



SPONGIFORM ENCEPHALOPATHY ADVISORY COMMITTEE
Minutes of reserved business of the 81st meeting held on 25th February 2004

At

Dti Conference Centre
Victoria St
London

Members:	Professor P. Smith (Chairman)	
	Mr J. Bassett	
	Dr D. Brown	
	Mr C. Browne	
	Professor G. Bulfield	
	Professor R. Carrell	
	Dr J. Chambers	
	Professor N. Hooper	
	Mr P. Jinman	
	Dr C. Lasmezas	
	Professor I. McConnell	
	Dr J. Manson	
	Dr G. Medley	
	Dr P. Rudge	
Technical Advisors:	Dr P. Barrowman	(Defra)
	Dr S. Dixon	(FSA)
	Mr P. Soul	(Defra)
	Dr J. Stephenson	(DH)
	Dr D. Matthews	(VLA)
Assessors:	Dr M. Bailey	(Defra)
	Mr A. Harvey	(FSA)
	Dr R. Jecock	(DH)
SEAC Secretary:	Dr C. Boyle	
Secretariat:	Mr M. Pemberton	
	Dr B. Jeffery	
	Dr P. Keep	
	Ms T. Dale	
	Dr C. Ravirajan	

ITEM 1 - CHAIR'S INTRODUCTION

1. The Chair welcomed Dr Lorenz Amsler (Federal Office of Public Health, Division of Epidemiology and Infectious Diseases, Bern), Dr Markus Glatzel (Institute of Neuropathology and National Reference Centre for Prion Diseases, Zurich), Dr David Hilton (Department of Histopathology, Derriford Hospital, Plymouth), Dr. Peter Bennett (Department of Health) and Professor John Kearney (National Blood Service) who were in attendance to present agenda items.

ITEM 2 – APPROVAL OF DRAFT RESERVED BUSINESS MINUTES FROM 26TH NOVEMBER 2003 (SEAC 80)

2. The Chair informed members that as the morning session had finished late, there was limited time for the afternoon session. The Chair asked members if they would provide written comments on the draft minutes to Dr C Boyle by post and the changes would be approved by Chairman's action.

ITEM 3- CJD SURVEILLANCE IN SWITZERLAND

3. At the November 2003 meeting the committee requested an update on the epidemiology of sporadic CJD (sCJD) in Switzerland. At the invitation of the secretariat, Dr Lorenz Amsler from the Swiss Federal Office of Public Health (SFOPH) and Dr Markus Glatzel, from the Institute of Neuropathology and National Reference Center for Prion disease, Zurich presented this item.
4. Dr Amsler presented work by the SFOPH, which investigated the increased incidence of sCJD reported between 2001 and 2002. A yearly incidence of 2.6 deaths per million from sCJD were reported between 2001 and March 2002, compared to a lower rate in earlier years. In 2003, the incidence rate continued to be high and around 16 sCJD cases had been reported but more cases were expected because of the delay in the onset of symptoms and subsequent diagnosis of disease.
5. A study of the Swiss sCJD cases (n=48) between 2001-2003 examined three hypotheses for the increased incidence; (1) increased ascertainment, (2) iatrogenic transmission, or (3) zoonotic disease. The statistical power of the study was limited given the small numbers of sCJD cases. The 2001-2003 cohort of CJD patients exhibited similar characteristics of sporadic CJD (median age of 70 years, range 48 to 83) compared to cases reported between 1996-2000 (median age 67 years). There was no statistical significance in the gender difference between the 1996-2000 and 2001-2003 cases.

