Response to the UKAS Report on a case of atypical scrapie in VLA project SE1931

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April 2007
Introduction

The VLA welcomes receipt of the final report of the UKAS audit conducted in the week commencing 27th November 2006. The thrust of the report concurs with the VLA’s own investigations, in that the case of atypical scrapie reported (G320) was indeed a genuine and correct diagnosis. Furthermore, we also conclude that it is was not possible at the time of the report to determine the origin of infection. Subsequent investigations and testing at the VLA have been unable to resolve this issue.

While we acknowledge references to lapses of biosecurity, none of which can be clearly identified as responsible for the introduction of scrapie, we believe that with the exception of the few points raised below, these issues are most appropriately discussed in the context of the scientific discussion about the case and factors identified.

We are however concerned about some misinterpretations in the report. As the report will be in the public domain we have commented below on the most important issues. Most importantly, recognising that the challenge of interpreting the facts in a scientific context will fall to SEAC and others, we refrain from beginning that debate in this response.

Specific comments

Executive summary
Paragraph 3 – Examination of culled sheep.
In the executive summary, but not elsewhere, the report states that a decision was taken in February 2006 not to examine the brains of culled sheep of resistant genotype, making it difficult to determine the extent of atypical scrapie in the flock. The auditors were reporting a note of a meeting which was not subsequently actioned as worded, and that their consequential comment about the loss of opportunity to screen for atypical scrapie is invalid. We acknowledge the fact that not all culled sheep were screened, but the exclusion generally related to animals culled at a young age. Their examination for the presence of scrapie was judged, on the basis of scientific knowledge at the time, to be unlikely to have identified any case of scrapie, whether atypical or classical, especially as it would be in a “low risk” context (low exposure rates) where long incubation periods might be expected. This seriously limited the power of any post-mortem screening of young culled animals.

Furthermore, the vast majority of animals born in the flock were distributed to TSE related studies at the VLA and elsewhere, where exposure to scrapie or BSE was anticipated, further compromising the interpretation of post-mortem results. Nevertheless, we acknowledge, with the benefit of hindsight, that a policy that required the examination of the brains of surrogate dams received in lamb by other institutes (ie. mature sheep not exposed at the receiving institute) would have been informative in the context of our retrospective investigations.
A total of 711 animals, of all ages and genotype, were culled from the breeding flock, out of a total of 4868 sheep that had left the flock at the time of investigation. Of the 711, a total of 447 were not examined at all for the presence of scrapie. Only 13 were over 12 months of age at the time of culling, and most of the remainder would have been under 6 months old. The flock breeding plan is managed in order to meet demands for lambs of particular genotypes in the following lambing season. It is inevitable that there will be some that will be surplus to demand because their genotypes do not match current needs. If not required for later breeding, it is therefore costly to retain such animals indefinitely, thus increasing pressure to reduce numbers sooner rather than later. Accommodation at the site is a limiting factor, so the flock size must be carefully managed to ensure that there is no overcrowding with consequential health and welfare problems.

Of the total culled and untested, only 46 coded for arginine at codon 171 (heterozygote or homozygote), and of these, only seven were over 12 months of age. Only one was aged four years. Four of the seven were homozygotes, the genotype that would have been classified as scrapie-resistant until atypical scrapie was first detected through active surveillance in the UK national flock in 2002. We therefore continue to feel that monitoring of young animals for the presence of scrapie would be disproportionate and that targeting of older animals continues to be justified.

Paragraph 6 - The report concludes that with J19, cross-contamination of inoculum cannot be ruled out. The VLA interpretation of data relating to J19 is that they remain equivocal. While at the time of the investigation the absence of corroborating positive immunochemical results, and apparent negative status of parallel bioassays in France and Germany, did suggest quite strongly that a cross-contamination incident at the time of inoculation might have been the logical interpretation, doubts remain. Further analysis of ongoing assays, at the VLA and in France and Germany, are however consistent with an interpretation that J19 may have been infected, at low titre, and that the detection of infectivity in the VLA assay in tg338 mice could reflect the greater sensitivity of inoculation by combined i.c.+i.p. routes (total volume of inoculum 120µl) as opposed to the i.c. route (total volume 20µl) used by our collaborators. VLA also uses larger group sizes for each assay. We stress that the interpretation of incomplete assays is potentially misleading. Nevertheless, in comparing parallel assays of atypical scrapie in tg338 at the VLA and in France, incubation periods are consistently, and significantly, shorter at the VLA. These results relate to high titre inocula, and therefore the difference may be more critical at low titre. This dilemma may be resolved shortly as we believe that most of the TgshpXI mice challenged with J19 inoculum in Germany are ready for examination. If not resolved, it is possible that bioassay of further aliquots of the J19 inoculum may assist our interpretation.

Paragraph 7 - The report recommends consideration of future waste management plans in view of the fact that the flock is presumed infected and waste is spread on an arable commercial farm. We are not aware that such spreading is precluded where scrapie is naturally acquired in commercial
flocks, and consideration of the risk should not attribute a disproportionate risk to this flock which is derived from scrapie free sources, and maintained as closed flock with significantly greater monitoring than any commercial flock maintained in the UK.

