

Neoproterozoic 'snowball Earth' glaciations and the evolution of altruism

R. A. BOYLE, T. M. LENTON AND H. T. P. WILLIAMS

School of Environmental Sciences, University of East Anglia, Norwich, UK

ABSTRACT

We hypothesize that a demographic and ecological effect of Neoproterozoic 'snowball Earth' glaciations was to increase the fitness of group-level traits and consequently the likelihood of the evolution of macroscopic form. Extreme and repeated founder effects raised genetic relatedness – and therefore the influence of kin selection on the individuals within a group. This was permissive for the evolution of some highly costly altruistic traits, including those for macroscopic differentiation. In some eukaryotic species, the harsh and fluctuating abiotic conditions made a macroscopic physiology advantageous, perhaps necessary, for collective survival. This caused population-wide group viability selection, whereby non-altruist 'cheat' genotypes killed the groups they were in, and therefore themselves, by reaching fixation. Furthermore, dispersal between refugia would reach zero under anything near a 'hard snowball', which would protect altruists at high local frequency from the influx of cheats from neighbouring groups. We illustrate our hypothesis analytically and with a simple spatial model. We show how removal of between-group dispersal, in a population with initial between-group variation in cheat frequency, causes the relative frequency of altruists to increase while the population as a whole decreases in size, as a result of group death caused by cheat invasion. This may be of particular relevance to animal multicellularity because irreversible differentiation (highly altruistic in that it imposes a high fitness cost on the individual cell) is more prevalent than in other multicellular eukaryotes. The relevance of our hypothesis should be scaled by any future consensus on the severity of snowball Earth, but it is theoretically plausible that global-scale glaciations had a systematic influence on the level of selection during Earth history.

Received 12 March 2007; accepted 16 May 2007

Corresponding author: R. A. Boyle. Tel.: 01603 591373; fax: 01603 591327; e-mail r.boyle@uea.ac.uk.

INTRODUCTION

The Neoproterozoic era saw the most recent examples of global-scale glaciation, followed by a qualitative increase in the taxonomic abundance and morphological diversity of macroscopic life during the Ediacaran period (Vincent *et al.*, 2004; Corsetti *et al.*, 2006; Knoll *et al.*, 2006; Butterfield, 2007). The possibility of a causal connection between the Neoproterozoic glaciations and the evolution of the biosphere has been discussed for some time (e.g. Hoffman *et al.*, 1998; Hedges, 2004; Vincent *et al.*, 2004) but has lacked a mechanistic evolutionary basis. Similarly, a major gap in the 'snowball Earth' hypothesis (Kirschvink, 1992; Hoffman *et al.*, 1998; Hoffman & Schrag, 2002) is its failure to explain how large-celled, and potentially macroscopic eukaryotes could survive such an extreme climatic event. Many authors have used the assumption that the effect of a complete 'hard' snowball Earth on life would have been intolerably harsh, as part of the motivation for investigating partial ice cover 'slushball' solutions (e.g. Pollard & Kasting,

2005). But we argue herein that rather than presenting a paradox concerning how 'complex' life could have survived a snowball Earth event, the demographic and ecological impact of a global-scale glaciation strongly favoured the evolution of group-level traits. Small, highly genetically related populations largely isolated from each other would have resulted in strong kin selection, relatively favourable to the costly 'altruistic' life-history strategies required for macroscopic differentiation. We argue that the 'bottleneck and flush' style of evolution suggested as part of the original snowball Earth hypothesis (Hoffman *et al.*, 1998) is entirely consistent with the levels of selection problem, and that causality between snowball Earth and the emergence of highly costly macroscopic form should be taken seriously as a hypothesis.

'Snowball Earth' as a geological pattern

The idea of glaciation at or near global in its spatial extent is not new (Harland & Rudwick, 1964), but has received recent

attention via the ‘snowball Earth hypothesis’ championed by Hoffman *et al.* (Kirschvink, 1992; Hoffman *et al.*, 1998; Hoffman & Schrag, 2002). This hypothesis describes how glaciation supposedly results from extensive CO_{2(g)} drawdown by weathering of low-latitude continents, and eventually ends through CO_{2(g)} build up from tectonic outgassing. Globally synchronous glacial tillites with primary equatorial remnant magnetism occur in three intervals during the late Proterozoic era at 710, 635 and 580 million years ago (Ma) (Evans, 2000). These date estimates are imprecise, ranging by 5–10 million years (Myr) or more, with estimates frequently overlapping within analytical error (Fanning & Link, 2004). Estimated duration of the glacial period is similarly imprecise, but in the region of 6–12 Myr (Hoffman & Schrag, 2002). Some glacial tillites contain incompletely oxidized ‘banded’ iron formations, invoking deep ocean anoxia and therefore an ocean at or near isolation from the atmosphere through surface sea ice cover. Most are overlain with metre-thick ‘cap’ carbonates, suggesting sudden exposure of the ocean to an atmosphere extremely high in CO₂ at the end of the glaciation. The carbon isotope ¹³C is enriched prior to some glacial periods then dramatically depleted during and immediately after (e.g. Kennedy, 1996; Misi & Veizer, 1998; Kennedy *et al.*, 2001; Rothman *et al.*, 2003) with an equivalent pattern in ³⁴S (Hurtgen, 2002, 2005) – suggesting a crash in global productivity. Alternative models used to explain the palaeomagnetic evidence include high orbital obliquity (Meert & Torsvik, 2004) and true polar wander (Kent & Smethurst, 1998) but are much less parsimonious than the idea of global-scale glaciation in integrating many of the other separate lines of evidence (Evans, 2000, 2003, 2005). The latitudinal extent of ice progression remains equivocal. Glacial scouring – therefore hydrological activity (Rice & Hofmann, 2000; Hoffman & Schrag, 2002) has been used to invoke ‘slushball’ solutions with some open tropical oceans (Hyde *et al.*, 2000; Baum & Crowley, 2001; Peltier *et al.*, 2004; Pollard & Kasting, 2005), but is not necessarily impossible under a ‘hard snowball’ with a minimal hydrological cycle including sublimation (Baum & Crowley, 2003; Pierrehumbert, 2005). The incompletely oxidized iron sediments and cap carbonates argue for an ocean that is highly anoxic and largely sealed from the atmosphere.

A change in evolutionary tempo?

Biomarkers suggest the possible existence of eukaryotes as far back as the late Archaean (Brocks *et al.*, 1999), and multicellular eukaryotes can be identified from diagnostic cell-division patterns, and later macroscopic form, from the Mesoproterozoic onward (Hofmann, 1990; Javaux *et al.*, 2003; Butterfield, 2004, 2005a,b, 2007; Knoll *et al.*, 2006). But there are no macroscopic animals prior to the late Neoproterozoic. The Mesoproterozoic 1800–850 Myr eukaryotic fossil record documents the divergence of major eukaryotic clades, but exhibits increasing but relatively low taxonomic abundance

and little or no macroscopic differentiation (Hofmann, 1990; Knoll *et al.*, 2006; Huntly *et al.*, 2006). During the Cryogenian period 850–635 Ma encompassing the two most severe glaciations, acritarch taxonomic diversity crashed, before peaking sharply after the period’s ending (Knoll, 1994; Knoll *et al.*, 2006; Grey *et al.*, 2003; Narbonne, 2005; Huntly *et al.*, 2006). The subsequent early Ediacaran period 632–560 Ma exhibits a marked increase in morphological diversification in algae (Xiao, 2004a,b), (potential) animals (Xue *et al.*, 1992; Li *et al.*, 1998; Grotzinger *et al.*, 2000; Narbonne & Gehling, 2003), and unassigned macroscopic eukaryotes (Knoll *et al.*, 2006; Butterfield, 2007). Subsequent radiation of the Ediacaran biota 575–542 Ma gave rise to the earliest examples of macroscopic differentiation into what are discernably organs (Xiao, 2004; Narbonne, 2005), prior to the extinction of the group during the rise of the Eumetazoa in the Cambrian explosion. Molecular clock studies have consistently found early divergence times for eukaryotic phyla since their inception (Brown *et al.*, 1972; Feng, 1997; Wang *et al.*, 1999; Heckman *et al.*, 2001; Hedges *et al.*, 2001; Hedges, 2004). With some exceptions (Peterson *et al.*, 2005; Peterson & Butterfield, 2005), molecular clock studies tend to place the origin of major animal clades significantly earlier than does the fossil evidence. This is despite the fact that molecular clocks’ largest consistent source of potential error, ‘long branch attraction’ (the probability of saturated therefore undetectable point mutation increasing in proportion to the divergence time between the taxa), makes them more likely to underestimate divergence times than overestimate them (Page & Holmes, 1996). It is therefore reasonable to suppose that Ediacara and animals were present before the end of the glacial events in microscopic forms, but some change occurred during and after the glaciations to cause the radiation in size and differentiation seen in the Ediacaran fossil record. It is notable that the increase in acritarch diversity coincides with the similar phase change in evolution of the Ediacara. Similar patterns in different taxa invoke a common ecological cause. It is clear that eukaryotic differentiated multicellularity was present in algae and fungi significantly before any glacial event (Butterfield, 2000, 2005a, b). Therefore it is incorrect to think of global-scale glaciation as having been a necessary condition for the evolution of multicellularity or architecturally complex form. But after the glacial events, macroscopic form unequivocally became more taxonomically abundant and morphologically diverse. And we think it is also simplistic to assume that climatic events on the scale of the Neoproterozoic glaciations did not have a systematic influence on evolution.

