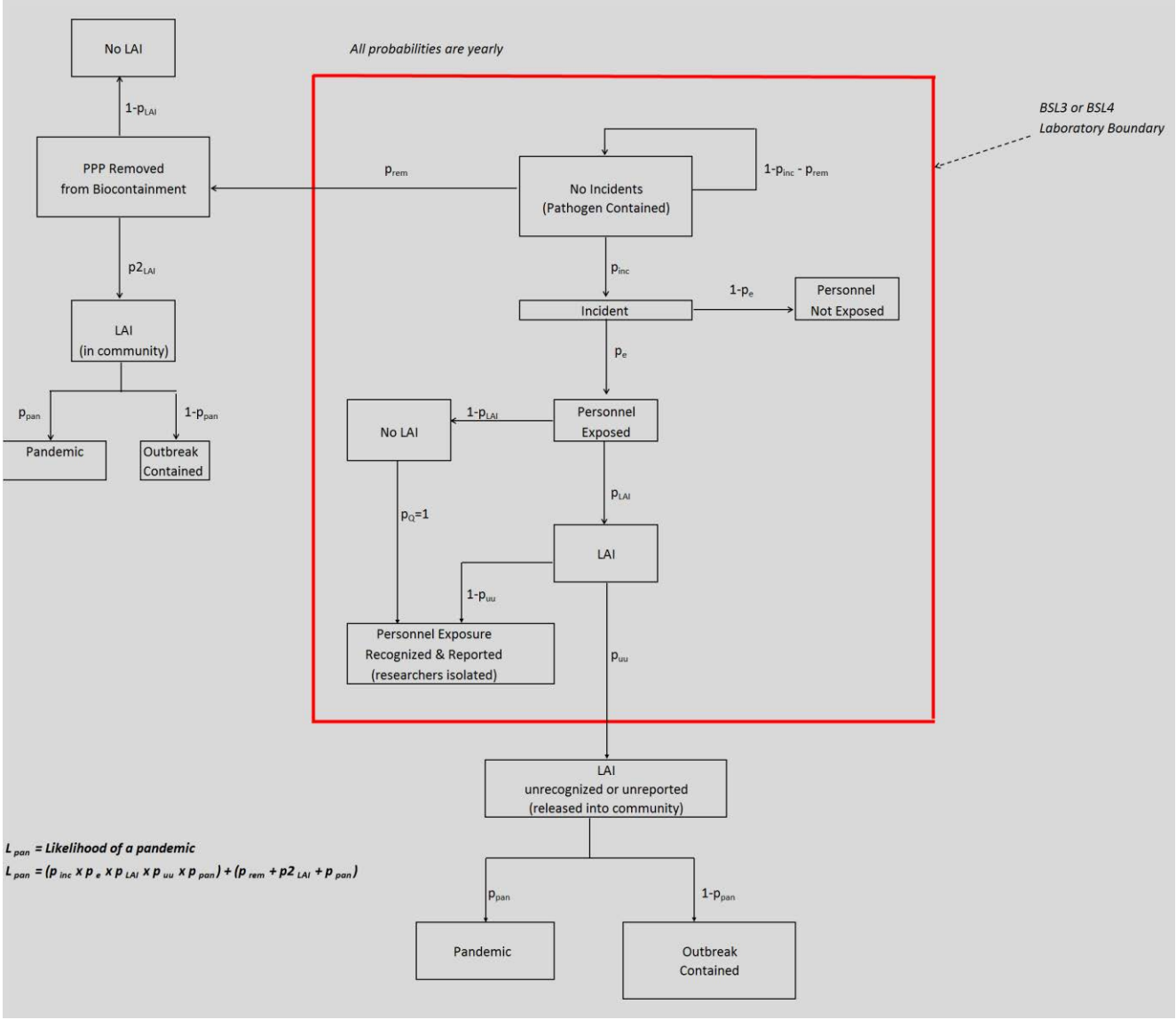

Figure 1. Flow chart of events leading to a lab escape and a pandemic.

In the Figure 1 rendering, there are two independent paths for escape: (1) the undetected or unreported LAI path (top to bottom) and (2) the purposeful removal from containment path (to the left).

For path (1), the likelihood (probability) of a pandemic is $L1_{pan} = p_{inc} \times p_e \times p_{LAI} \times p_{uu} \times p_{pan}$. Here, p_{inc} is the probability that there is an incident is the first place. p_e is the probability that the incident involves exposure of one or more lab personnel. p_{LAI} is the ratio of LAIs to exposures (not strictly a probability because it includes multiple LAI from each exposure). p_{uu} is the probability that the LAIs are undetected or unreported, so infected persons leave the laboratory into the community. In the flow chart, the undetected or unreported LAI moves outside the red laboratory boundary into the community. Finally, p_{pan} is the probability that a pandemic results.

For path (2), the likelihood of a pandemic is $L2_{pan} = p_{rem} \times p2_{LAI} \times p_{pan}$. Here, p_{rem} is the probability that a matHPAI is purposely removed from the laboratory. This could happen for a number of reasons, a common reason being that a researcher has mistakenly believed that the pathogen has been made inactive and is removed for research in a BSL2 lab or removed for transport to another facility.

The overall rate at which pandemics occur (effectively, the probability of generating a pandemic per calendar year) is

$$L_{pan} = (p_{inc} \times p_e \times p_{LAI} \times p_{uu} \times p_{pan}) + (p_{rem} \times p2_{LAI} \times p_{pan})$$

All probabilities in this analysis should be estimated for one year and one lab, as this is the basic probability from which many-lab, many-year escape probabilities can be readily calculated.

Determining values for the probabilities

For path (1), start with p_{inc}. It is a probability that should be obtainable with reasonable accuracy from incident data for many labs over many years. Gryphon should already have this data. I would guess that it is possible that every lab would experience some reportable incident each year, for instance a spill. So, p_{inc} might be 50% or greater. To be a bit more conservative, I will assume that p_{inc} =0.2, which assumes a lab will experience on average one incident every five years (1/0.2). This is likely a generous probability reduction.

In a telephone conversation with Rocco Casagrande, he commented that only 2% of incidents result in personnel being exposed. In analyzing incidents that result in LAIs[3] (Table 1), clearly exposure has occurred.

Thus, the probability that an incident escapes containment and a lab worker is exposed is p_e = 2% = 0.02. So 98% of the time incidents involve no personnel exposure ($1-p_e$ = 98%) and no LAI could occur. Gryphon should be able to comment on the accuracy of the 2% number--that is, how much data supports it. This is a key number.

To estimate the other probabilities, I turn to a table of reported lab incidents collected for the *Final Supplementary Risk Assessment for the Boston University National Emerging Infectious Diseases Laboratories (NEIDL).* (http://www.bu.edu/neidl/files/2013/01/SFEIR-Volume-III.pdf) This 2,716 page risk assessment is abbreviated as the SFEIR (Supplemental Final Environmental Impact Report).

An informative table in the SFEIR is Table D-7, “Recent Reported Incidents Involving U.S. BSL-3 laboratory Facilities.” The table is 27 pages long and lists and summarizes 118 incidents, with 23 incidents involving viruses. The table does not report the number of laboratories reporting and their years of operation, so probabilities for each of the different kinds of incidents cannot be ascertained (the frequently encountered “denominator” problem). However, it does provide a way that allows the probabilities downstream of p_e in Figure 1 to be estimated, using as denominator the 118 incidents.

The table covers 1984 through 2010, with most reported incidents after the year 2000. I sorted the table to collect all the LAIs together. The sorted table, including only confirmed LAIs, with a few columns deleted and a few non-substantive changes, is presented in Table 1 below.