6. Members asked if there was an association between the incidence of sCJD and geographical location. Dr Amsler confirmed that the place of residence of each case had been investigated 10,15 and 30 years before death and no unusual distribution had been found. Members asked if the difference in age between the Swiss sCJD cases (70.1 years) reported between 2001-2003 compared to cases reported between 1996-2000 (67.3 years) was significant. Dr Amsler confirmed that the difference between the two groups was not statistically significant.
7. No association had been found between incidence of sCJD and lifetime occupation. All but one of the sCJD cases had undergone some form of surgery (n=47/48). The type of surgical or dental procedure, blood transfusion, hypertension treatment and overseas travel requiring vaccination and dietary history did not appear to be a risk factor for sCJD disease.
8. Investigation of the temporal pattern of sCJD showed that an increase in sCJD cases occurred in 2000, in the same year that testing for protein 14-3-3 was introduced. Dr Amsler suggested that ascertainment bias could explain the increase in the number of reported cases of sCJD. An increased awareness rather than a real increase in the number of cases could explain the rise in disease incidence. Further investigations are planned, including a case-control study to investigate iatrogenic transmission as a possible risk factor for sCJD.
9. The Chair commented that improved surveillance and good communication in a small country was one possible explanation for the observed increase in cases. Dr Amsler also commented that the majority of patients that died in hospital agreed to post mortem which meant many sCJD diagnoses being confirmed by neuropathology.
10. One member commented that this was similar to the situation in Austria where post mortem were mandatory under the Rokitansky legislation. However although the majority of sCJD cases in Switzerland, were confirmed at autopsy, diagnosis was made using clinical symptoms.
11. Dr Markus Glaztel presented a summary of the research conducted by the Swiss National Reference Centre for Prion diseases. Western blot analysis of the Swiss CJD cases did not show a glyco-type profile consistent with variant CJD. Of the 41 cases of sCJD reported between 2000-2003, 59% were methionine homozygotes (MMs), 19% were methionine/valine heterozygotes (MVs) and 22% valine homozygotes (VVs) at codon 129 of the prion gene. This compared to 42% MMs, 45% MVs and 13% VVs in the control group (n=74). Dr Glatzel reported an increase in the number of sCJDMV2 cases in the 2000-2003 cohort.
12. A 55 year old, female sCJD case had been identified with atypical disease, which was not classical vCJD. The patient had PrP^{Sc} type 1 glyco-type (as defined by the Gambetti classification) but had plaque lesions in the

cerebellum and throughout the whole brain. Further research was being conducted on this case.

13. Dr Glazel described published work¹ reporting that a sensitive western blot method had detected PrP^{Sc} in the spleen (n=10/28) and skeletal muscle (n=8/32) of sCJD cases. PrP^{Sc} accumulation was not muscle type specific and not associated with the observed increased incidence in sCJD between 2000 and 2003. Further work would involve the strain typing of sCJD case tissue compared to UK CJD, BSE and Swiss Scrapie and Chronic wasting disease samples.
14. Members commented that an increased incidence of CJD had been reported in countries following the introduction of protein 14-3-3 testing. This added to the evidence that ascertainment bias may explain the increased incidence. French investigators² have reported a 50% increase in the number of CJD cases between 1991 and 1997. Dr Amsler informed members that testing for protein 14-3-3 had been introduced in Switzerland, 18 months before the observed increase in incidence. The committee agreed that the one-year delay between patient referral and diagnosis that took place in that time was an argument against ascertainment bias.
15. Members asked if the sCJD cases reported between 2000-2003 in Switzerland, showed a bias towards males (n=31/48). This gender difference was not statistically significant.
16. Members agreed that ascertainment bias could be one of a combination of factors contributing to the increase in cases.
17. Members agreed that strain typing experiments were important. The committee agreed that lesion profiles provided a clear distinction between strains compared to biochemical analysis (i.e. western blots). Members noted that wild type mice were relatively resistant to sCJD and the use of transgenic models was more appropriate. In response to a question on the implications of UK CJD surveillance, Dr Ward commented that in the UK strain typing experiments were limited by the amount of tissue available due to the low post mortem examination rate.
18. The Chair suggested that a future update on the strain typing experiments of BSE cases would be useful background information for the committee.

ACTION: SECRETARIAT

19. The Chair thanked Dr Amlser and Dr Glatzel for their presentations.

¹ Glatzel M, Abela E, Maissen M, Aguzzi A. Extraneural pathologic prion protein in sporadic Creutzfeldt-Jakob disease. *N Engl J Med.* 2003 Nov 6;349(19):1812-20.