Cooperation and altruism

There exist many circumstances in which a population is better off collectively if each individual produces a costly extracellular substance that is available to the whole local population (e.g. a bacterial siderophore) (West *et al.*, 2006). The short-term

fitness benefit an individual genotype can gain by forgoing this cost and investing extra effort in its own reproduction, but benefitting from production of the substance by its neighbours, increases its relative frequency – and therefore limits the level of the costly substance. This exemplifies the ‘tragedy of the commons’ that the average fitness of the population is lower than that of a population in which all members acted altruistically and produced the costly substance. The definitive link that such ‘public goods’ traits have with cell density and group phenotypes means that understanding when such costly traits are adaptive is important in describing the origins of multicellular and macroscopic form.

Natural selection frequently operates hierarchically on large groups of replicators (like multicellular organisms) that are composed of smaller individual replicators (like cells) (Michod, 1997). Many traits have an evolutionary impact on both the bearer and the individuals with which the bearer interacts – such traits are referred to as ‘social’. Often, the trait value that maximizes the fitness of the bearer is different to the trait value that has the greatest positive impact on other individuals – such as when genes for unrestricted cell growth lead to tumours in animals and plants. This leads to a tension between conflicting selection pressures at the individual and at the trait group levels. A trait that increases the fitness of the individuals with which the bearer interacts is termed cooperative, and a cooperative trait that imposes a fitness penalty on the bearer is termed altruistic (e.g. Lehmann & Keller, 2006), though of course no sentence is implied. If an individual interacts with an altruist, it receives a benefit, if it interacts with a non-altruist ‘cheat’ individual, it receives nothing and may experience decreased fitness, depending on its own genotype. Whether or not altruistic traits are adaptive depends on the nature of the relationship between the focal ‘actor’ individual bearing the trait, and the ‘recipient’ individual experiencing the trait’s results. There are a number of scenarios that can give rise to cooperation, all involving some constraint on the ratio b/c , of fitness benefit to recipient b , and fitness cost to altruist c . Each scenario involves some measure of the probability that an altruistic strategy will encounter and/or cause benefit to, a copy of itself. Different circumstances that can give rise to evolutionarily stable cooperation are (from Nowak, 2006):

(a) Kin selection ($b/c > 1/r$) (Hamilton, 1964). This process describes the effect on the fitness of an altruistic gene of the probability that the recipient of altruism also carries a copy of that gene. This probability is termed relatedness, r , but is defined specifically with respect to the focal locus and measures the probability of a correlation in trait values – which is not necessarily identical to a correlation in descent. Kin selection is normally referred to in the context of ‘Hamilton’s rule’ $rb > c$ (Hamilton, 1964, 1972). The conditions under which kin selection is relevant to a given trait depend on the balance between kin selection and kin competition, and consequently on the demographics – other more complex forms of Hamilton’s rule have been derived to incorporate the influence of

population viscosity under spatially continuous conditions (e.g. Van Baalen & Rand, 1998), as well as varying ploidy and sex ratios (Hamilton, 1972).

(b) Group selection (e.g. Sober & Sloan-Wilson, 1998). This is a means of expressing the way in which natural selection operates hierarchically on group replicators (such as multicellular organisms) that are themselves composed of smaller individual replicators (such as cells). In circumstances where trait values that increase the short-term fitness of the smaller replicators simultaneously decrease the long-term fitness of the larger replicators of which they are a part, then evolution of cooperation requires $(b/c) > [1 + (n/m)]$, where n is the maximum number of individuals per group, and m is the number of groups in the higher level population (Traulsen & Nowak, 2006). This condition arises from the assumption that a group of altruists will either split more frequently (‘group fecundity selection’) or have a higher probability of survival (‘group viability selection’) than will a group of cheats. It is arguable that in nature the probability of two individuals sharing a gene is reasonably approximated by $r \equiv [m/(m+n)]$ – i.e. that kin selection is an explicit genetic description of one type of group selection (Frank, 1998; Nowak, 2006).

(c) Reciprocity (Trivers, 1971). If an altruistic act by an individual increases the probability of that individual being the recipient of altruism, the trait may proliferate. Direct reciprocity requires $(b/c) > (1/w)$, with w , the probability of a repeated social interaction between the same two individuals, necessary to scale the mutual benefit from an interaction between two altruists by the probability that this interaction will happen again (Trivers, 1971; Axelrod & Hamilton, 1981). Reciprocity may also occur in other contexts, including a spatial framework $(b/c) > k$, where k is the average number of neighbours per individual (Nowak & May, 1992).

A MECHANISTIC HYPOTHESIS

We hypothesize that global-scale Neoproterozoic glaciations caused unicellular eukaryotic individuals to experience high within-group relatedness and therefore strong kin selection ((a) above). Because between-group dispersal was at or near zero on a planet covered with ice, cheats could not spread over the population and individual strategies involving kin competition had a reduced fitness payoff compared to non-glacial conditions. The benefit of altruistic strategies was increased by the need for resource sequestration and accentuating disequilibrium with the environment, which (we think) is more efficient with the division of labour possible with macroscopic, organ-level differentiation (Boyle & Lenton, 2006). After reaching or approaching fixation by local kin selection, such multicellular-scale physiology had consequences that made groups of altruists significantly more likely to survive the harsh abiotic conditions than groups of cheats – causing strong between-group viability selection (encompassed within (b), above). Therefore by reaching local fixation, cheat

genotypes caused the group they were within to experience dramatically curtailed growth, or (more likely) dramatically lower survival probability. Snowball Earth thus gave rise to a uniquely strong sampling process favouring altruism, of kin selection within groups and group viability selection between groups, a process that operated over a geological timescale and a global spatial scale.

Relatedness and founder effects

Snowball Earth would decimate the mass of the biosphere and scatter remaining populations into isolated refugia, in which individuals were descended from a number of 'founder' individuals dramatically fewer than under non-glacial conditions. Fluctuation in hydrological and temperature conditions would shorten the growth season and make resource availability erratic – analogous to modern arctic biomes (e.g. Vincent *et al.*, 2004). This would cause dramatic, repeated founder effects on a global spatial scale and a multimillion year temporal scale, leading to small, highly related populations with little or no dispersal between them. The promotion of the evolution of altruistic traits by repeated founder effects has been noted before (Cohen & Eshel, 1976; Eshel, 1977), though without tangible examples with sufficiently long duration and large spatial extent to have had an important influence on the history of life. In general, increasing demographic mobility (i.e. immigration and emigration) has long been appreciated to impede the evolution of altruism by reducing the probability that the recipient of altruism also carries the altruistic gene (Wright, 1945; Eshel, 1972; Fix, 1985; Van Baalen & Rand, 1998). The amount of immigration required to hinder the evolution of altruism is negatively related to the size of the group (Cohen & Eshel, 1976). The biotic refugia present in a world nearly or entirely covered by ice would be small and intermixed with each other to a minimal extent, with complete between-group isolation from each other during a 'hard' (equatorial) snowball. Although the data do not allow us to specify a number of founder effect cycles, we think that uniquely high kin relatedness, fluctuating demographic conditions, and a spatial structure conducive to altruism are a reasonable description of the conditions prevailing on a planet with minimal biotic refugia interspersed with near global glacial cover.