LAI Category	Research agent	Description	Results	Action
Detected	West Nile virus (WNV)	A microbiologist working under BSL-3 conditions suffered a finger puncture from a hypodermic needle harboring WNV being harvested from infected mouse brain (Centers for Disease Control and Prevention 2002).	The wound was cleansed and bandaged. Serologic testing showed evidence of acute WNV infection. Mild symptoms developed and resolved.	CDC determined that applicable handling and biocontainment protocols were followed.
Undetected or unreported	Sabia virus+	A research virologist discovered a leaking vessel upon opening a sealed aerosol biocontainment centrifuge rotor outside of a BSC. Personal respiratory protective equipment consisted of a surgical mask. The incident was not reported (Altman 1994).	Symptoms began 8 days afterward. Two days later the infection was correctly diagnosed.	Antiviral therapy cured the nearly fatal infection. Two external committees strongly criticized the researcher and institution. The university agreed to implement all recommendations. No secondary infections were found among the 142 subsequent human contacts (ref needed)
Undetected or unreported	*Neisseria meningitidis*	A microbiology researcher at BU sought medical attention for laboratory-acquired bacteremia and meningitis. Molecular typing determined the infecting strain was the same strain he had been working with. Work with *N. meningitis* is conducted at BSL-2 using BSL-3 precautions (respiratory protection provided by Class II BSC) (Boston University 2009; Smith 2009, 2009)	Intravenous antibiotics were administered and the researcher recovered fully.	University experts determined the researcher did not consistently wear appropriate personal protective equipment, and did not consistently follow appropriate safe microbiological practices. It was surmised that the researcher touched his gloved hand to his face while working with the bacterium
Undetected or unreported	*Coxiella burnetii*	Previously undiagnosed exposures to *C. burnetii* are diagnosed in three laboratory workers by serologic testing (Centers for Disease Control and Prevention 2007, 2007). As many as ten workers might have been infected (further information is unavailable (Subcommittee on Oversight and Investigations 2007).	Responsible officials did not report these infections to federal authorities as required by federal law.	CDC issued a cease and desist order to TAMU on April 20, 2007 that was expanded on June 30 to include work with all Select Agents. Other serious violations were found during a site visit inspection in July 2007.
Undetected or unreported	*Brucella melitensis*	A researcher contracted undiagnosed brucellosis during improper disinfection of aerosolization chamber. She later required prolonged administration of intravenous and oral antibiotics (Centers for Disease Control and Prevention 2007, 2007; Subcommittee on Oversight and Investigations 2007).	Responsible officials did not report this infection to federal authorities, as required by federal law, until April 11, 2007 in response to an inquiry from the Sunshine Project. (Texas A&M University 2007)	CDC issued a cease and desist order to TAMU on April 20, 2007 that was expanded on June 30 to include work with all Select Agents. Other serious violations were found during a site visit inspection in July 2007.
Undetected or unreported	*Francisella tularensis*	Researchers were working under BSL-2 biocontainment protocol with what was believed to be a non-infectious vaccine strain of the bacterium. Later, it was determined the bacterial culture also contained the infectious wild-type strain that requires BSL-3 biocontainment precautions. Investigation was unable to determine the cause for the mixed culture (Anonymous 2005, Barry 2005, Lawler 2005, Dalton 2005).	Two researchers became infected with *Francisella tularensis* in May and were not correctly diagnosed until a third scientist became infected with the bacterium in September.	An investigation revealed that researchers had failed to follow proper BSL-2 biocontainment protocol, and that the University failed to identify work- related illness in laboratory staff and failed to immediately report suspicious work-related illness to local and state health departments. Biosafety policies and SOPs were revised accordingly. The Chief of Infectious Diseases was replaced.
Undetected or unreported	*Brucella* species	A laboratory worker became feverish months after handling a culture of *Brucella* sp (The Associated Press 2009).	Infection was confirmed in July by laboratory testing.	It was determined that employee had handled the culture without using proper biocontainment precautions. The employee eventually returned to work.
Detected	West Nile virus (WNV)	A microbiologist, working under BSL-2 conditions using a Class II BSC, lacerated a thumb with a scalpel during necropsy of a bird infected with WNV (Centers for Disease Control and Prevention 2002).	The superficial wound was cleansed and bandaged. Symptoms began 4 days post injury; medical attention was sought 7 days after injury. Infection was self- limiting and was confirmed by serologic testing.	CDC recommends BSL- 3 biocontainment measures for WNV. However, CDC does accept BSL-2 biocontainment facilities that incorporate certain elements of BSL-3 biocontainment measures.
Undetected or unreported	*Bacillus anthracis*	A lab worker used an incorrect disinfectant; failed to wear disposable gloves; and failed to cover a pre-existing skin defect (facial cut from shaving) (Centers for Disease Control and Prevention 2002)	Cutaneous anthrax resulted following skin exposure to a contaminated surface.	Patient was successfully treated using antibiotics. CDC reviewed proper biosafety measures with laboratory personnel.
Undetected or unreported	*Burkholderia mallei*	A research microbiologist routinely failed to wear disposable gloves, and became infected. A primary care physician prescribed antibiotics without knowledge of the specific etiology (Srinivasan et al. 2001; Centers for Disease and Prevention 2000).	The patient improved but relapsed to a life- threatening condition. Culture revealed specific etiology and appropriate antibiotics resulted in cure.	A review of laboratory procedures was conducted but no further information is available.
Undetected or unreported	*Mycobacterium tuberculosis*	PPD skin test conversion was noted for a laboratory technician (Johnson 2009).	Source of infection suspected to be from samples sent by outside laboratories. Samples were to have been inactivated prior to receipt, but validation was uncertain.	Policies and SOPs for sampling handling were revised to assume that samples could be infectious. HVAC systems were upgraded. Air flow alarms were added to BSCs. Aerosol-containment centrifuge was added.
Undetected or unreported	*Chlamydia trachomatis*	Researcher was diagnosed with a lung infection soon after working with the pathogen (Johnson 2009).	Policies and SOPs for safe handling of the pathogen were found to be inadequate.	New requirements for PPE (respiratory protection), use of a BSC to open centrifuge rotors/buckets, and correct use of BSC were instituted.
Undetected or unreported	*Mycobacterium tuberculosis*	A retrospective survey was sent to 56 state and territorial public health laboratories to determine, by skin tests results, the frequency of probable laboratory-acquired tuberculosis.	Seven laboratory workers were determined to have laboratory-acquired infections (Kao et al. 1997).	CDC guidelines for preventing LAI tuberculosis, and recommendations for regular skin testing of laboratory employees, were re-emphasized.
Undetected or unreported	*Brucella melitensis*	A laboratory worker thawed a frozen vial of bacterial suspension and inoculated a plate culture on the open bench top instead of within a BSC (Staszkiewicz et al. 1991)	Eight laboratory workers became infected, one being asymptomatic. The outbreak was most consistent with airborne spread.	The 7 symptomatic workers were given antibiotic therapy. One relapsed and required alternative therapy. Enhancements to laboratory SOPs were recommended by the Department of Epidemiology and the Infectious Diseases Division.
Undetected or unreported	*Mycobacterium tuberculosis*	Three researchers became skin-test positive for tuberculosis after using a newly acquired aerosolization chamber for experimental infection of animals (Washington Department of Labor and Industries 2004).	Infections were sub- clinical. Prophylactic treatment typically is employed in such cases	Investigation revealed multiple faulty seals in the device, and researchers were not fully familiar with proper operation of the device.
Uncertain soure of infection	Venezuelan equine encephalitis virus.	A laboratory worker was found to have a high rise in anti-VEE virus titer. No occupational exposure was confirmed (Subcommittee on Oversight and Investigations 2007).	APHIS/CDC form 3 (Report of Theft, Loss, or Release of Select Agents and Toxins) was filed.	No further information available
Undetected or unreported	H1N1 influenza A virus (swine)	Two people, working in separate ABSL-3 rooms, each became symptomatic and were diagnosed with influenza 1.5 days after collecting nasal specimens from experimentally infected pigs (Wentworth et al. 1997).	Genetic analyses determined the workers had become infected with the same virus used to infect the pigs.	Investigation determined that an incorrect mask had been supplied to the workers for 1 day, and it is possible this error facilitated infection of personnel.
Uncertain whether detected or not	Various pathogens: U.S. Army Medical Research Institute of Infectious Diseases, Fort Detrick, Frederick, Maryland	A retrospective review of institute records showed that 67 people were evaluated for likely or highly likely exposure to infectious agents (Rusnak, Kortepeter, et al. 2004)	3 LAI from BSL-3 pathogens were confirmed in 3 cases: (Chikungunya virus, Venezuelan equine encephalitis virus, and *Coxiella burnetii*); LAI was likely in a 4th case (*Yersinia pestis*); post-flooding contamination of a lab with *B. anthracis* was detected.	NA (retrospective review)
Undetected or unreported	*Francisella tularensis*	A USAMRIID military scientist was reported to have been diagnosed with tularemia, as a result of her work with *F. tularensis*. (USAMRIID United States Army Medical Research Institute of Infectious Diseases 2009, 2009; Bhattacharjee 2009).	Oral antibiotics were started on an outpatient basis, followed by inpatient administration of intravenous antibiotics. Recovery was expected	No further information available

Table 1. Excerpts from Table D-7 from the Supplemental Final Environmental Impact Report

For the 118 reported incidents in Table 1, 19 involved LAIs in laboratory personnel, some incidents with multiple infected persons. These 19 are shown in the table. In my reading of the table descriptions, 15 of the 19 incidents involved undetected and unreported LAIs, where presumably the infected persons left the lab and entered the community before they were later diagnosed with infection; that is, the pathogen escaped the laboratory. *This is contrary to Gryphon's claim that most exposures would be detected, the infected persons would be quarantined until found to be not infected or until the infection cleared.*

A direct estimate of the probability that an LAI is undetected or unreported, p_{uu}, from these data would be 15/19= 79%. A very cautious matHPAI research lab might quarantine those who thought that they *may have been* exposed. For calculation purposes, p_{uu} = 0.20 or 20% will be used. This may be a generous reduction, as laboratory management and researchers may be reluctant to be quarantined based only on a thought.

Backing up on the flow chart to p_{LAI}, of the 118 reported incidents 17 resulted in LAIs. Taking into account that some incidents involve more than one LAI, the total number of LAIs was 38 (red-highlighted in Table 1). No fatalities were reported, which likely would not be the case with matHPAI. Thus, the probability or rate of LAIs per incident is p_{LAI} = 38/118 = 0.32 or 32%.

The probability values are summarized in Table 2, along with their source and rationale for values used in the analysis.

Parameter Symbol Value Used in Analysis	Definition	Direct Estimate & Source	Rationale for Value Used in Analysis
p_{inc} = 0.2 or 20%	probability there is a reportable incident	likely that every lab would experience some incident each year (e.g. a spill with or without a potential exposure)	assumes, conservatively, one incident every five years, years = 1/0.2 per lab
p_e = 0.02 or 2%	probability a lab worker is exposed in incident	probability is 2% according to Rocco Casagrande comment	2% value used in the analysis implies one exposure every 50 years = 1/0.02
p_{LAI} = 0.32 or 32%	rate of LAIs per incident	118 reported incidents with 38 total LAIs; 38/118 rate or LAIs per incident	32% value used in the analysis
p_{uu} = 0.2 or 20%	probability that an LAI is undetected or unrported	from the LAI data 15 of 19 LAIs were undetected or unreported (uu), implies p_{uu} = 15/19 = 79%	cautious lab might quarantine those *who thought* they may have been exposed, so p_{uu} reduced from 79% to 20%
p_{rem} = ?	probability that an matHPAI is purposely removed from the laboratory	difficult to obtain	not used in the analysis
$p2_{LAI}$ = ?	probability that removed matHPAI will result in an LAI	different from and greater than p_{LAI}	not used in the analysis

Table 2. Summary of probabilities used in the analysis.
(http://osp.od.nih.gov/office-biotechnology-activities/biosafety/institutional-biosafety-committees/incident-reporting)

Although not a large data set, there is enough data here to carry out a preliminary estimate of the likelihood or probability of escape from a lab, L_{esc}.

$$L_{esc} = p_{inc} \times p_e \times p_{LAI} \times p_{uu} = 0.2 \times 0.02 \times 0.32 \times 0.2 = 0.000256 \text{ or } 0.025\%$$

In addition, the 0.025% does not include escapes from purposeful removal from a laboratory. For purposeful removal, probability data might be obtainable from a larger number of incident reports than those collected for Table 1. There is one example of purposeful removal in Table 1, and we know of several more from past human errors and for recent human errors at the CDC and Dugway.

The flow chart and the analysis here should identify explicitly those probabilities where more data might be sought. Even though the probabilities can be made better with more data, those used in the analysis here are likely good enough to provide a fair estimate for the absolute probability of laboratory escape and subsequently the likelihood of a pandemic.

It has been argued that labs working with matPAI are designed to be safer mechanically than other BSL3 and BSL3+ labs. I agree. But human errors dominate. Table D-7 in the SFEIR risk assessment bears this out:

- 82 likely human errors
- 19 likely mechanical or equipment failures
- 3 non applicable incidents
- 14 incidents where it was unclear if human error was involved.

So of the 118 incidents, 82 errors or 69% are human errors, not mechanical or equipment failure. In the bulleted list, I say likely because in a few of the incidents, the descriptions are not clear enough to classify them definitely. Nevertheless, my conclusion holds that many more incidents involve human error than mechanical or equipment failure. Comments in the Gryphon RBA also agree that human errors dominate.

In many of the 118 incidents reported in Table D-7, for example needle sticks, animal bites and other clearly direct exposures “no further information was available.” These are not shown in Table 1, but some may have resulted in LAIs. Most pathogens were not highly contagious or deadly and easily treatable, so I expect the worker could go home.

All that remains is to determine the likelihood of a pandemic from a lab escape from an LAI in the community. For this probability, I consulted Figure 4 in the Lipsitch *et al.* (2003) paper (http://science.sciencemag.org/content/sci/300/5627/1966.full.pdf). The figure is reproduced below for convenience to the reader.

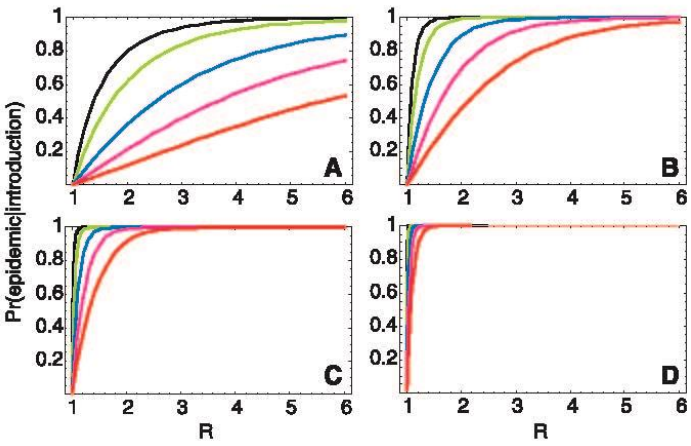

Fig. 4. The probability of an outbreak of SARS in a susceptible population for a range of values of *R*, approximated by the probability of nonextinction of a branching process (*22*) in which the number of secondary cases is given by a negative binomial distribution with a mean of *R* and a variance-to-mean ratio ranging from 1 (for which the negative binomial reduces to the Poisson distribution) to 20 [from left to right: 1 (black), 2 (green), 4 (blue), 10 (magenta), 20 (red)] after the introduction of (**A**) a single infectious case, (**B**) 5 infectious cases, (**C**) 20 infectious cases, and (**D**) 100 infectious cases.

The graphs were generated using branching theory, a pure mathematical construct, which requires only two parameters, the mean R_0 (the reproductive number or the average number of people an infected person infects) and the variance to mean ratio k, which measures the variation in number of people each infected person infects. For instance, some people infected with SARS will infect many other people (super spreaders) and others will infect no one; this implies SARS has a large variance to mean ratio k. I assume for mtHPAI, the subject of this analysis, k will be smaller, perhaps 1 to 2.

Estimating $R_0 = 2$ and k = 2 and a single LAI, the probability of a pandemic, p_{pan}, is about 50% from the green curve in Figure 4a. For more than one LAI entering the community, the probability rises steeply (e.g, Figure 4B for 5LAIs).