² : Brandel JP, Delasnerie-Laupretre N, Laplanche JL, Hauw JJ, Alperovitch A. Diagnosis of Creutzfeldt-Jakob disease: effect of clinical criteria on incidence estimates. *Neurology.* 2000 Mar 14;54(5):1095-9.

ITEM 4 - RETROSPECTIVE STUDIES OF THE PREVALENCE OF PrP^{Sc}

20. The Chair welcomed Dr David Hilton from the Department of Histopathology, Derriford Hospital. Dr Hilton is the lead scientist and pathologist for a DH-funded study and presented this item. The Chair welcomed Dr Azra Ghani who had done some modelling of the human epidemic using the results from Dr. Hilton's study.
21. Dr Hilton presented the pre-publication results from a two-centre in England and Scotland retrospective survey of human appendix and tonsil samples. The study was set up to determine the prevalence of detectable PrP^{Sc} in these tissues. The survey was unlinked and anonymised and had received ethical approval on this basis. A total of 16,703 samples were collected, (14,964 appendectomies and 1,739 tonsillectomies). In this sample approximately 60% were from 20-29 age group. 25% of samples (n=4029) were excluded from the final analysis because of inadequate amounts of lymphoid tissue (<5 secondary lymphoid follicles). Out of the remaining 12,674 samples three appendix samples tested positive for PrP^{Sc}. One immuno-reactive sample showed a staining pattern similar to that observed in vCJD cases with a coarse granularity in one out of six secondary lymphoid follicles. In two of the positive appendix samples, the immunostaining showed a fine granular pattern confined to the follicular dendritic cells which was not similar to that typically observed in vCJD cases. In all three samples there was no histopathological evidence of inflammation or neoplasia.
22. Dr Hilton suggested that possible explanations for the two atypical results included false positive immuno-staining, existence of a PrP^{Sc} carrier state or a phenotype with a longer incubation period than currently observed in vCJD cases.
23. Members asked if the positive samples had been genotyped for codon 129 of the PrP gene. Dr Hilton informed the committee that ethical approval for genotyping had not been obtained for this study. Also, repeat testing of the positive samples meant that a limited quantity of tissue was available, which prevented the possibility of genotyping in the future.
24. Professor Noel Gill (Health Protection Agency) provided the committee with details of a national tonsil archive which will collect tonsil samples prospectively. This will also be an unlinked and anonymised survey which will require further ethical approval for genetic analysis of samples. Professor Gill commented that the Swiss authorities had started a prospective linked study, which would allow a trace back to tissue donors should an effective treatment for CJD become available.
25. Members asked Dr Hilton if the PrP^{Sc} observed in the lymphoid follicles of the appendix tissue was co-localised with B- or T- lymphocytes or with

other phenotypic changes to the tissue architecture associated with inflammatory disease. Dr Hilton confirmed that inflammation was not present in areas of the tissue where PrP^{Sc} had been detected. Members commented that it was not known in what stage of the incubation period, if any, these samples represented. However previous researchers had detected PrP^{Sc} in the appendixes of vCJD patients, 8 months and 2 years before clinical onset of disease.

26. Dr Azra Ghani provided the committee with a revised statistical analysis of the estimated prevalence of vCJD based on the three positive samples (in 12,674 tested). The revised estimate of the prevalence was 237 infections per million population (95% CI 49-692 per million). Assuming this estimate relates to those aged 10-30 years (83% of the sample), then the best estimate is 3,808 individuals (95% CI 785-11,128) aged 10-30 incubating vCJD. Dr Ghani indicated that these predictions do not match the observed data of 18 deaths in 2003 and should be considered with the caveat that only a subgroup of the population had shown susceptibility to disease (methionine homozygotes).
27. The committee was informed about the proposal to analyse tonsil samples from the prospective study in batches of 5000 (until approximately 100,000 had been collected in total). The MRC prion unit had already collected 3000 tonsil samples. Members endorsed the decision to batch analyse the collected tonsil biopsies and recommended that ethical approval be obtained for the genotyping of positive samples. The Chair agreed to write to the Chief Medical Officer to emphasise the importance of speeding the conduct of further research using freshly collected tonsil specimens. This would increase the sample size on which prevalence estimates might be based. SEAC will be kept informed of progress with this work.
28. Dr Hilton informed the committee that the data was under review by a pathology journal and they hoped to publish by summer 2004. The Chair thanked Drs Hilton and Ghani for presenting to the committee.