The advantage of 'cheating'

Altruism can evolve in viscous spatial populations by kin selection (e.g. Van Baalen & Rand, 1998), however under some conditions kin competition can outweigh kin selection (e.g. Griffin & West, 2002). Consequently, highly related populations are not always synonymous with increasing altruism. The balance between the two processes depends on the details of the trait and of the species, and the benefit to be gained from a non-altruistic strategy in terms of extra

offspring. If productivity is low and chances of dispersal to a new group are at or near zero, then a unicellular eukaryotic genotype that does not contribute to any higher level physiology would not have significantly more offspring than the relatives with which it was competing, i.e. survival becomes a more important component of fitness than does fecundity. More importantly, any detrimental impact that non-altruist strategies had on the group might significantly decrease the survival probability of groups in which kin competition proliferated. Hence, we think that snowball Earth also reduced the fitness payoff for kin competition, swaying the balance of within-group evolution towards kin selection. In addition to processes sensitive to kin relatedness, reciprocal altruism is promoted by a small number of neighbours with low spatial demographic mobility (Nowak & May, 1992). Assuming the existence of some rudimentary cellular recognition mechanism, greatly reduced or absent between-group dispersal and persistence of life in small populations, would increase the probability of repeated interaction between the same individuals. This means that the kin selection processes discussed may have been complemented by an element of reciprocity. Furthermore, if altruism is necessary for group survival, then in order to survive, non-altruist strategies must either avoid reaching fixation, or disperse to a new group before the death of any group that they come to dominate. Building a macroscopic Ediacaran 'proto-animal' is a costly evolutionary innovation, probably requiring extensive terminal differentiation. In the majority of refugia, the ratio of the cost of altruism to the benefit would be too high, cheats would reach fixation, and the group would go extinct. But not in all cases. And removal of virtually all between-group dispersal by the glaciations would mean only those rare groups where altruism was adaptive would leave descendants. Consequently, the species-wide relative frequency of altruistic strategies increased over a global spatial scale and multimillion year timescale.

Selection pressure for group level differentiation

The very existence of bacterial 'public goods' such as siderophores demonstrates how a group of cells acting collectively can increase the average availability of a scarce resource through a scavenging behaviour linked to cell density. It is reasonable to suppose that the erratic, low-energy hydrological cycle, and dramatic decrease in terrestrial weathering occurring during a global-scale glaciation would have made virtually all resources scarce and available only erratically during, increasing the evolutionary incentive for such group-level traits. We have argued elsewhere (Boyle & Lenton, 2006) that the greater capacity of groups to undergo differentiation better predisposes them to tolerate fluctuation in the physical environment. Greater longevity and size makes groups more likely to encounter such fluctuation, giving rise to a positive feedback to exploit this potential to differentiate. I.e. under conditions likely prevailing on a snowball Earth, a macroscopic system of highly related

eukaryotic cells would increase the likelihood of its survival and reproduction by differentiation into organ structures that allowed it to (for example) sequester resources and maintain constant water potential. We think that such a structure would leave significantly more descendants than a collection of autonomous single cells of equivalent size. Similarly, fluctuating climatic conditions will actively select for a latent reproductive stage in order to survive periods of suboptimal environmental conditions. Spatial heterogeneity and extreme gradients in redox conditions, water potential, temperature and resource conditions would be prevalent on a snowball Earth. Hence we propose that during the Neoproterozoic glaciations, the imposition of confined but heterogeneous spatial structure, as well as harsh and fluctuating physical conditions, increased the fitness of differentiated group-level form with efficient division of labour. Within-group kin selection was therefore complemented by between-group viability selection for a division of labour efficient enough to survive the harsh conditions.

A SIMPLE MODEL

We now illustrate analytically the two distinct phases that we propose describe the influence of snowball Earth on the evolution of life-history strategies in relevant eukaryotic species:

Phase 1: Repeated founder effects increase relatedness, altruism becomes necessary for group survival

Let t_{crit} be a sufficient number of generations for altruism to gain a sufficient impact on fitness that a group filled with cheats will go extinct. For example, t_{crit} could be a sufficient number of generations (of a species of unicellular eukaryotes) over which Hamilton's rule continuously holds, for a costly altruistic trait for multicellular aggregation to change from having a positive but trivial collective fitness payoff, to having a qualitatively higher fitness payoff such that a group of cheats would be at an extreme fitness disadvantage. A useful comparison is that between an essentially autonomous unicellular species with facultative multicellular grouping, and a species with a multicellular physiology sufficiently coordinated and efficient that autonomous reproduction is either maladaptive or obsolete. We assume that such a physiology would impose extensive fitness penalties on individual cells, requiring many generations over which highly costly altruism was adaptive, in order to become established. We think this provided a constraint on the evolution of such traits. Extreme glaciations would force eukaryotic species into small populations experiencing an erratic, short growth season, fluctuating hydrological cycle and only brief intervals of habitability. Extreme and repeated founder effects drove up relatedness causing strong kin selection. Such conditions would have had to hold for the required t_{crit} generations. Let E be some representation of

the state of the abiotic environment that has influence on the fitness of altruistic traits. We lump all systematic change in the kin selection parameters over the focal time interval into the effect that the changing abiotic environment has on these parameters, so that (by the chain rule):

$$\frac{\partial}{\partial t}(rb - c) = \frac{\partial}{\partial E}(rb - c) \frac{\partial E}{\partial t} \quad (1)$$

Implicit in this formulation is our assumption that the strictly biotic constraints on the evolution of altruistic, macroscopic physiology (i.e. any constraint in terms of 'laws of form') did not systematically vary over the Neoproterozoic era (and in our opinion probably over all of Earth history) in any way that was not dictated by how different physiologies would fair in different abiotic environments. Certain physiologies could only evolve in abiotic environments that made their high cost an adaptive trade off. Recalling that even in a permissive environment, many (t_{crit}) generations would be needed for such a costly macroscopic physiology to become coordinated enough for this trade off to be achieved, we require the abiotic environment E to allow Hamilton's inequality to hold consecutively for this length of time:

$$\prod_{t=1}^{t_{crit}} \frac{1}{2} \left(1 + \frac{\int_0^t \frac{\partial E}{\partial t} \left(\frac{\partial}{\partial E}(rb - c) \right) \partial t}{\left| \int_0^t \frac{\partial E}{\partial t} \left(\frac{\partial}{\partial E}(rb - c) \right) \partial t \right|} \right) > 0 \quad (2)$$

Because snowball Earth operated over timescales unequivocally evolutionary (as opposed to ecological) in length, and because the dynamics of populations of proto-multicellular eukaryotes were likely orders of magnitude faster than their modern descendants, we think that this would have been possible during the Neoproterozoic glaciations. The requirement for altruism to become necessary for group survival is a prerequisite for the group viability selection we suggest below to be effective. But macroscopic form exists, multicellular animals and plants with excessive tumours die, so altruist frequency did gain such an influence at some point in Earth history. We think the radiation of the Ediacara immediately after a period demographically conducive to altruism is not merely coincidental. Finally, although ubiquitous small populations would increase the impact of genetic drift on evolution (e.g. Wright, 1945), this does not preclude an additional directional trend, particularly under uniquely high relatedness conditions. Macroscopic form involving terminal differentiation is a physiological elaboration on mechanisms of differentiation present in bacteria and more simple eukaryotes (e.g. Rainey & Rainey, 2003). We emphasize that we are not proposing any novel mechanism for the evolution of altruistic form, merely an abiotic environment that biased evolution towards strong kin selection and low kin competition, releasing an evolutionary constraint on highly costly group-level traits.

Phase 2: Complete removal of between-group dispersal – ‘cheats’ drive themselves extinct

If the presence of at least some altruists is a prerequisite for group survival, the non-altruist strategy that will leave most descendents is one that either remains at intermediate frequency (i.e. is further on the continuum towards altruism), or one that migrates to a new group before the death of the current one. An example of this trade off is disease – virulence is a strategy that gives a pathogen high fitness only in circumstances in which dispersal to a new host is possible before the death of the current one (e.g. Day, 2003). In a situation in which dispersal between individual groups is not possible – and, simultaneously the presence of altruists is a prerequisite for survival, cheat genotypes that reach fixation will die and only those that are less harmful to the group and/or remain at intermediate frequency will leave descendents. For example, if altruism constitutes storage of a resource of which there is no external supply over significant periods of time, then a cheat genotype that consumes the resource without sequestering it will out-compete an altruist genotype that sequesters a fraction of the resource available to it. But once the cheat genotype reaches fixation the resource supply will be exhausted and the whole group will die. A similar argument might be made for a patch of cells that required the presence of some altruistic terminally differentiated cells to form a boundary against a harmful external environment (such as one of fluctuating osmotic conditions). Without the possibility of between-group dispersal, the death of all such altruists because they had been outcompeted by cheats would also constitute the death of the cheats. All macroscopic, multicellular form requires such altruistic strategies on the part of its constituent cells. And we think it is a reasonable assumption that emigration would be dramatically curtailed if not absent in a world in which life was persisting in scattered refugia with the intermediate space nearly or entirely covered with ice.