Gryphon claims that the probability would not be so high because of public-health efforts to mitigate the spread of community infections. Those of us who watched the 2009 H1N1 pandemic unfold know that such mitigation efforts are likely futile for fast spreading pandemic influenza viruses.

Thus the likelihood or probability of a pandemic for path (1) is estimated to be

$$L1_{pan} = L1_{esc} \times p_{pan} = 000256 \times 0.5 = 0.000128$$

This is the likelihood for a single lab for a single year.

Fatality burden for a single lab in a single year

Assuming the number of fatalities is 4 million, one-tenth of those from the 1918 pandemic flu, the fatality burden for a single lab in a single year is

Fatality burden = 0.000128 x 4 million = 512 fatalities

To put this fatality burden in perspective, no Institutional Review Board tasked with assessing human subject research would approve a proposed research project with an expected 512 fatalities per year.

It should be noted this fatality burden is considerably more than that calculated by me based largely on Gryphon's numbers in my commentary for the January 2016 NSABB meeting. In that calculation, I questioned that their pandemic likelihood was 50% too low, because of an additional 2% probability of unknown origin in the Gryphon analysis. I argue that my calculation using the probabilities estimated here is closer to the true probability of escape. I welcome a response from Gryphon to see if we can reconcile our differences.

For a research enterprise of ten labs conducting this research for ten years, the likelihood of a pandemic is about 100-times greater or 1.28%. I find it very worrisome that laboratory research which could spawn 4 million fatalities has a 1.28% chance of happening in the near future. The assumptions in this analysis are conservative; one reason being that labs in other parts of the world may be much less safe than labs in developed nations.

This live virus research is just too risky to carry out, especially since other means of identifying mutations that lead to airborne transmission in mammals are available. Thus, there is very little to be lost by banning this live virus research.

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[1] "Absolute probability" is the term used by Gryphon Scientific in its risk-benefit analysis (RBA). It seems like a contradiction in terms, since "probability" implies uncertainly, not something absolute. I prefer "direct" probability as it implies leading directly toward a goal. Nevertheless, I will stick with the Gryphon term throughout this analysis.

[2] Each variable p with a subscript is a conditional probability of the event in the chain leading to an accident, given that the previous event in the chain occurred, with two exceptions. p_{inc} is an annual probability (effectively a rate) that an incident occurs. p_{LAI} is a ratio of LAI to exposure, taking into account multiple LAIs in the same exposure event.

[3] Many incidents that must be reported to the NIH involve spills that did not lead to LAIs. The NIH reporting guidelines state "spills or accidents occurring in high containment (BL3 or BL4) laboratories resulting in an overt or potential exposure must be immediately reported." (http://osp.od.nih.gov/office-biotechnology-activities/biosafety/institutional-biosafety-committees/incident-reporting) Potential exposures imply loss of containment to me.

To: National Science Advisory Board for Biosecurity (nsabb@od.nih.gov)
National Academy of Sciences (GOF@nas.edu)

From: Lynn C. Klotz, Ph.D.
Senior Science Fellow
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322 4th St., NE, Washington, D.C. 20002

Home: 5 Duley Street
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Date: March 6, 2016

Addendum to my February 23 Commentary for the March NAS meeting on GOF: Toward absolute probabilities for escape from a laboratory

My February 23 Commentary presents a calculation of "direct" or "absolute" probability of escape from a laboratory of a potential pandemic pathogen, specifically mammalian-airborne-transmissible, highly-pathogenic avian influenza viruses (matHPAI). Absolute probabilities are necessary to calculate the probability of a laboratory escape and subsequently the likelihood of a pandemic from an escape, a key goal of Gryphon Scientific's risk-benefit (RBA) analysis.

To obtain data for my calculation, I employed Table D-7 of reported lab incidents collected for the *Final Supplementary Risk Assessment for the Boston University National Emerging Infectious Diseases Laboratories (NEIDL).* (http://www.bu.edu/neidl/files/2013/01/SFEIR-Volume-III.pdf) This 2,716 page risk assessment is abbreviated as the SFEIR (Supplemental Final Environmental Impact Report).

Table D-7 lists and summarizes 118 exposure or potential exposure incidents in BSL3 labs, up to the year 2010. Although not a large data set, there was enough data in Table D-7 to carry out a preliminary estimate of the likelihood or probability of escape from a lab, which I did in my February 23 Commentary.

This small data set can be considerably strengthened in several ways:

(1) It can be brought up to date by including data from 2011 through 2015.

(2) The original incident reports to NIH should be read to clarify the few cases where summaries were confusing. I assume Table D-7 was prepared by the group carrying out the SFEIR analysis, so it is a secondary source.

(3) Similar data should be available from the European Union, and should be included.

Gryphon Scientific should be well positioned to carry out these three tasks quickly. They may already have much of the data. The original reports to the NIH (and the EU) should be made publically available by Gryphon, with names redacted of course, so we can make our own assessments.

Since the absolute or direct probability of escape for a matHPAI is the most important probability in the risk analysis, every attempt should be made to find a reasonable estimate of it. The method I demonstrated in my preliminary analysis seems to me to be a good way of finding a reasonable estimate.

THE AMERICAN ASSOCIATION OF IMMUNOLOGISTS

Comments of The American Association of Immunologists (AAI) to the National Science Advisory Board for Biosecurity on Gain-of-Function Studies

Submitted on behalf of AAI by Lauren G. Gross, J.D.,
Director of Public Policy and Government Affairs
The American Association of Immunologists (AAI)
March 8, 2016

The American Association of Immunologists (AAI), the largest professional association of immunologists in the world, representing more than 7,700 basic and clinical immunologists, appreciates the opportunity to provide comments to the National Science Advisory Board for Biosecurity (NSABB) Working Group on Gain-of-Function (GOF) Studies.

AAI appreciates the careful and thorough investigation of the risks, benefits, and public health considerations associated with select GOF research studies. The resulting working paper is a well-thought out document that provides an excellent foundation for the final ruling on this topic.

AAI is largely in favor of the draft recommendations that have been provided by the Working Group. There are, however, some concerns that have not yet been fully addressed. Importantly, the steps for implementation of these recommendations are not clearly laid out. AAI strongly recommends that these recommendations be implemented very cautiously to avoid potential burdens, including:

1) negatively affecting beneficial research perceived as GOF, but posing little real danger to public health, and
2) increasing the administrative burden on investigators and/or grant reviewers, taking away time and effort from important experimental research.

To avoid these unintended consequences, AAI believes that Recommendation 2 (to utilize existing policy frameworks) is the most crucial aspect of these new guidelines.

AAI believes that, very unfortunately, an individual intent on using biomedical research for nefarious purposes would not be prevented from doing so by these recommendations, and that instead, restriction of GOF research studies could actually impede advances in discovering the function and transmission of, as well as potential countermeasures against, natural and man-made biological threats. Because the risk profile of GOF studies is similar to studies using select agents, it may, in many cases, be more appropriate to apply current Dual Use Research of Concern (DURC) policies to these studies.

From: Giga Gerard
Sent: Friday, March 11, 2016 4:17 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Public Comment

F. Gerard Lelieveld, Den Haag,
as a concerned citizen
I listened for four whole days to your deliberations at the NSABB and NAS.
Here I offer you my conclusion:

Stop genetic manipulation of ALL virusses and pathogens, all else remains open for debate and allows for malice!!

Thank you.

From: Annie De Groot MD CEO/CSO
Sent: Monday, March 14, 2016 4:48 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Cc: Leo Einck <leinck@epivax.com>; Lenny Moise <lmoise@epivax.com>; Bill Martin Martin <martinb@epivax.com>
Subject: Researchers at EpiVax, Inc. propose slight modification to GOF text

To whom it may concern,

We, researchers at EpiVax, commend the Academy for organizing the recent Gain-of-Function (GOF) Symposium. We participated by Webex and we appreciate the hard work that went into bringing all of the parties together for this important discussion.

We conduct in silico studies to direct vaccine development.

We also create and test synthetic, non-infectious vaccines, that are modified to improve vaccine efficacy, yet we were recently denied funding due to GOF concerns.

We commend the NSABB Working Group's draft paper suggestion to restrict the definition of GOF studies of concern to those which create pathogens. We further commend the text in lines 465 - 468 evaluating the GOF risk in vaccine development studies. We would suggesting edits to that paragraph as follows:

In general, GOF studies that were not considered by the working group to entail significant risks were those that would adapt human pathogens to mammals to generate animal models or enhance the growth of attenuated vaccine strains. Attenuated vaccine strains would conform to the NSABB recommendations regarding transmission, virulence and susceptibility to control measures. GOF studies of antigenic drift or immune evasion using both synthetic and vaccine constructs that are commonly used to guide vaccine selection pose minimal GOF risk.

Best regards

Annie De Groot MD

Annie De Groot M.D.
CEO/CSO EpiVax
EpiVax, Inc.
146 Clifford Street,
Providence, RI 02903

annied@epivax.com

From: Morgan, Kara [mailto:morgankm@battelle.org]
Sent: Monday, March 28, 2016 3:22 PM
To: GOF@nas.edu; National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Cc: BNBI - Fitch, Joseph P <Joseph.Fitch@nbacc.dhs.gov>; Baruch Fischhoff <baruch@cmu.edu>
Subject: Comments for GOF Committee on decision process, criteria and definitions

Members of NSABB,

It was a pleasure to present at the 2nd GOF symposium several weeks ago. I came away from the discussions deeply impressed by the complexity of the issues and also with the thorough efforts of the board to consider all the relevant perspectives and possible options before moving forward.

In thinking about the topic over the past few weeks, I have decided there is one specific area for which I would like to provide additional thoughts. Specifically, how can the system learn from the lessons in the institution-level case-by-case review of studies to determine those that may require additional oversight, and how can the system incorporate those learnings back into the knowledge being used to make those decisions? As a decision scientist, my initial reaction to the institution-level case-by-case review process concerns me because there are challenges in ensuring consistency and probably more importantly, no mechanism for capturing the expertise that is being applied in these decisions. However, from what I heard at the meeting, I understand that the GOF community is not at a point they can *a priori* identify an agreed-upon set of definitions or criteria or standards to apply to those decisions across the board, so that is not a feasible option.

My recommendation is that if "case-by-case" at the institution level is the way decisions are going to made for now, NSABB could institute a reporting requirement – for those institutional boards to submit information on studies that go forward and those that don't. Then, there can be a mechanism to learn from those reviews and decisions about the studies, and share back the learnings (definitions, criteria) to NSABB and to the community. If there seem to be emerging themes from this analysis that NSABB decides should be added to the "additional oversight" process, they can do that. Instead of using the criteria to drive the decision, this process would use the decisions to define the criteria.

This approach would allow for it to be acceptable to let things stay as they are for now in terms of having a "case-by-case" process at the institution level and being fuzzy on definitions, but would define a clear process (and I would recommend setting timelines) for moving forward to clear definitions (as it was stated researchers are looking for). In addition, this approach would include facilitated information sharing, learning and improvement across the enterprise. How NSABB would incentivize folks to share their study proposals and review decisions with you is a problem that will need to be solved, but it seems solvable.

Please feel free to contact me if you have questions or would like additional clarifications.

Sincerely,

Kara Morgan, Ph.D.
Research Leader
Health and Analytics
Office: 614.424-4944
morgankm@battelle.org

Comments on the May 6 NSABB Working Group Draft Report - Recommendations for the Evaluation and Oversight of Proposed Gain-of-Function Research 5-6-2016

Marc Lipsitch
Harvard T.H. Chan School of Public Health

Overall I congratulate the NSABB and its working group on incorporating a number of issues raised at prior meetings and at the NAS Symposium into this revised draft. This draft addresses key issues to a significantly better degree than prior versions.

The following comments are limited to the Findings and Recommendations.

Finding 1 is exactly correct.