Observations and Assessment Findings
Page 5, para 5 – Sheep G320 is stated to “code for phenylalanine at amino acid position 141, which is characteristic of atypical scrapie.” The coding of the PrP gene is a characteristic that applies to the host, in this case a sheep, and is not a phenomenon that is attributable to the disease – in this case atypical scrapie. In reality, the AFRQ PrP gene (the AFRQ allele) is found in approximately 20% of sheep identified as having atypical scrapie in Great Britain (Saunders et al, 2006). The AFRQ allele is certainly considered as being a significant risk factor for atypical scrapie in comparison to classical scrapie, where it is found in very few cases. The frequency for the AFRQ allele in the healthy sheep population has been estimated as around 8% of all animals.

Site suitability – para 1&2. These paragraphs are slightly inconsistent, in that the first paragraph suggests that there is “no known history of livestock since around 1960”, which implies that there may have been livestock kept of which we are unaware. Meanwhile paragraph two states – “confirming that no livestock has been maintained on the fields from that date (ie. 1960).” This last statement is correct – as ADAS has detailed farm management records to verify land use since they took over ownership. We therefore presume that the conflict arises due to grammatical error in para 1, and that no uncertainty remains about the history of the farm with respect to grazing by livestock.

At the time the sheep unit was constructed in 1997, ADAS could prove that the land had been free of livestock and animal manures for at least 34 years ie. since 1963 when ADAS took over management of the farm.

Site suitability – para 5. At the time of the site visit the auditors were not able to access details of the sewage sludge trial as they were stored on older files. Plot details were however forwarded later, along with clarification of procedures adopted. While it is true that the VLA staff member interviewed did not recall being aware of the sewage trial at the time the sheep project was established, subsequent record searches have confirmed that both the VLA and MAFF were aware of the trial, and it was taken into account at the time that the suitability of the site was being considered.

The trial took place between 1994 and 1997. The location of the sites of application, the methods of application, drainage arrangements and the relative heights of land in the sheep unit, trial plots and intervening road make run-off in the direction of the sheep unit a physical impossibility. Furthermore, in light of the culture of detailed record keeping at this site, this statement is reinforced by the fact that there is no record of flooding either.

Disease security – visitors – paragraph 1 – while it is agreed that site instructions did not require declaration of freedom from contact with TSEs *per se*, perhaps through the handling of infected tissues, in reality this has no impact on risk given that no such individuals have visited, or been allowed to visit, the site and contact the sheep. The VLA project leader, based at Weybridge, is the only individual with perceived exposure to infected animals, but has no access to other materials. He is subject to the same rules regarding exposure to ruminants. This is therefore a perceived weakness that has not represented a real risk of exposure.

Disease security – visitors – paragraph 3. Reference to the visit by shearers in May 2001 implies that the absence of a countersignature by ADAS staff indicates a real risk to the flock. The use of self-declaration forms on entry is however accompanied by supervision of entry by a member of ADAS staff, through the only point of entry. This routinely involves questioning regarding recent contact with livestock, and entry would not be gained solely based on the self-declaration. It is accepted that countersigning by local staff would confirm their involvement in approving entry.

Biosecurity lapses – have been mentioned at various places, although in each case real risk of exposure has not been identified. These issues are best addressed in the context of the scientific debate about the incident. It is inevitable that such perceived lapses must take into account factors outwith the farm, that the audit could not take into account, before there can be any consideration of whether or not biosecurity was really compromised. It is also important to stress that the flock was never managed in such a way as to exclude exposure to pathogens in general. Given our understanding of the science of prion diseases at the time of establishment and subsequently, measures were intended to exclude scrapie and similar diseases.

Appendix IV – report by the veterinary consultant, B Hosie

Again the key issues relating to the appendix, and implications, if any, to the introduction of atypical scrape, are best addressed in the context of the scientific debate. The statements are largely factual and correct, but interpretation is not possible without further discussion.

Background

One sentence in this paragraph states – “Endoparasites, ectoparasites and hay mites have all been proposed as reservoirs of infection” – appears to be a definitive statement. It is correct in that many scientists have hypothesised that each has a role in the epidemiology of scrapie. Definitive proof remains elusive however, and the evidence put forward can be challenged on many grounds. It is however difficult to understand the relevance of the statement in terms of the evidence reviewed during the audit.

Routine security serology

This section deals with the use of serology to monitor the general health of the flock, and to screen for potential introduction of viral or bacterial diseases. The latter would be potential indicators of a breakdown in biosecurity, but this
would be dependent on the nature of the pathogen identified, and its likely mode of introduction. It was never intended as a means of detecting the introduction of scrapie per se, and is acknowledged in the report.

It is not possible to use serological testing for the introduction of TSEs. They produce no detectable change in blood constituents, or an immune response, at least by currently available validated technology. The sentence – “scrapie and other TSEs are highly infectious and are caused by agents that are exceptionally resistant to disinfection” – therefore seems out of context, and alarmist, and should be qualified by many other factors that determine whether or not exposure to TSEs results in infection. It is not an absolute, and the disease is not considered to be contagious in the sense that viral or bacterial diseases would be.