Spatial implementation

A species exists in a population separated into G total groups. Each group has k neighbour groups, to which members of that group send offspring with probability D per individual, per generation. We are interested in the way in which the j th genotype changes over time, because we will later label j as either altruist A or cheat S . The j th genotype has fitness F_j , by which the genotype’s frequency changes by each timestep, a starting frequency of N_{j0} and changes over time according to:

$$\frac{dN_j}{dt} = \frac{d}{dt}(N_{j0}F_j^t) = N_{j0}\ln(F_j)F_j^t \frac{dF_j}{dt} \quad (3)$$

The change in the genotype’s frequency within a group (dN_j/dt)_{group} is dictated by the interaction between those

individuals already present (the within-group dynamics) and the contribution of movement in and out of the group (the between-group dynamics). The latter contribution is denoted by the operator $\partial/\partial k$:

$$\frac{dN_j}{dt}_{group} = \left(1 + Dk \frac{\partial}{\partial k}\right) \frac{dN_j}{dt} \quad (4)$$

The total change in the genotype’s frequency (dN_j/dt)_{total} across the whole population (i.e. over all G groups) is the sum of the changes in each group:

$$\frac{dN_j}{dt}_{total} = \sum_g \left(1 + Dk \frac{\partial}{\partial k}\right) \frac{dN_j}{dt}_g \quad (5)$$

For steady state frequency of the j th genotype, over the whole population of G groups (the change over which is denoted by the operator $\partial/\partial G$):

$$\frac{dN_j}{dt}_{total} = G \frac{\partial}{\partial G} \left(1 + Dk \frac{\partial}{\partial k}\right) \frac{dN_j}{dt} = 0 \quad (6)$$

There are four ways that this steady state for the j th genotype (dN_j/dt)_{total} = 0 can happen.

- (1) Total extinction $G = 0$. Although under a global glaciation, this will encompass the majority of cases, it is not interesting to us because the species to which it happened did not leave descendents.
- (2) No neighbours $k = 0$ and/or dispersal probability $D = 0$, without total extinction. As described, we think that a situation at or approaching a ‘hard’ snowball Earth removed between-group dispersal D and thus gave rise to the rare if not unique situation that the overall dynamics were the sum of the within-group dynamics across all groups, without any immigration or emigration:

$$\frac{dN_j}{dt}_{total} = \sum_g \left(1 + Dk \frac{\partial}{\partial k}\right) \frac{dN_j}{dt}_g = \sum_g \frac{dN_j}{dt}_g \quad (7)$$

- (3) A global viscous population with the same change in genotype frequency over all groups:

$$\frac{\partial}{\partial G} \left(1 + Dk \frac{\partial}{\partial k}\right) \frac{dN_j}{dt} = 0 \quad (8)$$

We do not think that this happened during the Neoproterozoic glaciations, when the world was covered with ice. Even populations in close proximity would be confined to (for example) springs derived from separate and confined sources of volcanic heat, therefore not spatially continuous.

- (4) A viable population within each group, with non-zero neighbours $k \neq 0$ and non-zero dispersal probability $D \neq 0$, experiencing a balance between the within- and between-group dynamics:

$$\left(1 + Dk \frac{\partial}{\partial k}\right) \frac{dN_j}{dt} = 0 \quad (9)$$

This describes a more comprehensive equilibrium encompassing a balance of between-group and within-group contributions to frequency change, an equilibrium relevant to most contemporary levels of selection problems. While we think that situation (2) is most relevant to describing the impact of snowball Earth, (4) may also have briefly applied in some species during the Neoproterozoic glaciations.

Recall that we are using the j th genotype N_j to denote either altruists N_A or cheats N_S . We want to write the fitness of these genotypes in terms of the relative frequency of altruists N_A , the benefit b to all members of a group (of total size $N_t = N_A + N_S$) due to the presence of altruists, and the cost c of altruism, that altruists incur per generation, but that cheats avoid. We therefore define the fitness of altruists and cheats respectively as:

$$F_A = \left(\frac{N_A}{N_A + N_S}\right) b - c \quad (10)$$

$$F_S = \left(\frac{N_A}{N_A + N_S}\right) b \quad (11)$$

Balance between within- and between-group dynamics implies $[1 + Dk(\partial/\partial k)](dN_A/dt) = [1 + Dk(\partial/\partial k)](dN_S/dt) = 0$. Therefore, an equilibrium involving both within- and between-group processes, persisting over some arbitrary time interval $t_{end} - t_{start}$ implies:

$$\int_{t_{start}}^{t_{end}} \frac{dN_A}{dt} - \frac{dN_S}{dt} dt = \int_{t_{start}}^{t_{end}} Dk \frac{\partial}{\partial k} \left(\frac{dN_S}{dt} - \frac{dN_A}{dt} \right) dt \quad (12)$$

With (12) we can examine the implications of this dynamic equilibrium for the cost c and benefit b parameters, as well as the starting frequencies of altruists N_{A0} and cheats N_{S0} . It is useful to note that the relative frequency of altruists within the group, which appears in both fitness definitions (10) and (11) is equivalent to the average relatedness between the altruist and the potential recipient of altruism as defined with reference to kin selection – i.e. $N_A/(N_A + N_S) = r$. Re-labelling in this way, and substituting from (3), (10) and (11), the definite integral (12) implies:

$$[N_A - N_S]_{t_{start}}^{t_{end}} = \left[Dk \left(r b^t \frac{\partial N_{S0}}{\partial k} + N_{S0} \frac{\partial}{\partial k} (r b^t) - (r b - c)^t \frac{\partial N_{A0}}{\partial k} - N_{A0} \frac{\partial}{\partial k} (r b - c)^t \right) \right]_{t_{start}}^{t_{end}} \quad (13)$$

This means that over a steady state interval $t_{end} - t_{start}$, the initial within-group difference in genotype frequency between altruists and cheats $N_A - N_S$, must be balanced by a

between-group component of that difference, with effect on both the initial frequencies and (probably to a lesser extent) the cost–benefit parameters themselves. As we have discussed, selection pressure for differentiation into (for example) organs for resource sequestration in the harsh abiotic conditions would have driven up the benefit b for such altruistic traits, and high founder effects would have dramatically driven up r . But snowball Earth would have been most significant in its effect on between-group dispersal probability, an issue we now illustrate.

Numerical simulations

Consider a system of $G = 100$ groups, each with $k = 2$ distinct neighbouring groups, using the parameters benefit $b = 1$ and cost $c = 0.1$, and with relatedness defined by the varying within-group altruist relative frequencies $N_A/(N_A + N_S) = r$. Within each group, the frequency of a genotype at one generation is the product of its frequency and its fitness at the previous generation. The fitness values of altruists and cheats were of the form of (10) and (11), respectively; i.e. altruists could potentially experience kin selection, but relatedness could be ‘diluted’ by the reproduction of unrelated cheat individuals that benefited from altruism but avoided the cost. Altruism was assumed necessary for group survival – and groups in which cheats reached fixation went extinct. Each group had a carrying capacity of 250 individuals, and was initialized with 25 individuals at the start of each simulation. An individual could potentially produce offspring both within its own group and in each of its neighbours, the latter according to the value of the reproductive dispersal probability D , per individual, per generation. The results we present refer to the end of a 200-generation simulation for various values of D . A group at its carrying capacity could not receive offspring from neighbouring groups, and an empty group was considered dead and therefore unsuitable for receiving offspring from adjacent groups. Genotype frequency at a given timestep was the product of fitness and genotype frequency at the previous timestep. For non-zero dispersal probability D , this frequency contained a within-group component, and an immigration component from each of the two neighbour groups. So in the G th group, with two neighbouring groups $G + 1$ and $G - 1$, from which individuals could add offspring according to reproductive dispersal probability D , at the t th generation, the number of altruists $N_{A,t,G}$ was:

$$N_{A,t,G} = \left[\left(\frac{N_{A,t-1,G}}{N_{A,t-1,G} + N_{S,t-1,G}} \right) b - c \right] \left[N_{A,t-1,G} + D \cdot (N_{A,t-1,G+1} + N_{A,t-1,G-1}) \right] \quad (14)$$

With the first square bracketed term being altruist fitness within the G th group, as defined in (10), the second bracketed