Finding 2 is overly optimistic. It makes no reference to the problems of conflict of interest, real or perceived, that arise when those performing oversight are employed by, or funded by, those who benefit from performing or sponsoring the research. It also acknowledges, but does not sufficiently emphasize, that these decisions are made without adequate quantitative data on the risks, and that this lack of data is a direct consequence of the secrecy requirements as interpreted by CDC and other agencies that both regulate and perform GOFROC, and that oversee biosafety and DURC issues more generally, including Select Agents. The recent *USA Today* article http://www.usatoday.com/story/news/2016/05/10/cdc-lab-secret-sanctions/84163590/ clarifies this point further, that as both regulator and subject of regulation, CDC continues to evade public scrutiny of repeated laboratory errors, including three more examples of improperly killed high-containment pathogens being transported out of high containment, thereby circumventing all the mechanical and biological protections specific to high-containment labs.

Finding 3 is correct but is too limited. The fact is that even during the period of highest scrutiny, the current funding pause, there have been NIH-funded GOF studies performed on coronaviruses that violate the spirit, and I believe the letter, of the funding pause, with very unclear explanations given (1). There has been federal funding cited for what is clearly influenza GOF as well (2), also during the funding pause. These are only the examples I have become aware of, and it is very likely that there are others. If even the funding pause ordered by the White House cannot for a short period stop federally-funded GOF research of concern, it is unclear why we should expect that those systems in place before the pause should be adequate.

Finding 4 is unclear as I am not sure what an “adaptive” policy is or what the alternative option would be.

The bold text of **Finding 5** is correct, but the explanatory text is confusing. None of the examples of unjustifiable research is an example of GOFROC, nor even are they all clearly examples of risks outweighing benefits (human subjects not giving consent is a concern for other reasons, not always to do with risks). In line 1159 the text “or that entail benefits that are

unjustifiable in the light of the risks“ appears to misstate what is meant “…entail risks that are unjustifiable in light of the benefits.” Risks must be justified; benefits are the justification.

I also disagree with the statements on lines 1161-2: “There may be GOFROC that should not be funded on ethical grounds but it is difficult to identify or describe such studies based on general or hypothetical descriptions.” Just as there are clear lines of unethical behavior in research involving human subjects, it should be considered unethical (for example) to conduct a study which imposes a risk of starting a large-scale outbreak or pandemic of a virulent pathogen, in order to gain scientific knowledge where similar scientific goals could be met or equivalent public health benefit could be gained through alternative approaches not involving pandemic or outbreak risk. This claim has not been generally accepted to date, and I would not argue that GOFROC to date has been unethical, but I would argue (and have argued in a peer-reviewed publication) that the same principles that lead us to accept restrictions on human subjects research – demanding humanitarian benefit when risks are significant, and only permitting significant risks to humans when alternatives are unavailable – should also restrict GOFROC (3).

Finding 6 seems correct, subject to the concerns about the inadequacy of current mechanisms noted above.

Finding 7 is correct but needs a corresponding recommendation for how to create international oversight, and this is lacking.

Recommendation 1 and supporting text are improved from prior drafts. The “resistance to countermeasures” criterion has been appropriately removed, but it is to some extent retained in the language:

> To be considered “capable of wide and uncontrollable spread in human populations” it must be judged that there would be limited options for controlling the spread of the pathogen other than patient isolation or quarantine. Such a determination might be made, for instance, if humans lack population immunity to the resulting pathogen, if the pathogen would evade or suppress the human immune response, if the pathogen would be resistant to medical countermeasures, or if existing countermeasures would be unavailable globally in sufficient quantities.

The idea that medical countermeasures alone would be sufficient to reduce the risk of spread of a novel infection is untenable, as recent events dramatically illustrate. Even the basic countermeasures of hygiene and safe burial, routinely available in the US, were not “available” enough to prevent the West African Ebola outbreak from infecting tens of thousands. The current Yellow Fever outbreak represents uncontrolled spread of a virus for which a nearly perfect vaccine has been available for decades. While the further spread of this virus will likely be exacerbated by vaccine shortages, the main problem leading to the current amount of spread is not a vaccine shortage but the fact that the vaccine has not been used in advance of

the epidemic in many places. For most anti-flu countermeasures global availability is extremely poor (4). At best, the "unavailable globally in sufficient quantities" proviso essentially is so universally true that the "'limited options" clause would apply to every infectious agent. At worst, it complicates interpretation. The "uncontrollable spread" aspect should be removed from the first criterion for clarity and brevity.

The *Yersinia* experiment of engineering greater pneumonic tropism for plague in an antibiotic resistant strain described in Appendix C is a good example of how the "limited options" proviso complicates the situation unnecessarily. Surely the same experiment to enhance transmissibility, performed in an antibiotic-susceptible strain, would create substantial risk of uncontrolled spread, given that (a) it might not be recognized and properly treated, even in places with good health infrastructure and (b) there are many parts of the world where treatment is not available on a widespread basis for pneumonic plague. This is exactly the sort of project where the "lack of countermeasures" criterion could create a false sense of security.

The paragraph at line 1274 and following is an important addition reflecting discussions at NAS.

The principles for consideration of GOFROC numbered i through viii are also improved. Principle iv speaks of "the same scientific question" while the explanatory text describes "provide the same or very similar information [as a GOFROC approach]." The two should be harmonized to "the same or similar," as one can always define a scientific question that can only be answered in one way, such as "what is the result of performing manipulation X on strain Y?" which can only be answered with one experiment. There should be no opportunity to circumvent this essential criterion by semantics.

I remain concerned that department-level review (which in practice currently means HHS) cannot be independent given the real conflicts of interest between funding and regulating such research. At a minimum, such a panel should include a substantial membership from non-government employees and/or other departments.

Overall, while the principles laid out in this recommendation have many strengths, I am concerned that the institutional arrangement may be essentially indistinguishable from that established by the 2014 HHS Frameworks, which were not judged adequate.

Recommendation 3 is excellent, as is Recommendation 3.1 in particular. It should be made explicit that the secrecy barriers currently in place should be reconsidered in light of the strong evidence that secrecy prevents effective learning from mistakes and accountability. Again the recent USA Today story on CDC lapses is very much on point http://www.usatoday.com/story/news/2016/05/10/cdc-lab-secret-sanctions/84163590/.

Recommendation 4 is appropriate, and I would suggest that specific types of experiments be added to the list of prohibited experiments under the Select Agent Rule, as has been suggested

previously http://www.cidrap.umn.edu/news-perspective/2016/03/commentary-six-policy-options-conducting-gain-function-research .

Recommendation 5 is very important and excellent, and 6 and 7 are very good as well. However more specific ideas for international oversight would be most welcome.

1. **Menachery VD**, **Yount BL**, **Debbink K**, **Agnihothram S**, **Gralinski LE**, **Plante JA**, **Graham RL**, **Scobey T**, **Ge X-Y**, **Donaldson EF**, **Randell SH**, **Lanzavecchia A**, **Marasco WA**, **Shi Z-L**, **Baric RS**. 2015. A SARS-like cluster of circulating bat coronaviruses shows potential for human emergence. Nature Medicine.

2. **Williams GD**, **Pinto AK**, **Doll B**, **Boon ACM**. 2016. A North American H7N3 Influenza Virus Supports Reassortment with 2009 Pandemic H1N1 and Induces Disease in Mice without Prior Adaptation. J Virol **90**:4796–4806.

3. **Evans NG**, **Lipsitch M**, **Levinson M**. 2015. The ethics of biosafety considerations in gain-of-function research resulting in the creation of potential pandemic pathogens: Table 1. J Med Ethics medethics–2014–102619.

4. **Fedson DS**. 2009. Meeting the challenge of influenza pandemic preparedness in developing countries. Emerging Infect Dis **15**:365–371.

****DRAFT****

A Proposed Oversight and Decision Mechanism for Creating and/or Researching Potential Pandemic Pathogens

Lynn C. Klotz, PhD,
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May 12, 2016

Introduction

About two years ago, the White House ordered (here and here) a "deliberative process and research funding pause" for gain-of-function studies on viruses that "would have enhanced pathogenicity [virulence] and/or transmissibility in mammals via the respiratory route." This White-House-ordered activity is now near completion; the National Advisory Board on Biosecurity has just issued its Draft Final Report

The viruses that are the subject of the White House order include highly pathogenic Asian influenza viruses that can transmit disease from mammal to mammal by the respiratory route (airborne transmission). Such viruses have already been created in the laboratory, in particular but not limited to the laboratories of Ron Fouchier and Yoshihiro Kawaoka. If one of these viruses escaped a laboratory, it could seed a pandemic with thousands to millions of human fatalities. These are called GOF studies of concern by the National Science Advisory Board for Biosecurity (NSABB), or simply studies of concern.

Any review mechanism for studies of concern must take into account risk-benefit, biosafety, biosecurity and other international consequences such as demands for reparations for morbidity and mortality from a laboratory escape. Allowing the most dangerous research to proceed sends a message to other nations that such research is acceptable; and it may send the wrong message that the U.S. is embarking on the most-dangerous-imaginable biological weapons development.

A proactive and on-going review process for studies of concern that involves several committees is proposed here:

- A means of identifying which studies could seed a pandemic in humans if a laboratory-created pathogen escaped.
- A Committee of Outside Experts (COE) to review such research to supplement the current Institutional Biosafety Committee (IBC) review and Federal review, presumably NIH internal review.[1]
- A White-House Committee (WHC) charged with making decisions when there is disagreement among the three committees whether the studies should or should not be conducted (banned) in the U.S.

The WHC could include members from the National Security Council, the Office of Science and Technology Policy, the Department of State, the Department of Health and Human Services (HHS) and perhaps others. This committee composition would help ensure that dual-use security concerns, biosafety risk to the community, and international ramifications are addressed. The WHC would recommend to the President to ban a particular study of concern.

The just released NSABB Draft Final Report in its Findings and Recommendations has come to some of the same conclusions as the proposal here; for instance, the possibility of banning some studies of concern:

> **"Finding 5.** There are life sciences research studies, including possibly some GOF research of concern, that should not be conducted because the potential risks associated with the study are not justified by the potential benefits."

Summaries of the current state of affairs, criticisms of the NSABB rules, and discussion of this Proposal follows:

Problems with the NSABB rules for identifying "studies of concern"

In the Gain-of-Function Research Symposium held at the National Academy of Sciences (March 10-11, 2016), the NSABB gave a presentation (Slides 12 and 13) summarizing its conclusions on funding and oversight for GOF studies of concern. The NSABB concluded:

> "Research proposals involving GOF studies of concern…should be reviewed carefully for biosafety and biosecurity implications, as well as potential benefits, prior to determining whether they are acceptable for funding. If funded, such projects should be subject to ongoing oversight at the NIH and institutional levels."

GOF studies of concern needed to be defined. The NSABB offered the following three rules:

> "A GOF study of concern is one that could generate a pathogen with all of the following attributes:
> 1. The pathogen generated is highly transmissible in a relevant mammalian model.
> 2. The pathogen generated is highly virulent in a relevant mammalian model.
> 3. The pathogen generated is more likely capable of being spread among human populations than currently circulating strains of the pathogen."

In its presentation, the NSABB emphasizes that all three rules must apply by underlining the word "all". The White House called for a "deliberative process and research funding pause" for GOF studies on viruses that "would have enhanced pathogenicity [virulence] and/or transmissibility in mammals via the respiratory route." The "and/or" was usually interpreted as "or". The NSABB changing now to the word "all" fundamentally changes the discussion, and could allow dangerous virus strains to escape their studies-of-concern designation.

In the Draft Final Report, the NSABB has dropped Rule 3, but still insists that both Rules 1 and 2 must be met to be a GOF study of concern. In slightly different language:

> "To be considered [Gain of function research of concern] GOFROC, the research must, in a single step or over the course of manipulations, be reasonably anticipated to generate a pathogen with both of the following attributes:

> i. The pathogen generated is likely highly transmissible and likely capable of wide and uncontrollable spread in human populations.
> ii. The pathogen generated is likely highly virulent and likely to cause significant morbidity and/or mortality in humans."