ITEM 5 - DEPARTMENT OF HEALTH RESEARCH UPDATE

29. Dr John Stephenson provided committee members with an update on the Department of Health research portfolio.
30. Members noted that DH had contributed over £4.5 million on TSE research for 2002-03. Research includes the diagnosis of CJD, blood safety, laboratory detection of PrP^{Sc}, decontamination and also to inform policy needs on human TSEs in the areas related to preventing the spread of CJD, and treating patients and people at risk.
31. Some progress has been made on both detection and inactivation of prions on surgical instruments. Several technologies have been marketed, or are close to being marketed. A pre-clinical test for CJD has not yet been

developed, and several research programmes on potential CJD therapy are under way and a clinical trial protocol has been developed.

ITEM 6 – BONE RISK ASSESSMENT

32. The Chair welcomed Dr Peter Bennett from DH to present and invited Professor John Kearney from the National Blood Service (NBS) to join the discussion on this item.
33. In November 2003 SEAC considered a draft risk assessment, produced by the NBS and the Department of Health's Economics and Operational Research (EOR) division, to examine the risk of transmission of vCJD via implantation of bone. This draft assessment had been revised to take account of members' comments.
34. Members were asked to consider the revised calculations for the probability of infection and advise on the comparative risks of the different bone products with regard to vCJD transmission given the scientific uncertainties of the level of infectivity present and the effect of processing, including γ -irradiation of bone.
35. Dr Peter Bennett informed members that revision hip replacement surgery could be performed on otherwise healthy elderly patients whose life expectancy was long enough to allow the development of vCJD should there be transmission from an infected bone product.
36. Members were informed that bone is sourced from either single femoral heads (from living patients undergoing primary hip replacement) or from cadaveric donors. The assessment compared the risks associated with different bone products i.e. processed or unprocessed, pooled or unpooled, under different scenarios of infectivity. At their previous meeting in November, the committee agreed that as uncertainties around infectivity were unlikely to be resolved soon, they endorsed the need to consider as many scenarios as possible with respect to the risks from the different bone products.
37. In November 2003, the committee had commented that infectivity in bone could not be discounted. Dr Bennett informed the committee that if infectivity could reside in bone the advantage from blood and marrow removal could be substantially negated. Members noted that in unprocessed bone, vCJD infectivity could also be present in non-leucodepleted blood and marrow. In addition the fat content of bone and the lipophilicity of prion protein were also factors that could effect the conclusions made from the risk assessment
38. Members noted that processed femoral heads could either be unpooled, from a single cadaveric donor, or pooled from living donors. The NBS estimated that at least 99% of blood and marrow is removed by processing. Members were informed that femoral heads from single

donors can be used after washing and centrifuging, which is estimated to remove 98% of blood and marrow (Lomas et al, 2000³). The committee noted that γ -irradiation of the bone might potentially reduce infectivity in the order of one log of magnitude and the calculations in the paper had been revised accordingly to consider alternative assumptions.

39. The risk assessment showed that the risk of transmission of vCJD from bone is affected by pooling and is dependent on the assumed level of infectivity. Processing of bone reduces the amount of blood and marrow, therefore the risk of infection from that source would be lowered. However if living (rather than cadaveric) donors are used, processing requires the pooling of typically 17 donations. This is due to the relatively small mass of each individual femoral head. Pooling donations may constitute a greater risk because of the possibility of sourcing infectious material from any one of multiple donors. The EOR paper addressed the balance of risk between processing and pooling in different scenarios.