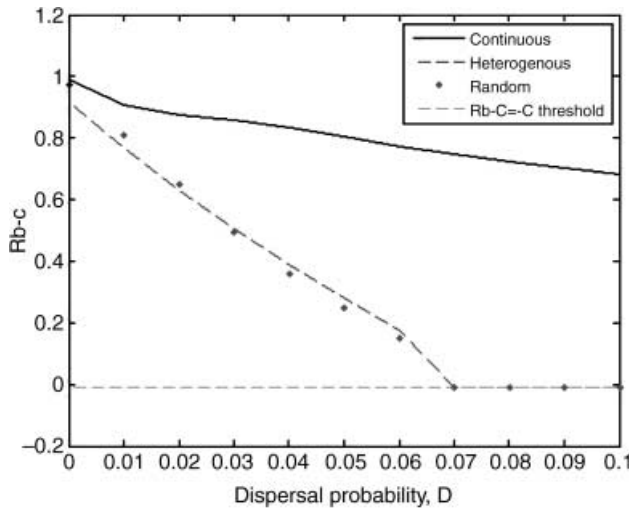


Fig. 1 Increased mixing between groups reduces the probability of Hamilton's condition being realized over a range of initial between-group genotype frequencies. *y*-axis shows $rb - c$ using relatedness $r = N_A / (N_A + N_C)$ averaged over the whole population. 'Continuous' (solid line) means each group has similar initial genotype frequency to its two neighbours, 'heterogeneous' (dashed line) means groups initialized at high relative altruist frequency are adjacent to groups initialized at low altruist frequency, 'Random' (dotted line) refers to (pseudo)random initial frequencies of altruists in each group. Line marked with squares shows threshold of zero benefit for altruism. Note that the continuous experiment shows a lower sensitivity to low but non-zero between-group dispersal, because it takes longer for cheats to spread across the population.

term being the frequency in the focal group and the neighbouring groups, with the latter scaled by dispersal probability D . The cheat genotype benefited from the presence of altruists according to their density, but lacked the fitness cost:

$$N_{S_{t,G}} = \left[\left(\frac{N_{A_{t-1,G}}}{N_{A_{t-1,G}} + N_{S_{t-1,G}}} \right) b \right] \cdot \left[N_{S_{t-1,G}} + D \cdot (N_{S_{t-1,G+1}} + N_{S_{t-1,G-1}}) \right] \quad (15)$$

Of course, the results from any such spatial set up depend on the starting frequencies. We present results from three initializations: (a) Continuous linear variation in initial altruist frequency, so that the first group began with 1% altruists, the second 2% etc. This caused relative genotype frequencies in neighbouring groups to be similar. (b) Heterogeneous non-random variation in initial frequencies, with the relative frequency of each genotype negatively correlated with that of neighbouring groups. This was achieved with the (admittedly arbitrary) set up of the first group 1% altruists (therefore 99% cheats), the second group 98% altruists and 2% cheats, the third 3% altruists and 97% cheats, and so on. (c) Random initial altruist frequency (with cheat frequency 1 - altruist frequency as above).

Figures 1 and 2 show the results for various dispersal probabilities D . Figure 1 shows the final value of Hamilton's condition $rb - c$. This approaches 1 for zero between-group

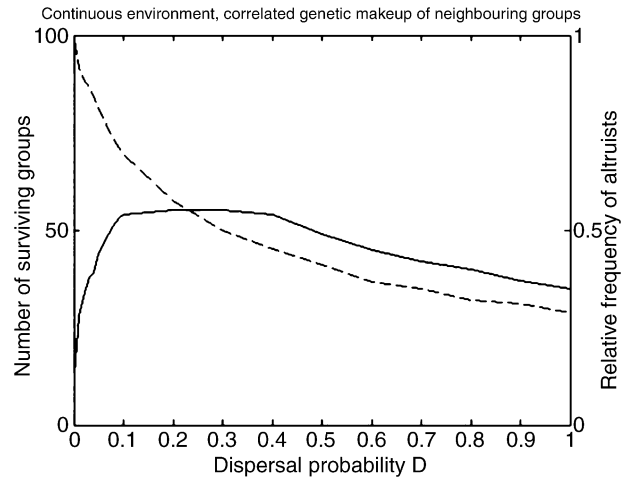


Fig. 2 Low between-group dispersal probability perpetuates spatial variation in genotype frequencies, protecting altruists from cheats in neighbouring groups. Right *y*-axis (dashed line) shows final relative altruist frequency averaged over the whole population, left *y*-axis (solid line) shows the number of groups from the original 100 surviving after the 200 generation simulation in which initial relative genotype frequencies were correlated between adjacent groups. At zero dispersal probability, most groups go extinct as cheats reach local fixation, but in the small remaining population, altruists dominate. As the population becomes more continuous, a larger number of groups survive but cheat frequency becomes higher, until at around $D = 0.3$, cheat fixation starts to decrease the size of the population despite the capacity for reproduction in neighbouring groups. Nonetheless, it is clear that altruists at relatively high local frequency are favoured by extremely low and zero dispersal values (see text).

dispersal but rapidly declines due to incremental cheat invasion when between-group dispersal is at positive values. This decline is sharpest in scenarios (b) and (c), in which groups with high relative altruist frequency are neighbored by groups filled with cheats, and less sharp in conditions of continuous gradation over the population (a). The reason that $rb - c$ is able to reach such high values for $D = 0$ or extremely low-dispersal probabilities rests in the variable population size (itself derived from our assumption of non-zero altruist frequency being a prerequisite for survival), and is illustrated in Fig. 2. At zero and extremely low-dispersal probabilities, population size drastically declines (around five groups from the starting 100), because groups the individuals within in which cheats reach fixation die without the opportunity to re-disperse, and the only remaining groups are those with high altruist frequency. As dispersal increases slightly, population size increases, as cheats reach higher relative frequency across the population but reach local fixation less frequently. At dispersal probabilities of around $D = 0.3$ and higher, population size again becomes limited as cheats reach local fixation in a wider number of groups. We emphasize that this model is only intended to be illustrative. The important point is that in a system with within-group kin selection, and group viability selection operating between groups, complete or near complete removal of between-group dispersal strongly favours altruists.

DISCUSSION

The temporal correlation between the glaciations and the emergence of complex animal life has been commented on repeatedly (Hedges, 2004; Butterfield, 2007; Canfield *et al.*, 2007). Butterfield (2007), for example, attributes diversification of the post-Ediacaran biosphere to a restructuring of macroecology caused by the introduction of the Eumetazoa, but does not address the reason for Ediacaran diversification before the Cambrian explosion. Canfield *et al.* (2007) attribute diversification of aerobic animal metabolism to increased photosynthetic O₂ production after the Gaskiers event, citing as evidence decreased reactive: total Fe, and increased ³⁴S fractionation after termination of the glacial deposits. Increased atmospheric oxygen will indeed cause aerobic heterotrophic metabolism on a macroscopic scale to become more adaptive, and might arguably have been a necessary condition for the radiation of the Eumetazoa, as is popularly acknowledged (e.g. Lenton & Watson, 2004; Canfield *et al.*, 2007). But macroscopic metazoan forms are necessary in the first place for evolution to alter their physiology. The capacity of a multicellular (proto) animal to differentiate in a coordinated, organ-scale manner is necessary before natural selection can make the oxygen assimilation of such an organism more efficient. Increased atmospheric oxygen will not give rise to this capacity to differentiate, therefore was not a sufficient condition for the evolution of macroscopic animals. Similarly, Baker (2006) argues that the impact of climatic stress on the 'heat shock' chaperone protein HSP-90 caused increased expression of latent genetic diversity in animal signal transduction pathways, increasing the rate of the evolution of developmental complexity and contributing to the Cambrian explosion. This hypothesis may also be true, but requires a level of pre-existing developmental complexity in order for evolution to iterate on a theme. The existence of such potential for developmental complexity is, we think, the pattern that requires an explanation. The mechanism we present herein describes the broader, more systematic influence that we think global-scale glaciation would have had on the evolution of physiology and form across many taxa.