To make the discussion more real, let's concentrate on one type of pathogen: mammalian-airborne-transmissible, highly-pathogenic avian influenza viruses (matHPAI). Some of these dangerous matHPAI strains created in Fouchier's and Kawaoka's laboratories might not qualify as studies of concern under the NSABB rules. For instance, a strain that is highly transmissible and only modestly virulent in ferrets might not be captured as a study of concern. We would certainly not like to see such a strain escape from a laboratory. The problem here is that both rules must apply to qualify according to the NSABB.

What exactly is meant by "highly virulent" or "highly transmissible" in Rules i and ii? Higher or lower virulence and airborne transmissibility of pathogens in ferrets cannot reliably be extrapolated to humans. We must take a careful approach by assuming many of these pathogen strains might seed an uncontrollable outbreak (pandemic), unless they are deemed not dangerous after careful analysis.

Proposed new rule for Identifying studies (research) of concern

Many of us active in the deliberative process use the expression "potential pandemic pathogens" to better identify pathogens of concern, which would focus disagreements on pandemic potential, not on the vague word "highly."

Pathogens that exhibit, or reasonably could be expected to exhibit, pandemic potential are abbreviated PPPs, obviously standing for potential pandemic pathogens.

"Reasonably could be expected to exhibit" is an important phrase here, as pathogens of concern are laboratory-created and are novel, so their pandemic potential has not been observed in nature. With this definition of a PPP, the two NSABB rules might be rewritten simply as a single rule:

A study of concern is one that creates in the laboratory or studies a live laboratory-created PPP not present in nature that reasonably could be expected to be virulent in humans or transmissible in humans by aerosol-droplets or other means of efficient transmission not requiring direct physical contact.

The focus for this proposal is narrowly defined to humans. The NSABB's "relevant mammalian model" is not necessary as part of the definition, although demonstration of mammalian airborne transmission of HPAI in ferrets was the original trigger for widespread concern and will remain a trigger for concern.

Ebola is an example of efficient (non-airborne) transmission with and without direct physical contact. "Not present in nature" excludes pathogens already in the community prepared from plasmids, as is common today for influenza viruses. It also excludes natural strains of pathogens (not laboratory-created) already in the community, such as MERS.

This rule is an attempt to find a rule(s) that is not too narrow so as to exclude some studies of concern, and not too broad so as to include safe studies. From the many discussions leading to this rule, it is clear that drafting a perfect rule is likely not possible. The Committees described here will sometimes have to make decisions to include or exclude particular studies based on their assessment of virulence, transmissibility, and other factors. With experience, the rule may well be modified.

A Committee of Outside Experts to supplement IBC and NIH review

An NSABB quote in this article refers to "NIH and institutional" review. History tells us that institutional review followed by NIH review has been ineffective.

Review by institutional biosafety committees (IBCs) has been incompetent to non-existent. See, for example, the discussion in Chapter 7, "Who's Minding the Store," in Breeding Bio Insecurity where it is suggested why IBC's do not effectively carry out their duties:

> "The root of these failures probably lies in the free-spirit culture of scientists unaccustomed to regulations and suspicious of them, and the inability of the already-dysfunctional Institutional Biosafety Committees to deal with the new era of security regulations."

The review and oversight process cannot begin unless IBCs contact NIH about questionable research project proposals. There should be stiff and enforced penalties for failure to report to the NIH.

The history of NIH review is concerning as well. Again, from Breeding Bio Insecurity:

> "[M]ost of the law's oversight provisions are guidelines and not legally enforceable…the NIH can withhold funding from those violating the guidelines. But the agency doesn't and won't: too much vital research might be impeded. Even prestigious universities pay only lip service to the guidelines, many not even that."

Recent NIH grant awards for the studies that created and researched live matHPAI viruses do not inspire confidence in that particular NIH review. It appears that these studies were funded with little questioning of their risk, certainly without public discussion.

IBC and NIH review should be supplemented by a Committee of Outside Experts (COE) review. From the scientists, ethicists, lawyers, and international policy experts who have participated in the deliberative process, it should be possible to put together a committee that represents all facets and views.

The NSABB Draft Final Report agrees that a third committee is needed:

> "**Finding 3.** Oversight policies vary in scope and applicability, and do not cover all potential GOFROC, therefore, current oversight is not sufficient for all GOF research of concern."

and

> "**Recommendation 1.** Research proposals involving GOF research of concern entail significant potential risks and should receive an additional, multidisciplinary review, prior to determining whether they are acceptable for funding."

Final decisions about proposed studies of concern

The kinds of decisions that might be made range from:

- Outright banning a particular study in the U.S.
- Allowing a study to proceed and be funded at an appropriate biocontainment level BSL3, BSL4 or BSL4+[2]

When the three committees (the IBC, NIH, and the COE) all agree on a decision that does not call for banning the study, the NIH can notify the researchers' Institution of the decision. If one or more of the three committees recommends banning the proposed research, the Final Decision will be made by the President from the advice of the WHC.

The obvious reason for high-level WHC review is that a lab escape of a live pathogen could cause an uncontrolled outbreak, with thousands to millions of fatalities. Even the relatively mild 2009 H1N1 pandemic flu killed over 200,000 people around the world.

But there are other reasons as well for Executive-branch review. Casualties outside the U.S. could make the U.S. liable for reparations, and certainly international condemnation. Also failure to ban the most dangerous research sends a message to the rest of the world saying that such research is acceptable; and it may send the wrong message that the U.S. is embarking on the most-dangerous-imaginable biological-weapons development.

There is already a framework in place to guide funding decisions for matHPAI research. The 2013 framework outlines the criteria for funding.

> 'Such proposals will undergo additional funding agency review as well as [HHS] Department-level review in order to determine its acceptability for funding by HHS…the funding agency will determine whether the proposed research is in accord with the following criteria:
> 1) The virus anticipated to be generated could be produced through a natural evolutionary process;
> 2) The research addresses a scientific question with high significance to public health;
> 3) There are no feasible alternative methods to address the same scientific question in a manner that poses less risk than does the proposed approach;
> 4) Biosafety risks to laboratory workers and the public can be sufficiently mitigated and managed;
> 5) Biosecurity risks can be sufficiently mitigated and managed;
> 6) The research information is anticipated to be broadly shared in order to realize its potential benefits to global health; and
> 7) The research will be supported through funding mechanisms that facilitate appropriate oversight of the conduct and communication of the research."

Presumably, this framework allowed funding of the Kawaoka and Fouchier matHPAI studies before the 2014 funding pause and deliberative process. A Committee of Experts could well decide that these studies should not be conducted. And the many scientists who signed the Cambridge Working Group statement feel that studies such as these should be "curtailed" until they are reviewed again.

> "For any experiment, the expected net benefits should outweigh the risks. Experiments involving the creation of potential pandemic pathogens should be curtailed until there has been a quantitative, objective and credible assessment of the risks, potential benefits, and opportunities for risk mitigation, as well as comparison against safer experimental approaches."

To be kept informed of decisions, an appropriate Congressional Committee or Caucus will be notified of the Final Decision, along with the three committee's decisions and explanations. The Congressional Biomedical Research Caucus[3] is perhaps the best congressional group to keep informed.

Conclusions

Completion of the NSABB deliberative process should not mean the funding pause should be lifted. All studies subject to the funding pause should remain unfunded by the NIH until a new review process, such as that proposed here, is put in place and new reviews are carried out for all existing studies of concern. The U.S. government should also consider stopping all studies of concern regardless of funding source until they are reviewed again.

This proposal does not address the dual-use concern that someone will use the research for hostile purposes. How to decide what is dual-use research of concern and decisions about its publication might follow a procedure similar to that outlined here.

I thank Richard Ebright and an anonymous reviewer with considerable expertise in controversial science/technology issues for many rounds of comments on this Opinion article, particularly on definitions, the rules, and whether the rules are too narrow or too broad.

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[1] Called Federal review by the National Science Advisory Biosecurity Board. Federal review is likely review by the NIH Recombinant DNA Advisory Committee (RAC) or the NIH Office of Biotechnology Activities (OBA). It may also include review by the Department of Health and Human Services (HHS).

[2] An additional level of biosafety -- call it BSL-4-plus -- that adds special protections for laboratory work with dangerous PPP research. BSL4+ differences from BSL4 include (1) Train full-time technical staff who are dedicated to working with highly dangerous pathogens. These staffers would carry out experiments directed by scientists who would never need to be present in the BSL-4+ laboratory. With modern audio-video technology, research scientists can remotely monitor lab work as if they were present. (2) Require lab staffers to follow up extended work shifts with periods of quarantine before they leave the biocontainment area. Such procedures would assure that no potential pandemic pathogen escapes from a BSL-4+ lab through a laboratory-acquired infection; anyone accidentally infected would show symptoms while still in quarantine.

[3] The Congressional Biomedical Research Caucus (CBRC)…is a bipartisan, bicameral Caucus…Seventy five Members of the House of Representatives and nine Members of the Senate comprise the Caucus Membership...
The Caucus seeks to support the excellent efforts of the congressional committees and Members of Congress with jurisdiction over the National Institutes of Health (NIH), the National Science Foundation (NSF), science research, and health issues.

Comments on NSABB May 6, 2016 Draft Report "Recommendations for the Evaluation and Oversight of Proposed Gain of Function Research"

Submitted by:
Tom Inglesby, MD
UPMC Center for Health Security
May 20, 2016

Finding 1: Agree with all

Finding 2: Main points unclear as written. In principle, yes I agree that there are places in the research cycle where risks could be managed – if the right policies and effective implementation were in place. But as written it implies that the correct US policies are already in place. It cites a range of guidelines and policies already in place and suggests that these policies together aim to manage and oversee GOFROC. But of those policies cited, only the HHS framework for guiding funding for GOFROC research directly relates to this work, and that framework only applies to H5N1 and H7N9 influenza, not for other influenza or for other respiratory viruses. All but the HHS framework were in place before the GOFROC concerns arose in 2012 and did not stop these experiments (or even flag them as of concern.) This finding also implies that federal advisory committees are responsible for oversight or managing risks, and it is unclear what this is referring to. It also implies that journal editors are responsible for oversight or managing risks – prominent journal editors have said clearly they do not agree that they should bear that responsibility and aren't constituted to implement that. It is correct to note that GOFROC research not federally funded does not currently appear to be subject to oversight. It is true that institutional oversight will vary widely, depending on local expertise and culture. It is true that data is limited regarding the rate and extent of laboratory accidents and near-misses and that no single entity collects all relevant accident data.

Finding 3: Agree with some of this, but it does not go far enough. Agree that current policies are not sufficient for all GOFROC. However, the Finding implies that research subject to Select Agent rule would be receiving oversight for GOFROC issues, and this is not true. It's also the case that DURC policies have not appeared to flag GOFROC research for additional oversight, so we shouldn't expect that policy to identify GOFROC. It is good to point out that GOFROC not funded by USG is currently outside of oversight processes, and that should change. Good to point out that other countries fund GOFROC and that the US policy has nothing to do with this, but the DRAFT recommendations should say more about what the US should do to try to reach international consensus in line with some of the major findings of these recommendations. It is important to point out that there are gaps in oversight in US, and that there are substantial implementation issues.

Figure 4: Unclear what "Adaptive Policy Approach" means. Would more clearly define this term. I do agree with the sentiment expressed that new information and data should influence the policies that are established for GOFROC as knowledge and experience gained. Publishing the series of HHS/NIH (and other federal agencies) reviews of proposed GOFROC research would be valuable to the research community in that it could assess more clearly how decisions are made. These reviews could be anonymized as needed and the particulars of new research ideas removed so that intellectual property protected. It will be important for the oversight and risk management process to get smarter with learning as it evolves.

Finding 5: Agree with bolded text. However I do think it is possible to identify GOFROC research as being unethical – i.e. proposed GOFROC research would be unethical if it exposes large numbers of the public to significant risk without the possibility of substantial public health gain, and if that gain cannot be made using any other safer approach.

Finding 6: Agree. But would be clear about what additional oversight and containment mechanisms are appropriate for GOFROC, either with definitive recommendations or at least illustrations of what additional mechanisms are needed.

Finding 7: Agree. Though draft recommendations should be more specific about what should be done internationally.

