BSE AND SCRAPIE RESEARCH

FROM: BSE Programme Management Group
7 February 1994

TO: Mr R Bradley

cc: Mr T E D Eddy
    Mr K C Taylor
    Dr J A Morris

1. Your minute of 1 February requested a consideration by the BSE Programme Management Group (PMG) of four possible areas/objectives for action as a result of your discussions with the CVO on your Concept Note. The BSE PMG has now met to discuss specifically the policy concern that the BSE agent could have become endemic in the British sheep population, this has been one of the subjects considered at recent meetings for future research to fulfil potential policy needs in order to formulate our research strategy. The following summarises the unanimous view of the PMG for the initial response requested.

2. It was agreed that there was evidence of scrapie in sheep as a result of food borne exposure. This is provided by the statistically significant increase in the incidence of sheep scrapie from 1985, as determined from analyses of the submissions made to VI Centres, and from individual case and flock incident studies. As the working hypothesis is that there has been recycling of infected cattle tissues which has augmented the epidemic in cattle the continued infection of sheep with the BSE agent, via the food borne source, cannot be excluded. There is therefore also a possibility that the BSE agent may have become endemic in the sheep population, but it is impossible to design any short-term research programme to elucidate this.

3. Taking the four possible areas and objectives for consideration in turn:

   3.1 "To determine if natural infection of BSE can be distinguished clinically or promptly by pathological methods post mortem"

   Considering pathological methods initially, in practical terms this requires a study of the brain changes, particularly the distribution and severity of vacuolar changes (the lesion profile) in clinical cases. This poses the following difficulties:
3.1.1 The lesion profile depends principally on strain of agent and genotype of the sheep and it is now well established that these are essentially constants for BSE in cattle and variables for natural scrapie in sheep. Previous work (Fraser 1976, Barlow unpublished, Wood et al unpublished) has indicated the difficulties of creating the necessary baseline to cope with these and possible additional (e.g. route of infection) interacting variables that affect the lesion profile. A population study of lesion profiles in sheep therefore presents considerable difficulties in both the number of sheep brains requiring examination and the detail of the profile analysis required to show small differences (see Wells et al 1992, Wells et al in press).

3.1.2 At present, the identification of a "BSE-profile" in sheep rests on the profiles obtained at first experimental passage of bovine BSE in sheep, that is across a species barrier. Should it prove possible to establish a distinctive lesion profile in this small sample there are no grounds for assuming that such a profile in sheep would be the same on passage in sheep (infection without a species barrier). A cattle-to-sheep barrier must be inferred otherwise there would be no point in executing any of the proposed studies. A second oral passage of BSE in sheep would be required to test the constancy of a "BSE-profile" in sheep which would not necessarily reflect the profile of naturally transmitted BSE in sheep because of variables due to possible non-oral routes of infection.

3.1.3 The profile approach can only identify the major strain(s) associated with the development of clinical disease. Food borne risks to other species depend on the amount and strains of infection in various tissues used for food. Lesion profiles cannot identify mixed infections (and clearly do not even identify infection at all if this is confined to the LRS).

Notwithstanding such difficulties there is merit in conducting a strictly limited exploratory assessment of existing material since a sufficiently characteristic profile in sheep (especially if it could be based exclusively on a medulla-obex examination) provides information which would be of value in further studies to diagnose the occurrence of clinical BSE in sheep. The feasibility of identifying a BSE profile in sheep could be explored in the short term from:

- study of the lesion profile in experimental BSE in sheep and goats (this is already in hand)
review of Mr J Wood's analysis of Pathology Dept, CVL sheep and goat scrapie pathology and possible re-examination of the material (initial access to Mr Wood's unpublished review is sought)

3.1.4 The potential for clinical distinction of scrapie and BSE in sheep is, given the variability and multiplicity of the clinical signs, less sensitive than the lesion profile approach and there are no data on which to base a preliminary assessment.

3.2 "To determine whether or not BSE agent exists in the national sheep flock (and, if so, later to determine at what incidence)"

Of the four objectives this is the only one which is fulfillable by a direct approach which seeks to identify strains of agent in circumstances where endemic infection with the BSE agent, if it occurs in sheep, would already be present. Sampling problems can be managed more economically because strain typing could be carried out with pools of tissue from a sufficiently large sample of animals.