Requirements for evolution of multicellular and macroscopic form

The emergence of multicellular form requires separation of a germ line and a soma, exploiting the evolutionary advantages of multicellularity by top-down physiological control probably requires sexual reproduction (Wolpert, 1994; Maynard-Smith & Szathmary, 1994; Kerzberg & Wolpert, 1998; Wolpert & Szathmary, 2002). The first step towards the evolution of multicellularity requires formation of cell clusters (Kerzberg & Wolpert, 1998; Pfeiffer & Bonhoeffer, 2003). Cells on the exterior of such clusters are more likely to die in harsh environmental conditions, perhaps forming a protective barrier

between inner cells and the environment. The earliest germline-soma separation may have been into one cell line predisposed to phagocytose neighbouring cells, and one cell line significantly less predisposed to do so (Kerzberg & Wolpert, 1998). Once such a mutation occurs, those cells that phagocytose their neighbours will, of course, leave many more descendents. Any developmental separation between the two cell lines early in development of the cluster will result in the germ line accumulating significantly less mutations than the soma; hence mutations disruptive to the higher level structure will not be passed on to descendents. This feature, coupled with increased relatedness due to passing through the egg cell bottleneck each generation, means that once it arose, the germline-soma separation was probably self-sustaining (Wolpert, 1994; Kerzberg & Wolpert, 1998). But why should this separation have occurred in the first place? It is reasonable to suppose that it would be adaptive for cells on the interior of a cluster to phagocytose dead cells on the exterior that had died as a result of (for example) low resource availability, but why should the cells on the exterior not behave similarly before reaching the point of starvation? If the first eggs were cannibalistic, the germline-soma separation is best described as evolution of a somatic mutation for restraint from cannibalism. This is an extremely altruistic trait. Pfeiffer & Bonhoeffer (2003) show that spatial clustering may have promoted multicellularity in clusters of unicellular organisms with rudimentary cooperative behaviour, by reducing average probability of interaction with non-cooperators, but acknowledge that the reason for the existence of such clusters in the first place is unknown. Changes in the cell cycle giving rise to multicellularity have been attributed to unspecified ecological signals before (Wolpert, 1994). But there has been no systematic consideration of the types of trait made most adaptive by long-term abiotic pressures occurring at around the time of emergence of the earliest Metazoa. Regardless of the physiological basis for the first germline-soma separation, we think that the evolution of an altruistic soma is a major evolutionary step that remains incompletely explained. We propose that the first multicellular clusters of the Ediacara (and implicitly therefore, the ancestors of the Metazoa) were imposed on the biosphere by constraints on space and resource availability in the abiotic environment, and that extremely high within cluster relatedness resulted from strong founder effects – promoting the evolution of an altruistic soma through exceptionally effective kin selection. Subsequent elaboration of physiology by organ level differentiation was probably achieved by qualitative genomic changes in the regulation of development (e.g. Wilkins, 2002), many of which required costly terminal differentiation – further promoted by this persistently high relatedness, i.e. high relatedness within multicellular clusters of the first animals, as well as being the result of passing through an egg cell bottleneck, preceded and promoted the separation into a germline and a soma, as well as other costly group-level traits. Furthermore, the presence of highly related

multicellular clusters was not initially an adaptation, but was imposed by extreme abiotic pressures.

Falsifiability: analogues for a snowball Earth biosphere

Temporal resolution of both the geological evidence for glaciation and the biological evidence for macroscopic differentiation is too coarse for the idea we present here to be anything other than a hypothesis, the relevance of which should be scaled by any future consensus on the severity of the Neoproterozoic glaciations. The strength of the founder effects and dispersal restrictions that we propose would be greatest under a complete 'hard snowball', but still likely prevalent under a 'slushball' if the hydrological cycle caused isolation into refugia and decimated net productivity. We fully acknowledge that the only appropriate test is an empirical one – the presence of diverse macroscopic Ediacaran 'animal' forms with organ-level differentiation prior to the Sturtian glaciation at 710 Ma would, of course, refute our idea. But none have been found, despite multicellular forms in other taxa. Furthermore, the qualitative transition in altruism that we propose may in fact be an internal change – such as a highly costly change in cellular compartmentalization, gene duplication or resource storage, which only later gave rise to the macroscopic form for which it was necessary. A more realistic means of falsifying our idea may therefore be to examine the evolution of cooperative traits in existing extreme environments.

Modern Arctic and Antarctic bacteria and eukaryotes tolerate extreme temperature decreases and extreme osmotic fluctuation, and decrease the risk of mechanical damage through ice crystal formation by a range of cellular and biochemical strategies, frequently being capable of maintaining significant enzymatic activity at near freezing temperatures (Vincent, 1988; Deming & Huston, 2000; Rothschild & Mancinelli, 2001). Communities consisting of a range of microbial extremophiles and eukaryotic algae also persist within brine channels separating crystals of sea ice (Thomas & Dieckmann, 2002). A particularly useful analogue for the communities likely present during a snowball event is provided by that of the thick sea ice shelves of the modern Arctic and Antarctic. Most of the biomass in these communities is contained within microbial mats, dominated by prokaryotes but including micro-algae and some micro-invertebrates (Vincent *et al.*, 2004). A range of microcosms provide niches for halophilic and halophobic, aerobic and anaerobic microbiota. Algae and cyanobacteria exhibit extensive diversification in light-harvesting photosynthetic pigments as well as photoprotective pigments; matching the range of light intensities and wavelengths penetrating different depths in water bodies amongst and below the ice. Sediment communities form microbial 'ice mats' around the layers of gravel in the ablation zone – consisting of loosely bound communities of cyanobacteria and diatoms (Vincent, 1988; Hawes *et al.*, 1993). Formation and destruction of these gravel communities are frequent but erratic, matching

the dynamics of ice formation and melting. Population densities are low but highly variable – mirroring the erratic hydrological cycle.

The evolutionary implications of global-scale glaciation for communities of this sort have been considered before (Vincent *et al.*, 2004; Hedges, 2004). Nisbet & Fowler (1999), as well as Vincent *et al.* (2004), note that the steep redox gradients within communities below the ice could accentuate differentiation within microbial mats – suggesting that this factor, as well as the relative stability of extracellular DNA under the ice-cooled conditions, might have facilitated lateral gene transfer and symbiosis. Most consideration has been given to the Palaeoproterozoic glaciations – diversifying selection under conditions of low productivity, as well as strong chemical nutrient gradients, having been proposed as a mechanism for eukaryogenesis (Vincent *et al.*, 2004). However, to our knowledge, there has been no systematic consideration of the type of evolutionary strategy that is most adaptive in the extreme demographic conditions prevailing in these refugia. Spatial heterogeneity of the kind present in these biomes has been suggested to give rise to altruistic low growth rate, high growth yield strategies in bacterial biofilms – films with a lower average growth rate but a more coherent structure being able to achieve a larger size (Kreft, 2004). Imposition of spatial heterogeneity is sufficient to cause morphological differentiation in bacteria (Rainey & Travisano, 1998), including cooperative group level differentiation detrimental to individual short-term success (Rainey & Rainey, 2003). It is therefore reasonable to hypothesize that the extreme resource and oxidative gradients within such arctic environments may cause equally extreme differentiation in (say) bacterial colonies today. The mechanisms we suggest for eukaryotes would be merely an extension of this idea over a longer timescale and conditions of higher kin relatedness. Additionally, any similar event causing the decimation of the biosphere and isolation into distinct refugia could in theory increase the fitness of group level traits. If similar demographic conditions resulted, an analogous evolutionary effect of the Palaeoproterozoic Makganyene glaciation (Evans *et al.*, 1997), perhaps increasing the developmental complexity of (unicellular) eukaryotes, may also be plausible.

Specific influence on animal evolution?

Certain differentiated organ-level structures impose a qualitatively greater short-term fitness cost on the individual cell than multicellularity *per se*, because they involve a greater decrease in potency than does a trait causing (say) cellular adhesion, but permitting some retention of reproductive autonomy. A cell that permanently differentiates into a macroscopic organ, and in so doing makes itself sterile makes a greater short-term fitness sacrifice than a cell that undergoes reversible differentiation within a multicellular structure, but retains the potential ability to independently reproduce if that structure breaks down. This idea may be relevant to why there

were multicellular algae and fungi before the Neoproterozoic, but no multicellular (ancestral) animals. Cells of higher plants and other photoautotrophs undergo extensive differentiation, but retain a far greater degree of developmental plasticity than do the differentiated cells of animals, in which potency becomes far more restricted (e.g. Weigel & Jurgens, 2002). One cannot extrapolate too precisely from the development of those multicellular eukaryotes present today to that of their relatives from 500 Ma. But we suggest that it is useful to view the individual cell as incurring a greater short-term fitness cost in macroscopic heterotrophic form than in photoautotrophic form, because the former involves a greater restriction on the number of possible future developmental fates the cell can adopt. I.e. animals have been more successful in mediating the levels of selection conflicts inherent in multicellular form than other multicellular eukaryotes, but this has been achieved by making terminal cellular differentiation (and hence cellular altruism) more intrinsic to development. Consequently, macroscopic heterotrophic Edicara (and the Metazoa that some such taxa probably gave rise to) may have actually required the unique evolutionary conditions of strong kin selection and low advantage to cheating, provided by snowball Earth, in order to gain tangible fitness values.