Recommendation 1: This recommendation is stronger and clearer than in earlier NSABB drafts, but some ambiguity remains. The two criteria to identify GOFROC are correct. However the first attribute could be clearer. If a newly created pathogen is highly transmissible, it is by definition capable of wide and uncontrollable spread in human populations -- these are not separate criteria. Having them listed as distinct can confuse understanding. The existence of a countermeasure for a given highly transmissible disease should have no impact on whether it is classified capable of wide and uncontrollable spread unless it is vaccine that is used nearly universally around the world routinely. In the example of GOFROC influenza, it should not matter that there exists a vaccine or therapeutic that is effective because the majority of the world will not be able to get such a vaccine or therapeutic. In addition, it will not be able to tell in advance of the GOFROC research whether a newly created GOFROC strain would still be protected against with existing vaccines or therapeutics. Appendix C is a good example of the kinds of teaching and guidance materials that will be useful to give to the research community. As noted above, I think a living catalogue of actual experiments that have gone through the GOFROC oversight process that is established, with details regarding how decisions were made, would be quite valuable to the community.

Principles for guiding review and funding decisions: I think the principles are good. However, these principles should dictate not just whether a project should be funded, but also whether it should be allowed to go forward even if not funded by the US government. Recommend that bolded text for criteria *iv* says "the same or very similar" question because while it may not be possible for an alternative approach to answer exactly the same question, it may be possible for an alternative approach to answer a very similar research question that provides equally or nearly equally valuable information.

Review Process for Proposals Involving GOFROC: **Step 1** – it will be important to assess whether most (all?) institutions receiving federal funding have review committees that are deemed (by themselves and HHS) capable of making these determinations. If not, then institutions should get help in getting ready to do this. **Step 2 –** this step seems to leave decisions about whether research is GOFROC to the funding agency program managers. This doesn't seem to be a change from the current status quo which had did not seem to have stopped any GOFROC experiments prior to the Deliberative Pause. It is not clear that program managers who funded the experiments that have now been determined to have been GOFROC (by these new NSABB definitions) would agree that these experiments should be named GOFROC. **Step 3** - A Department level review with a federal panel with diverse views from biosafety, biosecurity, ethics public health etc is an appropriate step, but it appears it will not be triggered unless program managers within the funding agency determine that something is GOFROC, which as noted above, may not occur. The language noted in this step about avoiding real

and apparent conflicts of interest should be applied to Step 2 as well. **Step 4 –** Agree risk management is appropriate step. Not clear who determines what is appropriate risk management. There have been arguments that GOFROC work should only be done in BL4, but NSABB does not take a position on that. And while it lists biocontainment in the text, it is not listed as measure in Box 4. **Step 5 –** Agree.

Recommendation 2: It is good to plan for the continued engagement of external advisory body on these issues, for the reasons articulated. For the committee to be available to all agencies and to be free of funding agency constraints, it should sit outside any one particular federal agency. Agree that the committee should be engaged with the research and public health communities that care about these issues, and it should be transparent and independent.

Recommendation 3: See comments above on need to define "adaptive policy approach. Also see above comment regarding how availability of a countermeasure will not something from being highly transmissible and easily spread. Even if a countermeasure exists, it will not be available for all or most in the world. (unless it is a universally available vaccine, such as a routine childhood vaccine, but it is hard to imagine something qualifying as GOFROC that is protected against by a childhood vaccine.)

Recommendation 4: I agree with this in principle - better to have fewer more comprehensive policies than fragmented ones. However existing policy frameworks have not been effective for GOFROC so far, so would need to ensure the proposals in this report are fully embraced into an existing framework if that is going to be the vehicle to make these changes.

Recommendation 5: Agree.

Recommendation 6: Agree

Recommendation 7: Agree with the recommendation and the text, and the goals stated around international engagement are very important. But it would be useful and important for this recommendation to provide additional concrete proposals for how to engage the international community. The international engagement efforts to date have not been highly attended by the international scientific and relevant policy communities and have mostly been limited to US and European representatives. It is important to expand those dialogues and to consider concretely what norms and international agreements might be established that address GOFROC.

From: ROLAN.CLARK
Sent: Tuesday, May 24, 2016 12:40 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: please insure words/processes have common meaning

NSABB,

Words are important, especially across different languages and I believe the INTENT of any word/process be determined to have a common standing.

I believe there should be a Federal oversight department for ALL biolabs to insure common meanings/processes are understood and a single source of reference.

Respectfully submitted,

Rolan O. Clark

From: Megan Joan Palmer [mailto:mjpalmer@stanford.edu]
Sent: Tuesday, May 24, 2016 2:00 PM
To: National Science Advisory Board for Biosecurity (NIH/OD) <NSABB@od.nih.gov>
Subject: Public comment that was missed

Dear Members of the NSABB –

Thank you for the opportunity to provide public comment. I regret not being able to join you in person. These comments do not reflect the positions of the organizations with which I am affiliated.

First, thank you for your hard work and dedication to public service in navigating a very complex task. Thank you also for ensuring the public comment remains open beyond today's meeting so that I - and others - can provide ongoing and more detailed feedback as your recommendations proceed to the next stage.

I've been encouraged to hear some thoughtful reflections on the role of the NSABB and some important questions regarding the implementation of your recommendations and the precedent they set.

I wanted to highlight and emphasize three points of discussion that have been raised yet I believe have not been sufficiently addressed.

First, the NSABB has an important opportunity at this stage to reflect critically on the success and failures of the process they have just undertaken – and to share these reflections with the government to inform what its role can and should be in the future. This reflection is especially important given the recommendations you have made regarding future advisory boards with roles that partly overlap with the originally envisioned role of the NSABB. There have been many challenging questions about the scope and authority of this group, and capturing these reflections will be important to deciding whether the narrow focus on GOF, the types of risk-benefit and ethical analyses, and the authority and composition of the board sets a good precedent.

A second related point is to encourage you to critically examine the potential unintended consequences of your recommendations being adopted beyond GoFRoC. It should be made more clear within your comments the extent to which you believe these set a meaningful precedent for the principles and structure of oversight beyond gain of function research. Choices made in the name of expediency – such as only examining human health – may not be something you want to promote more broadly

Last, there have been important questions about the clarity and specificity of your recommendations and how they might be perceived and implemented – and most importantly **who** might implement them with **what resources**. I realize that your role is advisory, that several of you have said you do not feel comfortable making more specific recommendations, and that ex-officio members have been clear that they are prepared to continue to dig into the details. However I wanted to encourage you to include a recommendation that the research into implementation – the design of the details - be performed as openly as possible. Many of you have expressed that it will be vital to promote learning between institutions and emphasizing that opening up this process is important to this learning - and will require resources – is an important message.

Best,

Megan J. Palmer, Ph.D.

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Policy Working Paper [Revision 0.9]

Beyond risk-benefit analysis: pricing externalities for gain-of-function research of concern

Owen Cotton-Barratt*†, Sebastian Farquhar†, Andrew Snyder-Beattie*

March 2016
*Future of Humanity Institute, University of Oxford
†Global Priorities Project, Centre for Effective Altruism

Executive summary

The recent US moratorium on certain types of Gain-of-Function‡ (GoF) research[1] made it clear that a new approach is needed to balance the costs and benefits of potentially risky research.[2] Current risk management tools work well in the context of most laboratory risk, where risks are local.[3] However, in the case of potential pandemic pathogens, even a very low probability of accident could be unacceptable given the consequences of a global pandemic. Although quantitative assessment is feasible for these low-probability, high-stakes risks, simultaneously comparing these risks with the qualitative benefits of such research is an especially difficult task.[4]

In this policy working paper we outline an approach for handling decisions about GoF research of concern. Our central policy objective is that:

> *Proposals for research projects with the possibility of catastrophic accident should have an independent estimate of the expected damage, and this figure should be explicitly included in the cost of the research project.*

Our policy objective has three key advantages:

1. It keeps decisions about which science is worth funding in the hands of scientists.
2. It incentivizes sponsors to fund research only when the scientific merit outweighs the costs because the negative externalities are considered as part of the cost of the research project - without the need for a direct benefit-cost analysis.
3. It provides a generalizable solution, which can be applied to other emerging risks from science and technology.

We propose and compare two different approaches to achieving the policy objective.

‡ In this paper we use the term 'Gain-of-Function' to refer only to the research covered by the recent White House moratorium.

Notes: We are grateful to Carrick Flynn for research assistance and to Anthony Aguirre, Leah Broad, Marc Lipsitch, Kathryn Mecrow, Piers Millett, and Stefan Schubert for their comments on drafts.

The first is to establish strict liability for any damages that result from GoF research of concern, and to require grant-holders to purchase liability insurance as part of the grant. The strength of this approach is market-based and incentivizes insurers to price externalities correctly.

The second approach is to centrally commission assessments of absolute risk and require a payment to a state or non-state body to cover the expected cost. The strength of this approach is that it works even if there may be no clear liability after the fact, so could address biosecurity as well as biosafety risks.

Framing the issue

Recent controversy has emerged around certain types of GoF research. Scientists remain deeply divided on both the benefits and the risks of such research.[5]

The controversy culminated in a moratorium on GoF research of concern pending an independent assessment of the risks and benefits. The NIH commissioned an assessment from Gryphon Scientific, released in December 2015. The report did not draw firm conclusions on whether the benefits of such research outweighed the risks.[6]

Challenges of risk-benefit analysis for scientific research

In principle, analysing the risks and the benefits of research and weighing these against each other is the correct way to determine whether to pursue risky research. In practice, both sides of this are very difficult to analyse. Gryphon Scientific was able to present a tentative quantitative analysis of absolute biosafety risk, but only a qualitative analysis of the benefits of the research and of the biosecurity risk.[7] That this major review was not able to analyse risks and benefits on a common scale demonstrates the difficulty of this type of analysis.

Existing solution: the scientific grant process

The scientific grant-making process is the primary mechanism for assessing the uncertain benefits of research against their costs, including the opportunity cost of unfunded research. Although it is hard to judge quantitatively, expert reviewers assess the potential for scientific excellence in different proposals. They must regularly make trade-offs between projects with disparate and uncertain benefits.

GoF experiments are the outcomes of successful grants. But these are currently assessed primarily on the basis of scientific merit and potential benefits, with comparatively little emphasis on the scope of possible risks. Risks from research, just like the benefits, impose an externality on the public. Because the risks are not considered as explicitly, a risky project could get funding over a safer one which has equal or only slightly lower expected benefit. This means that the public is implicitly subsidising risky research relative to safe research.

Our policy approach

Since the benefits are difficult to assess, any direct comparison of risks and benefits is extremely difficult, even when the risks are well-quantified. Rather than employing a direct comparison, we

suggest using an absolute risk assessment to price the expected risk, and to explicitly include this cost in grant proposals. This allows the scientists making grant allocations to use their judgement to pick projects with the greatest potential benefits, given their true social costs.

A generalizable solution

Biotechnology is not the only research area that could create the potential for small probability, high impact risks. Other fields might need to grapple with similar governance issues.[8] A solution that could be extended to other fields, with moderately straightforward generalizations, might avoid harmful controversy and delays while the issues are resolved. Our approach is likely to be generalizable in this sense.

Policy target: have risks priced into grants

In this section, we outline the intended results of pricing risk externalities into grants. We explain what this would look like, why we think it would be beneficial, and how it could perform better than existing safety approaches. In the next sections, we explore two potential mechanisms for achieving this.

Key policy target

Our central aim is:

> *Proposals for research projects with the possibility of catastrophic accident should have an independent estimate of the expected damage, and this figure should be explicitly included in the cost of the research project.*

If the cost is explicitly included, the project would internalise the negative externality associated with risks to the public. For now, we set aside the issues of where the independent estimate comes from, or where the money to cover this explicit cost goes. We will return to these questions in the next two sections.

Benefits of achieving this target

The principal benefit would be to keep decisions about experiments in the hands of scientists, who are best-placed to evaluate the potential benefits, while removing the implicit subsidy for risky research over safer research.