It also provides the most sensitive approach, and reduces the risk of false negative results. More specifically, it is capable of identifying the BSE agent:-

(a) in sheep infected by natural routes and doses
(b) in both LRS and CNS tissues
(c) in mixed infections
(d) even when present as a quantitatively minor component

As a result, this objective was considered to be the most important identified in relation to the policy concern. A ROAME proposal is therefore being prepared and is outlined below.

3.3 "To determine the pathogenesis of experimental BSE in sheep following oral challenge with BSE brain, the tissue distribution of the agent during the incubation period and the temporal progression of infectivity titres in infected tissues"

This has the fundamental problem that it is an indirect approach. It does not accommodate natural levels of exposure and non-oral routes of infection and involves the experimental exposure of BSE to sheep. A more direct approach would be necessary to expose sheep experimentally to brains from BSE affected cattle and conduct the
ultimate pathogenesis study using passaged material from these sheep, to inoculate uninfected sheep. This does not, however, overcome the underlying problem that a negative or positive result with respect to detectable LRS infectivity, in sheep in this second passage study, would not be sufficient to discount natural transmission of the BSE agent in sheep, because natural transmission does not depend exclusively on LRS infectivity and because of the insensitivity of bio-assays across the species barrier (i.e. in mice). The absence of an invariable, 100 per cent, detection rate of the scrapie agent in placenta of affected ewes is clearly relevant in the consideration of this approach.

3.4 "To determine whether or not maternal transmission occurs in experimental BSE in sheep following oral challenge, and if so with what incidence"

This approach has a number of weaknesses which cannot be removed by modification of the initial idea. Primarily, it is scientifically invalid because of the unnatural dose, source and routes of infection as past studies of the contagious spread of scrapie revealed the extent to which experimental infection of sheep with scrapie does not reproduce all of the important features of the natural disease. It is also impossible to design a study which could simulate natural conditions as husbandry practices cannot be dismissed as important factors in the natural transmission of scrapie. The approach is also based on the premise that maternal transmission provides the main means by which scrapie is maintained in the sheep population; whereas there is evidence that horizontal transmission is at least of equal importance and the precise means by which scrapie is transmitted between sheep remains an unknown. If such an approach was pursued it would create uncertainties in the interpretation of a positive result, and a negative result would be inadequate, on its own, as evidence against the natural transmission of the BSE agent in the sheep population.

4. In the third paragraph of your minute you asked for existing information on the possible exposure of sheep to the BSE agent via feed and any evidence that maternal transmission does or does not occur. On the first point there is no evidence, other than that summarised above, and on the second aspect maternal transmission is certainly likely to occur, but is unlikely to be the means of transmission to maintain scrapie in the sheep population.

5. Finally, we are producing a ROAME proposal to investigate the policy concern that the BSE agent may now be endemic in the British sheep population. This has been designed to have added value in that it provides data which will be invaluable to other aspects of BSE R&D that have policy
implications. At the risk of over-simplification, this can be summarised as involving the following activities:

- the collection of a representative sample of brains from scrapie-affected sheep, using criteria to exclude cases which could have been infected from a primary food borne source, which is likely to be a mix of scrapie and BSE.

- the initial genotyping of all pathologically confirmed cases, using a case definition based on the results of histological and EM examinations to determine the occurrence of polymorphisms at codons 136 and 171.

- the oral exposure of one group of cattle to a pool of brains from sheep of one of these genotypes and another to the alternative genotype.

- strain typing of these two homogenates in mice

- strain typing, in mice, of clinical cases occurring as a result of this exposure.
There are three possible outcomes to this study:

<table>
<thead>
<tr>
<th>OUTCOME</th>
<th>Transmission to cattle</th>
<th>Strain typing of scrapie in mice</th>
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</thead>
<tbody>
<tr>
<td>One</td>
<td>Positive</td>
<td>No BSE agent</td>
</tr>
<tr>
<td>Two</td>
<td>Positive</td>
<td>BSE agent</td>
</tr>
<tr>
<td>Three</td>
<td>Negative</td>
<td>No BSE agent</td>
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The second of the above outcomes would provide evidence that the BSE agent has become endemic in the British sheep population. Outcomes One and Three would be evidence against this phenomenon.

REFERENCES


J W WILESMITH
Chairman BSE PMG