If the evolutionary effects that we postulate here are reasonable, snowball Earth may have temporarily increased the degree of altruism that was adaptive and promoted an increasing degree of terminal differentiation. This paved the way for larger, more elaborate form and irreversibly redirected eukaryotic evolution.

ACKNOWLEDGEMENTS

We are grateful to Ilan Eshel for providing some of his previous work linking founder effects and altruism. We are very grateful to Nick Butterfield and two other (anonymous) referees for useful comments. We thank Liz Farmer for pointing out literature. TML's work is supported by a Philip Leverhulme prize. RAB thanks the school of Environmental Sciences at UEA for funding, and TML and Andy Watson for supervision.

REFERENCES

- Axelrod R, Hamilton WD (1981) The evolution of co-operation. *Science* **211**, 1390–1396.
- Baker ME (2006) The genetic response to Snowball Earth: role of HSP90 in the Cambrian explosion. *Geobiology* **4**, 11–14.
- Baum SK, Crowley TJ (2001) GCM response to late Precambrian ice-covered continents. *Geophysical Research Letters* **28**, 583–586.
- Baum SK, Crowley TJ (2003) The snow/ice instability as a mechanism for rapid climate change: A Neoproterozoic Snowball Earth model example. *Geophysical Research Letters* **30**, GL017333.
- Boyle RA, Lenton TM (2006) Fluctuation in the physical environment as a mechanism for reinforcing evolutionary transitions. *Journal of Theoretical Biology* **242**, 832–843.
- Brocks JJ, Logan GA, Buick R, Summons RE (1999) Archean molecular fossils and the early rise of eukaryotes. *Science* **285**, 1033–1036.
- Brown RH, Richardson M, Boutler D, Ramshaw JA, Jefferies RPS (1972) The amino acid sequence of cytochrome c from *Helix aspera* (garden snail). *Biochemical Journal* **128**, 971–974.
- Butterfield NJ (2000) *Bangiomorpha pubescens* n. gen., n. sp.: implications for the evolution of sex, multicellularity, and the Mesoproterozoic/Neoproterozoic radiation of eukaryotes. *Paleobiology* **26** (3), 386–404.
- Butterfield NJ (2004) A Vaucherian alga from the middle Neoproterozoic of Spitsbergen: implications for the evolution of Proterozoic eukaryotes and the Cambrian explosion. *Paleobiology* **30**, 231–252.
- Butterfield NJ (2005a) Probable proterozoic fungi. *Paleobiology* **31** (1), 165–182.
- Butterfield NJ (2005b) Reconstructing a complex early Neoproterozoic eukaryote, Wynniatt formation, Arctic Canada. *Lethaia* **38**, 155–169.
- Butterfield NJ (2007) Macroevolution and macroecology through deep time. *Paleontology* **50** (1), 41–55.
- Canfield DE, Poulton SW, Narbonne GM (2007) Late Neoproterozoic deep-ocean oxygenation and the rise of animal life. *Science* **315**, 92–95.
- Cohen D, Eshel I (1976) On the founder effect and the evolution of altruistic traits. *Theoretical Population Biology* **10** (3), 276–302.
- Corsetti FA, Olcott AN, Bakermans C (2006) The biotic response to Snowball Earth. *Paleogeography, Paleoclimatology & Paleoceanography* **232**, 114–130.
- Day T (2003) Virulence evolution and the timing of disease life history events. *Trends in Ecology & Evolution* **18** (3), 113–118.
- Deming JW, Huston AL (2000) An oceanographic perspective on microbial life at low temperatures with implications for polar ecology, biotechnology and astrobiology. *Journey to Diverse Microbial Worlds: Adaptation to Exotic Environments*. Kluwer, Netherlands and/or Norwell MA, USA, pp. 149–160.
- Eshel I (1972) On the neighbour effect and the evolution of altruistic traits. *Theoretical population biology* **3** (3), 258–276.
- Eshel I (1977) On the founder effect and the evolution of altruistic traits: an ecogenetical approach. *Theoretical Population Biology* **11** (3), 410–424.
- Evans DAD *et al.* (1997) Low latitude glaciation in the Paleoproterozoic era. *Nature* **386**, 262–266.
- Evans DAD (2000) Stratigraphic, geochronological, and paleomagnetic constraints on the Neoproterozoic climatic paradox. *American Journal of Science* **300**, 347–433.
- Evans DAD (2003) True polar wander and supercontinents. *Tectonophysics* **362**, 303–320.
- Evans DAD (2005) Proterozoic low orbital obliquity and axial-dipolar geomagnetic field from evaporite paleolatitudes. *Nature* **444**, 51–55.
- Fanning CM, Link PK (2004) U-Pb SHRIMP age of Neoproterozoic (Sturtian) glaciogenic Pocatello formation, southeastern Idaho. *Geology* **10**, 881–884.
- Feng DF (1997) Determining divergence times with a protein clock: update and re-evaluation. *Proceedings of the National Academy of Sciences of USA* **94**, 13028–13033.
- Fix AG (1985) Evolution of altruism in kin-structured and random subdivided populations. *Evolution* **39**, 928–939.
- Frank SA (1998) *Foundations of Social Evolution*. Princeton University Press, Princeton, New Jersey.
- Grey K, Walter MR, Calver CR (2003) Neoproterozoic biotic diversification: Snowball Earth or aftermath of the Acraman impact? *Geology* **31**, 459–462.
- Griffin AS, West SA (2002) Kin selection: fact and fiction. *Trends in Ecology & Evolution* **17**, 15–21.
- Grotzinger JP, Watters WA, Knoll AH (2000) Calcareous metazoans in thrombolithic bioherms of the terminal Proterozoic Nama group, Namibia. *Paleobiology* **26**, 334–359.