This would have a number of instrumental benefits. First, it should mean that experiments are funded precisely when the benefits outweigh the costs (including both the risks *and* the opportunity costs of not funding other experiments). Second, it would incentivize scientists and laboratories to look for alternative ways to run experiments that would reduce the risk, as this could reduce their extra costs.

While there are significant existing biosafety measures, in many nations these are driven by regulations focused primarily on occupational health (the safety of lab workers), rather than public health.[9] This focus accurately reflects the median historical risk, since most lab acquired infections

have not been passed on. However, it does not reflect the risks posed by experiments which could cause pandemics, where most of the risk exists in the small chance of catastrophic public damage. Our proposal would incentivize effective ways to minimise these risks.

Effects on grant process and domain of applicability
The independent risk assessment could take place before grant applications are submitted (at the request of the group intending to apply for a grant) or after the grant application is submitted (at the request of the grant body). In either case, some time and work would be needed for a proper risk assessment.

If it affects many areas of research, this requirement would significantly increase bureaucratic overhead. Accordingly, we recommend that if implemented, it initially apply only to the GoF research which is covered by the recent US moratorium. In the future, it could potentially be extended to other areas which pose significant risk to public health.

Effects on funding
Laboratories currently receive an implicit subsidy because they do not fully internalize the probabilistic costs of their dangerous activities. If the larger research community were asked to internalize this cost out of their existing limited budget allocation, it would represent an additional unfunded overhead expense and a functional shrinking of the budget available for actual research. Since research is potentially of great benefit to humanity, this may not be desirable. Instead we recommend that the government proportionally increase funding for life sciences research to compensate for this additional expense. Although this would increase explicit expenditure by the government in the form of larger research budgets, governments are already responsible for public health crisis management. This essentially transfers expenditure, from crisis management to crisis prevention, by making the implicit subsidy explicit. The primary advantage of this budget reallocation is that it allows for the same functional cost to the public, while removing the distortion of incentives created by the hidden externalities.

Reporting and safety culture

Any approach which penalises laboratories for reporting accidents and near misses in a timely way might harm biosafety and biosecurity in the long run. Reduced reporting makes it harder to use lessons from mistakes to improve lab design and impairs accident response. Mechanisms for pricing risk will work best if they avoid creating perverse incentives around reporting, and we believe that the mechanisms we describe below can be constructed in a way that does so.

First potential mechanism: mandatory liability insurance

Our first approach is market-based. Laboratories conducting experiments in the appropriate class could be mandated to purchase insurance against liability claims arising from accidents associated with their research. Ideally, this research should be explicitly classified as an "inherently dangerous activity" by the legislature. This will establish strict liability for any damages caused by accidents,

which means that laboratories would be liable even if there was no negligence. Strict liability is already legally established for other inherently dangerous activities analogous to this research, and might well be the legal standard used in many common law jurisdictions in a GoF case even without legislative intervention. The advantage of making this clearly established is that it would provide laboratories with strong incentives to minimise risk.

It is beneficial to require insurance, rather than just ensure there is liability, because of the "judgement proof problem."[10] Many universities currently self-insure against the damage of accidents in their research. This makes sense for occupational and small-scale public health issues, but for cases where there is a small chance of catastrophic damage, the institution may not have enough assets to cover the potential damage. Additionally, a blanket policy of self-insurance may mean that financial planners within universities do not even carefully consider liability risks of their specific research activities.

Advantages of the liability approach

There are a number of advantages to taking this market-based approach. First, it is a relatively light intervention, requiring less ongoing work from the state. Second, it incentivizes insurers to accurately estimate risks, reducing possible politicisation of the risk assessment process. Scientists and engineers would also be incentivised to devise effective safety protocols to reduce their institutions' insurance premiums. Imposing liability has been seen to improve outcomes in other domains such as occupational safety, medicine, and general risk management in non-profits and governmental agencies.[11]

Possible issues with the liability approach

A big question about mandatory insurance is whether insurers would in fact be willing to insure against these outcomes. There are two main reasons why they might not.

The first is that the potential risks are simply too large. A bad global pandemic could kill hundreds of millions of people, and even the largest reinsurers would be unable to absorb this cost without bankrupting themselves (costs above this level will be implicitly backed by the state or the public in any case). It is better to be explicit, and cap liability at a specific industry-wide figure. If the cap were sufficiently high, the effect would be improved risk aversion, even if the tail risk for the insurer were not fully internalised.

Secondly, the risks are hard to model and the market would be small. Developing models to estimate the risk could be more costly than the expected profit from participating in the market. Moreover, developing these models is a difficult task requiring rare expertise. However, insurers already have models for other hard-to-anticipate risks, such as terrorism and global pandemics. If necessary, the development of appropriate models to facilitate this insurance could be explicitly subsidised.

Aside from whether or not insurers are willing to take on the risk, there are challenges to international adoption. For example, it may prove difficult to harmonise liability laws across jurisdictions, particularly in international collaborations.

Aside from the question of whether any firms would be willing to insure against this risk, liability insurance can potentially increase moral hazard, by making actors less responsible for the consequences of their actions. If the deductible/excess on the insurance were set correctly, it could reduce the moral hazard while averting the judgement-proof problem. It also seems that in the particular case of GoF experiments undertaken to benefit public health, the nonfinancial consequences for any scientist who was involved in an accident with major harms to the public would be great enough to serve as a deterrent to reckless behaviour, even if financial consequences were mitigated by holding liability insurance.

Finally, depending on which legal framework is used in these types of cases, the market-based approach might not be able to capture various biosecurity risks. This is because it will likely be difficult in these instances to attribute a disaster to a specific project. This is particularly true with information biosecurity risks. It may, however, be possible to employ other methods of attribution, such as 'market share' liability. In contrast, the approach in the next section could potentially treat biosecurity risks in the same way as biosafety risks.

Second potential mechanism: centrally-commissioned risk assessments

The second approach is to centralise risk assessments. When an area of potential concern is identified, a body commissioned by the state would perform an analysis of the risks involved. This might be similar to the recent Gryphon Scientific analysis, except that it would not attempt to analyse the benefits, and it would focus only on producing the best-estimate absolute risk analysis for different kinds of work, rather than leaving it at a qualitative level. This absolute risk analysis would present its outcomes in monetary terms, using Value of Statistical Life figures to convert fatalities into a cost.

In order to do work of the relevant type, laboratories would be required to pay the corresponding cost to a central authority. This would most naturally be the body or bodies likely to absorb the cost in the event of catastrophe; such as the government's public health and disaster management agencies. It could also be used, in part or whole, to support the cost of the risk assessments.

Advantages of centrally-commissioned risk assessments
Compared to the market-based approach, centralising the risk assessments has two main benefits. First, it can be done without needing to persuade insurers to enter the market. Second, it does not suffer from the same legal uncertainties as the market-based approach. This is especially relevant in the biosecurity context, where attribution might be difficult. The Gryphon Scientific report concluded that the biosecurity risks looked at least as large as the biosafety risks, so this may be a significant benefit.

Questions and issues for centrally-commissioned risk assessments
Three unresolved questions are:

1) Who would make the risk assessments? This could potentially be an independent agency or

an outside contractor. It might be difficult to build the capacity to do the assessments well; note that the Gryphon Scientific report did not fully support the absolute risk analysis it presented, and did not offer any absolute risk analysis for biosecurity risks.

2) Who should receive the money that is included in the price of the grant? Should it be retained at the national level, or shared internationally (since the risks are global)?
3) How would this system work with international collaboration or non-domestic accidents?

A potentially larger issue is ensuring fair and accurate risk assessments. In the case of liability insurance, market forces help align the incentives to motivate insurers to make accurate risk assessments. If assessments are centrally commissioned, there is no such force keeping them in check, which means they would be at risk of becoming politicised.

Comparisons

We have outlined the advantage of our approach, which works by aligning the incentives for scientists and funding bodies more closely with those of society as a whole. This may be the only way to keep the assessment of the benefits of scientific research purely in the hands of scientists, while also reducing risks when appropriate. We have explored two different ways to achieve this. Each has its own advantages and disadvantages.

Overall, the liability approach is more market based. As a result, the risk-assessors have a financial incentive to accurately estimate risk, and political pressures are diminished. It might also be easier to use as a template internationally. Since the risks are global and the potentially risky research is not being pursued in just one country, being able to build global solutions is valuable.

The main benefit of the centrally-commissioned analysis approach is that it bypasses potential legal difficulties with attribution. It may therefore be a more general tool, able to correct incentives for a larger class of risks (such as biosecurity risks and information hazards).

For both approaches, it is important not to punish reporting of laboratory acquired infections or other accidents and near misses. Accurate information and a culture of open reporting are vital for laboratory safety and disease prevention.

Conclusions

Our approach is not to suggest a specific policy, but rather to outline different options which would facilitate a better evaluation of benefits and risks. We have suggested two quite different methods for achieving this. One relies more on market mechanisms, while the other depends on central oversight. Both would require the strong support of regulatory bodies. Each of them has a number of advantages and disadvantages. We do not feel we are in the right position to conclude decisively in favour of one over the other. We would like to encourage discussion among stakeholders of the relative merits of the two approaches.

References

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Cc: Jo Husbands <jhusband@nas.edu>; Viggiani, Christopher (NIH/OD) [E] <christopher.viggiani@nih.gov>
Subject: On NSC, OSTP, and Department of State GOF policy development

Dear NSC, OSTP, and Department of State Senior Officials

The NSABB's final recommendations on gain-of-function research will soon be turned over to you to develop policy, which is why I am now sending you my attached response to the May 24 NASBB meeting. It is titled "Commentary and Revised Proposal: Decision Mechanism for Creating and/or Researching Potential Pandemic Pathogens."

I know it is a presumptuous of me to suggest to you a detailed mechanism for deciding what gain-of-function research-of-concern (GOFROC) should or should not be allowed to proceed in U.S. laboratories. Clearly, the NSC, OSTP and the Department of State will work out policy on this issue; nevertheless, there are reasons for sending my proposal to you.

(1) I am concerned that the NSABB/NIH will decide almost all this research should proceed, even though there is a large group of us who believe the most dangerous research should not proceed (See for instance, http://www.cambridgeworkinggroup.org/ and https://safesci.org/).

(2) From discussion at the May 24 meeting, it seems the NSABB wants to keep decisions within the NIH, with experts outside the NIH just looking in on the decision process from time-to-time. In my view, experts must be intimately involved in decision-making. In my proposal, I indicate one way to involve experts.

Institutional review followed by NIH review has been ineffective in the past. Historically, review by institutional biosafety committees (IBCs) has been incompetent to non-existent. IBCs can be unduly influenced by their institution's scientists, who often have a free-spirit culture unaccustomed to regulations and suspicious of them. Furthermore, I am concerned with the inability of the already-dysfunctional institutional biosafety committees to deal with the new era of security regulations.

The history of NIH review is concerning as well. Most of the NIH's oversight provisions are guidelines and not legally enforceable. The NIH can withhold funding from those violating the guidelines. But the agency doesn't and won't. Even prestigious universities pay only lip service to the guidelines, many not even that.

Recent NIH grant awards for the studies that created and researched live mammalian-airborne-transmissible highly-pathogenic-avian-influenza (matHPAI) viruses do not inspire confidence in that particular NIH review. It appears that these studies were funded with little questioning of their risk, certainly without public discussion.

(3) The NSABB has turned a deaf ear to us. Only after the March meeting at the NAS has the NSABB began to pay any attention to our concerns.

The attached proposal discusses my concerns with the NSABB decision-making rules and principles and outlines one way in which review of GOFROC might be better carried out for the safety of all of us.

Sincerely yours,
Lynn Klotz, PhD
Senior Science Fellow
Center for Arms Control and Non-proliferation

Commentary and Revised Proposal: Decision Mechanism for Creating and/or Researching Potential Pandemic Pathogens

Lynn C. Klotz, PhD,
Senior Science Fellow, Center for Arms Control and Non-proliferation.
Dr. Klotz may be reached at lynnklotz@live.com

June 7, 2016

The NSABB defines Gain of Function Research of Concern (GOFROC) as "research that has the potential to generate pathogens with pandemic potential in humans…"[1] While GOFROC accurately defines the research we are concerned with here, saying and reading the word "GOFROC" is unappealing. In this commentary and revision, I will use the NSABB's old term "studies of concern," to mean the same as GOFROC.