- Hamilton WD (1964) The genetical evolution of social behaviour, II. *Journal of Theoretical Biology* **7**, 17–52.
- Hamilton WD (1972) Altruism and related phenomena, mainly in social insects. *Annual Review of Ecology and Systematics* **3**, 193–232.
- Harland WB, Rudwick MJS (1964) The infra-Cambrian ice age. *Scientific American* **211**, 28–36.
- Hawes I, Howard-Williams C, Pridmore RD (1993) Environmental control of microbial biomass of the McMurdo Ice shelf, Antarctica. *Archiv fur Hydrobiologie* **127**, 27–287.
- Heckman DS, Geiser DM, Eidell BR, Stauffer RL, Kardos NL, Hedges SB (2001) Molecular evidence for the early colonisation of land by fungi and plants. *Science* **293**, 1129–1133.
- Hedges SB, Chen H, Kumar S, Wang DYC, Thompson AS, Watanabe H (2001) A genomic timescale for the origin of eukaryotes. *BMC Evolutionary Biology* **1**, 4.
- Hedges SB (2004) Molecular clocks and a biological trigger for Snowball Earth and the Cambrian explosion. *Telling the Evolutionary Time: Molecular Clocks and the Fossil Record*. CRC Press, Boca Raton, Florida, pp. 27–40.
- Hoffman PF, Schrag DP (2002) The Snowball Earth hypothesis: testing the limits of global change. *Terra Nova* **14**, 129–155.
- Hoffman PF, Kauffman AJ, Halverson GP, Schragg DP (1998) A Neoproterozoic Snowball Earth. *Science* **281**, 1342–1346.
- Hofmann HJ (1990) Ediacaran remains from intertillite beds in northwestern Canada. *Geology* **18**, 1199–1202.
- Huntley JW, Xiao S, Kowalewski M (2006) 1.3 Billion years of acritarch history: An empirical morphospace approach. *Precambrian Research* **144**, 52–68.
- Hurtgen MT (2002) The sulphur isotopic composition of Neoproterozoic seawater sulphate: implications for a snowball Earth? *Earth and Planetary Science Letters* **203**, 413–429.
- Hurtgen MT (2005) Neoproterozoic sulfur isotopes, the evolution of microbial sulphur species, and the burial efficiency of sulphide as sedimentary pyrite. *Geology* **33** (1), 41–44.
- Hyde WT, Crowley TJ, Baum SK, Peltier WR (2000) Neoproterozoic ‘snowball Earth’ simulations with a coupled climate/ice-sheet model. *Nature* **405**, 425–429.
- Javaux E, Knoll AH, Walter M (2003) Recognizing and interpreting the fossils of early eukaryotes. *Origins of Life and Evolution of the Biosphere* **33**, 75–94.
- Kennedy MJ, Christie-Blick N, Prave AR (2001) Carbon isotopic composition of Neoproterozoic glacial carbonates as a test of paleoceanographic models for snowball Earth phenomena. *Geology* **29**, 1135–1138.
- Kennedy MJ (1996) Stratigraphy, sedimentology and isotope geochemistry of Australian Neoproterozoic postglacial cap dolostones: deglaciation $\delta^{13}\text{C}$ excursions, and carbonate precipitation. *Journal of Sedimentary Research* **66**, 1050–1064.
- Kent DV, Smethurst MA (1998) A Shallow bias of paleomagnetic inclinations in the Paleozoic and Precambrian. *Earth Planet Science Letters* **160**, 391–402.
- Kerzberg M, Wolpert L (1998) The origin of Metazoa and the egg: a role for cell death. *Journal of Theoretical Biology* **193**, 535–537.
- Kirschvink JL (1992) Late proterozoic low-latitude global glaciation: the Snowball Earth. In *The Proterozoic Biosphere: A Multidisciplinary Study* (eds Schopf JW, Klein C). Cambridge University Press, Cambridge, UK, pp. 51–52.
- Knoll et al. (2006) Eukaryotic organisms in Proterozoic oceans. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences* **361**, 1023–1038.
- Knoll AH (1994) Proterozoic and Early Cambrian protists: Evidence for accelerating evolutionary tempo. *Proceedings of the National Academy of Sciences of the USA* **91**, 6743–6750.
- Kreft J (2004) Biofilms promote altruism. *Microbiology* **150**, 2751–2760.
- Lehmann L, Keller L (2006) The evolution of co-operation and altruism – a general framework and a classification of models. *Journal of Evolutionary Biology* **19**, 1365–1376.
- Lenton TM, Watson AJ (2004) Biotic enhancement of weathering, atmospheric oxygen and carbon dioxide in the Neoproterozoic. *Geophysics Research Letters* **31**, 1–5.
- Li CW et al., (1998) Precambrian Sponges with cellular structures. *Science* **279**, 879–883.
- Maynard-Smith JM, Szathmary E (1994) *The Major Transitions in Evolution*. Oxford University Press, Oxford, UK, pp. 40–58, 99–121.
- Meert JG, Torsvik TH (2004) Paleomagnetic constraints on Neoproterozoic ‘Snowball Earth’ continental reconstructions. The extreme Proterozoic: geology, geochemistry and climate. *Geophysical Monograph Series* **146**, 5–11.
- Michod R (1997) *Darwinian Dynamics: Evolutionary Transitions in Fitness and Individuality*. Princeton University Press, Princeton, New Jersey.
- Misi A, Veizer J (1998) Neoproterozoic carbonate sequences of the Una group, Irecé Basin, Brazil; chemostratigraphy, age and correlations. *Precambrian Research* **89**, 87–100.
- Narbonne GM (2005) The ediacara biota: neoproterozoic origin of animals and their ecosystems. *Annual Reviews of Earth and Planetary Sciences* **421–442**.
- Narbonne GM, Gehling JG (2003) Life after snowball: the oldest complex Ediacaran fossils. *Geology* **31**, 27–30.
- Nisbet EG, Fowler CMR (1999) Archaean metabolic evolution of microbial mats. *Proceedings of the Royal Society of London. Series B, Biological Sciences* **266**, 2375–2382.
- Nowak MA, May RM (1992) Evolutionary games and spatial chaos. *Nature* **359**, 826–829.
- Nowak MA (2006) Five rules for the evolution of co-operation. *Science* **314**, 1560–1563.
- Page RDM, Holmes EC (1996) *Molecular Evolution: A Phylogenetic Approach*. Blackwell Science, Oxford, UK.
- Peltier WR, Tarasov L, Vettoretti G (2004) Climate dynamics in deep time: modeling the snowball bifurcation and assessing the plausibility of its occurrence. The extreme Proterozoic: geology, geochemistry and climate. *Geophysical Monograph Series* **146**, 107–124.
- Peterson, Butterfield (2005) Origin of the Eumetazoa: Testing predictions from molecular clocks against the eukaryotic fossil record. *Proceedings of the National Academy of Sciences of the USA* **102** (27), 9547–9552.
- Peterson KJ, McPeck MA, Evans DAD (2005) Tempo and mode of early animal evolution: inferences from rocks, Hox, and molecular clocks. *Paleobiology* **31** (2), 36–55.
- Pfeiffer T, Bonhoeffer S (2003) An evolutionary scenario for the transition to undifferentiated multicellularity. *Proceedings of the National Academy of Sciences of the USA* **100** (3), 1095–1098.
- Pierrehumbert RT (2005) Climate dynamics of a hard Snowball Earth. *Journal of Geophysics Research* **110**, D01111.
- Pollard DP, Kasting JF (2005) Snowball earth: a thin-ice solution with flowing sea glaciers. *Journal of Geophysics Research* **110**, C07010.
- Rainey PB, Rainey K (2003) Evolution of co-operation and conflict in experimental bacterial populations. *Nature* **425**, 72–74.
- Rainey P, Travisano M (1998) Adaptive radiation in a heterogenous environment. *Nature* **394**, 68–72.
- Rice AHN, Hofmann CC (2000) Evidence for a glacial origin of Neoproterozoic III striations at Oaibaccannjar’ga, Finnmark, northern Norway. *Geological Magazine* **137**, 355–366.

- Rothman DH, Hayes JM, Summons RE (2003) Dynamics of the Neoproterozoic carbon cycle. *Proceedings of the National Academy of Sciences of the USA* **100**, 8124–8129.
- Rothschild LJ, Mancinelli RL (2001) Life in extreme environments. *Nature* **409**, 1092–1101.
- Sober E, Sloan-Wilson DS (1998) *Unto Others: The Evolution and Psychology of Unselfish Behaviour*. Harvard University Press, Cambridge, Massachusetts.
- Thomas DN, Dieckmann DS (2002) Antarctic sea ice – a habitat for extremophiles. *Science* **295**, 641–644.
- Traulsen A, Nowak MA (2006) Evolution of co-operation by multi-level selection. *Proceedings of the National Academy of Sciences of the USA* **103**, 10952–10955.
- Trivers R (1971) The evolution of reciprocal altruism. *Quarterly Review of Biology* **46**, 35–57.
- Van Baalen M, Rand DA (1998) The unit of selection in viscous populations and the evolution of altruism. *Journal of Theoretical Biology* **193**, 631–648, 191–192.
- Vincent WF (1988) *Microbial Ecosystems of Antarctica*. Cambridge University Press.
- Vincent WF, Mueller D, Van P (2004) Glacial periods on early Earth and implications for the evolution of life. *Origins: Genesis, Evolution and Diversity of Life* (ed. Joseph Seckbach). Kluwer, Dordrecht, pp. 481–501.
- Wang DY, Kumar S, Hedges SB (1999) Divergence time estimates for early history of animal phyla and the origin of plants, animals and fungi. *Philosophical Transactions of the Royal Society of London B. Series B, Biological Sciences* **266**, 163–171.
- Weigel D, Jurgens G (2002) Stem cells that make stems. *Nature* **415**, 751–754.
- West SA, Griffin AS, Gardener A, Diggle SP (2006) Social evolutionary theory for micro-organisms. *Nature Review of Microbiology* **4**, 597–607.
- Wilkins AS (2002) *The Evolution of Developmental Pathways*. Sinauer, Sunderland, Massachusetts, pp. 467–501.
- Wolpert L, Szathmary E (2002) Evolution and the egg. *Nature* **420**, 745.
- Wolpert L (1994) The evolutionary origin of development: cycle; patterning, privilege and continuity. *Development Supplement* **79–84**.
- Wright S (1945) Tempo and mode in Evolution—a critical review. *Ecology* **26**, 415–419.
- Xiao S (2004a) Neoproterozoic glaciations and the fossil record. The Extreme Proterozoic: Geology, Geochemistry and Climate. *Geophysical Monograph Series* **146**, 199–214.
- Xiao S (2004b) New multicellular algal fossils and acritarchs in Doushantuo Chert Nodules. *Journal of Paleontology* **78** (2), 393–401.
- Xue Y, Tang TF, Yue CL (1992) Discovery of the oldest skeletal forms from the Upper Sinian Doushantuo formation in Weng'an, Guizhou, and its significance. *Acta Paleontologica Sinica* **31**, 530–539.