Presumably, NSABB's role will end with suggesting a detailed procedure for making decisions:

> "[T]he White House Office of Science and Technology Policy and Department of Health and Human Services today announced that the U.S. Government is launching a deliberative process to assess the potential risks and benefits associated with a subset of life sciences research known as "gain-of-function" studies…The NSABB will serve as the official Federal advisory body for providing advice on oversight of this area of dual-use research, in keeping with Federal rules and regulations…[and] will inform the development and adoption of a new U.S. Government policy regarding gain-of-function research."

A chain of steps should be followed to make a decision whether a study of concern should or should not be conducted. The steps are:

1. Identifying studies of concern
2. Expert review of studies of concern
3. Making decisions on particular studies of concern.
4. Continuing oversight of conducted studies of concern

In order to protect us from an escape of a laboratory-generated potential pandemic pathogen (PPP) each link in the chain must be strong. The main goal of this commentary is to make suggestions to strengthen each link in the chain.

1. Identifying studies of concern

The review and oversight process cannot begin unless Institutional Biosafety Committees (IBCs) report to NIH potential studies of concern at their institution. There should be severe penalties for failure to report. Penalties should be at least as severe as those for violations of the Select Agent Program, which are up to $500,000 fines, and for some violations up to five years imprisonment. Severe penalties should

force IBCs to err on the cautious side. We must take a cautious approach as potential pandemic pathogens could seed an uncontrollable outbreak.

The IBCs must understand clearly the definition of studies of concern. As of May 24, 2016, the NSABB defines GOFROC (studies of concern) as

> "GOF research of concern is research that can be reasonably anticipated to generate a pathogen with both of the following attributes:
> 1. The pathogen generated is likely highly transmissible and likely capable of wide and uncontrollable spread in human populations.
> AND
> 2. The pathogen generated is likely highly virulent and likely to cause significant morbidity and/or mortality in humans." [2]

These two rules define PPPs. But the definition is a bit complicated, and taking literally the connecting conjunction "AND" could result in not reporting some research that should be reported as studies of concern.

The phrase "can be reasonably anticipated to generate a pathogen" with certain attributes appears to convey the same message as the word "likely" in the two rules; however, the word "likely" modifies the words "highly transmissible…in human populations" and "highly virulent…in humans" What exactly is meant by "highly virulent" or "highly transmissible" in rules 1 and 2? Higher or lower virulence and airborne transmissibility of pathogens in ferrets or mice cannot be extrapolated reliably to humans. We don't know how virulent or transmissible lab-generated PPPs will be in humans, as they are not present in human populations.

With a slight change in wording from my previous commentary to re-state and slightly expand upon the latest NSABB language, I rewrite the two NSABB rules as:

> A study of concern is research that can be reasonably anticipated to generate a pathogen or employs a live laboratory-created pathogen, not presently in nature, with one or both of the following attributes:
> 1. could cause significant morbidity and/or mortality in humans.
> OR
> 2. could be transmissible in humans by aerosol-droplets or other means of transmission not requiring direct physical contact.

The rules have been expanded to include research that employs a previously-created live PPP, as escape from a laboratory would be just as dangerous. The rules have also been expanded to include pathogens such as Ebola virus, which is an example of efficient (non-airborne) transmission without direct person to person physical contact. "Not present in nature" excludes pathogens already in the community prepared from plasmids, as is common today for influenza viruses. It also excludes natural strains of pathogens (not laboratory-created) already in the community, such as MERS.

The word "could" takes into account that we don't know if lab-generated pathogens will be less or more virulent or transmissible in humans than animals, as they are not present in human populations.

The word "AND" in the NSABB definition has been changed to "OR". The conjunction, or, has the logical meaning of and/or. Changing to "or" would give expert reviewers more latitude in deciding whether a study should not be conducted. In some instances, it might make sense to decide that only one of the two both attributes would be necessary to decide that a study should not be conducted. In others, both might be necessary.

To make the discussion more real, take two examples of already conducted research.

2009 pandemic H1N1 and H3N2 influenza viruses were made potentially capable of causing more widespread infection by selecting mutants that could escape immune responses generated against the parent viruses. These antigenic escape mutants could infect and be transmissible in those previously immune. This research might not be captured as a study of concern with the NSABB definition, since the virus strains might not be judged to be highly virulent in humans because the 2009 H1N1 flu virus was not particularly deadly. Regardless of anyone's definition of "highly" that pandemic did cause significant morbidity and mortality world-wide, and so should be considered a study of concern to be reviewed.

Mammalian-airborne-transmissible, highly-pathogenic avian influenza viruses (matHPAI): Some of these potentially dangerous matHPAI viruses have been created in the laboratories of Ron Fouchier and Yoshihiro Kawaoka. If one of these viruses escaped a laboratory, it could seed a pandemic with thousands to millions of human fatalities. By almost everyone's definition, these strains would qualify as studies of concern because they "could" be highly virulent and highly transmissible in humans.

Both the NSABB rules and my rules are focused narrowly to humans. Similar rules and decision-making chains for plant and agricultural animal pathogens should be considered.

2. Expert review of studies of concern

A proactive and on-going review process for studies of concern should involve three committees. After an Institutional Biosafety Committee (IBC) refers a potential study of concern to NIH for review and recommendations[3], the actual decision process should then be in the hands of a third committee (here called the White-House Committee or WHC). This committee would include members from the National Security Council (NSC), the Office of Science and Technology Policy (OSTP), the Department of State, and the Department of Health and Human Services (DHHS).

WHC membership should include non-government experts as well--in particular, members of the National Academy of Sciences (NAS) and other non-government experts. This group would include experts in molecular virology, epidemiology, public health, ethics, and international law. The non-government experts would have an advisory non-voting role on decisions for each study of concern. Because many of the nation's most respected scientists are NAS members, the NAS should have the major role for nominating non-government experts to the WHC.

This WHC Committee composition would help ensure that dual-use security concerns, biosafety risk to the community, and international ramifications are addressed.

3. Final decisions about proposed studies of concern

Final decisions about proposed studies of concern--whether the study could be conducted, protocols and restrictions that must be followed, and biosafety requirements—will be made by the WHC. The kinds of decisions that might be made range from:

- Outright banning a particular study in the U.S.
- Allowing a study to proceed and be funded at an appropriate biocontainment level BSL3, BSL4 or BSL4+[4]

with many possible decisions in between.

Decisions must take into account risk-benefit, biosafety, biosecurity, ethical, and international consequences such as demands for reparations for morbidity and mortality from a laboratory escape or theft. Allowing the most dangerous research to proceed sends a message to other nations that such research is acceptable; and it may send the wrong message that the U.S. is embarking on the most-dangerous-imaginable biological weapons development.

There is a framework in place to guide funding decisions for studies of concern. The 2013 framework outlines the criteria for funding. The NSABB has suggested a somewhat different framework for studies of concern:[5]

> **"Principles that should guide the review of and funding decisions about research proposals anticipated to involve GOF studies of concern:**
> i. The research proposal has been evaluated by a peer-review process and determined to be scientifically meritorious, with high impact on the research field(s) involved.
> ii. The pathogen that is anticipated to be generated must be judged, based on scientific evidence, to be able to arise by natural processes.
> iii. An assessment of the overall potential risks and benefits associated with the project determines that the potential risks as compared to the potential benefits to society are justified.
> iv. There are no feasible, equally efficacious alternative methods to address the same scientific question in a manner that poses less risk than does the proposed approach.
> v. The investigator and institution proposing the research have the demonstrated capacity and commitment to conduct it safely and securely, and have the ability to respond rapidly and adequately to laboratory accidents and security breaches.
> vi. The results of the research are anticipated to be broadly shared in compliance with applicable laws and regulations in order to realize its potential benefits to global health.
> vii. The research will be supported through funding mechanisms that allow for appropriate management of risks and ongoing Federal and institutional oversight of all aspects of the research throughout the course of the project.
> viii. The proposed research is ethically justifiable."

I have some concerns over the wording in this new framework. In general, they should be labelled Guidelines, not Principles. My main specific concern is over Principle iv. The mutations responsible for airborne transmission of matHPAI strains found by Fouchier can be found by alternative methods that do not employ live viruses, so the risk of a lab escape seeding a pandemic is effectively zero. There is always a risk that a strain will escape a laboratory by accident or theft. Alternative methods are likely faster at finding these mutations. However, they don't prove a role in transmissibility, only suggest it, but that disadvantage is more than offset by the fact that risk is essentially eliminated. There are a number of publications that compare gain-of-function methods with alternative methods (for instance, here, here, here, and here[6]).

I would judge the alternative methods "equally efficacious;" however, in general, this judgement should be a consideration of the reviewing committee on a case-by-case basis, not stated as an absolute Principle. The words "equally efficacious" should be deleted. Principle iv would then read: "There are no feasible, alternative methods to address the same scientific question in a manner that poses less risk than does the proposed approach.

Some final thoughts

To be kept informed of decisions, an appropriate Congressional Committee or Caucus will be notified of the final decisions and explanations. The Congressional Biomedical Research Caucus[7] is perhaps the appropriate congressional group to keep informed.

Completion of the NSABB deliberative process should not mean the funding pause should be lifted. All studies subject to the funding pause should remain unfunded by the NIH until a new review process, such as that proposed here, is put in place and new reviews are carried out for all existing studies of concern.

The U.S. government should also consider pausing all studies of concern regardless of funding source until they are reviewed again. The many scientists who signed the Cambridge Working Group statement feel that studies such as these should be "curtailed" until further reviewed:

> "For any experiment, the expected net benefits should outweigh the risks. Experiments involving the creation of potential pandemic pathogens should be curtailed until there has been a quantitative, objective and credible assessment of the risks, potential benefits, and opportunities for risk mitigation, as well as comparison against safer experimental approaches."

In order to maintain continuity in the review and decision-making process, civil service employees should manage that process, as most political appointments will change with changing elected officials.

This proposal does not address the dual-use concern that someone will repeat the research for hostile purposes.

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[1] Joseph Kanabrocki May 24 presentation titled: *NSABB Working Group Draft Report: Recommendations for the Evaluation and Oversight of Proposed Gain-of-Function Research*

[2] ibid

[3] Called Federal review by the National Science Advisory Biosecurity Board. Federal review is likely review by the NIH Recombinant DNA Advisory Committee (RAC) or the NIH Office of Biotechnology Activities (OBA). It may also include review by the Department of Health and Human Services (HHS).

[4] An additional level of biosafety -- call it BSL-4-plus -- that adds special protections for laboratory work with dangerous PPP research. BSL4+ differences from BSL4 include (1) Train full-time technical staff who are dedicated to working with highly dangerous pathogens. These staffers would carry out experiments directed by scientists who would never need to be present in the BSL-4+ laboratory. With modern audio-video technology, research scientists can remotely monitor lab work as if they were present. (2) Require lab staffers to follow up extended work shifts with periods of quarantine before they leave the biocontainment area. Such procedures would assure that no potential pandemic pathogen escapes from a BSL-4+ lab through a laboratory-acquired infection; anyone accidentally infected would show symptoms while still in quarantine.

[5] Joseph Kanabrocki May 24 presentation *op. cit.* titled: *NSABB Working Group Draft Report: Recommendations for the Evaluation and Oversight of Proposed Gain-of-Function Research*

[6] M Lipsitch, *Comment on "Gain-of-Function Research and the Relevance to Clinical Practice"*, J Infect Dis., in press.

[7] The Congressional Biomedical Research Caucus (CBRC)…is a bipartisan, bicameral Caucus…Seventy five Members of the House of Representatives and nine Members of the Senate comprise the Caucus Membership...
The Caucus seeks to support the excellent efforts of the congressional committees and Members of Congress with jurisdiction over the National Institutes of Health (NIH), the National Science Foundation (NSF), science research, and health issues.