40. Dr Bennett described the possible scenarios involving the processing and pooling of bone.

- If infectious doses transmitted remain below 2 ID_{50S}, then pooling does not change the expected number of infections for a given donor prevalence. Pooling of donations would result in a greater number of recipients receiving a small dose. Under a linear dose-response relationship, these considerations would cancel out. If infectivity were confined to blood and marrow, the key consideration would be the effective removal of this by processing. In these scenarios, the risks associated with unprocessed femoral heads would be greater than for any of the other products.
- However in scenarios where doses exceed 2 ID_{50S}, the risk of infection is mainly determined by the number of donors in the pool. In high-infectivity scenarios, the disadvantages of pooling would outweigh the benefits of processing. This could occur if the mix of blood and marrow had high potential infectivity and/or if infectivity could reside in the bone itself.

41. Dr Bennett informed the committee that pooling of femoral heads was never advantageous, unless there was a threshold dose of infectivity. Batch dedication of bone donations from a single cadaver would decrease the risk from vCJD. Batch dedicated donations could be used instead of processed products containing a quantity of bone, equivalent to half a femoral head which could double the number of donors to each recipient.

42. Professor Kearney commented that the amount of bone in one unit sourced from a pool of 17 processed femoral heads was equivalent to one

³ Lomas R, Drummond O, Kearney JN. Processing of whole femoral head allografts: A method for improving clinical efficacy and safety. Cell and Tissue Banking 1:193-200,2000.

unprocessed femoral head. Therefore the risk from vCJD infection were lower using one femoral head from a cadaver.

43. Given the assumptions made in the assessment and uncertainties around infectivity in bone,

- processed femoral heads (even if pooled) were of lower risk than unprocessed, in scenarios with low infectivity in blood and marrow.
- However in high infectivity scenarios, and/or if bone itself was infective, unpooled femoral heads were of lower risk than pooled donations (even if unprocessed).

44. Members were informed that the NBS is a supplier of approximately 50% of the bone used in England and Wales, of which three quarters comes from femoral heads from living donors and one quarter from cadaveric donors. Professor Kearney informed the committee that femoral heads could not be morcellised and then washed due to the limitations of the apparatus used. Typically two or three femoral heads were individually washed before morcellisation, freeze-dried and irradiated. Eight times as much bone was collected obtained from a cadaveric donor compared to that taken during hip replacement from a living donor. Members asked if the long bones could be collected but Professor Kearney informed the committee that cancellous bone used for hip replacements, was collected from knee joints and femoral heads.

45. Members agreed that as a precautionary approach, they endorsed the use of a linear dose response model with no lower threshold of infectivity. Members questioned the starting scenario of 2 ID₅₀/mL infectivity for blood. This had been calculated from a dose of 10ID₅₀/mL from intracerebral (i.c.) challenge, and assuming the intravenous (i.v.) route of infection was five times less efficient. If, as suggested by some recent studies in primates, the i.v. route of infection was as efficient as the i.c. route, an estimate of 10 ID₅₀/mL may be more appropriate. It was noted, however, that this would not alter the main conclusions of the paper. The committee asked if experimental research could further inform on the level of infectivity in blood. The Chair responded that research was limited by assay sensitivity.

46. Members agreed with the approach using relative risks between the different bone products and agreed that the risk of infection would also depend on the prevalence of vCJD in the general population. SEAC was content with the assumptions made in the assessment and members agreed on the overall conclusion offered in the paper i.e. that use of cadaveric bone (batch dedicated if possible) minimises the risk of vCJD infection within the population, followed by single, washed/centrifuged femoral heads.

47. Members were informed that a wider consultation amongst healthcare professionals was planned which would examine the effect of CJD risk management options on the bone supply chain as well as non CJD-related

health issues, such as the mechanical properties of the various bone products.

ITEM 7 - TISSUE RISK ASSESSMENT

48. The Chair welcomed Dr Peter Bennett from the Department of Health (DH) to present this item.
49. The Department of Health's Economics and Operational Research (EOR) division and the National Blood Service (NBS) produced a draft assessment on the risks of vCJD transmission from tissue transplantation. A quantitative risk assessment was not thought feasible because of the scientific uncertainties surrounding the issue of infectivity in tissues.
50. Dr Bennett explained that the aim of the approach was to identify areas of concern and priorities for tissue infectivity experiments. The UK blood services Standing Advisory Committee on Transfusion Transmitted Infections (SACTTI) have previously advised on the potential infectivity of tissues. This supplements WHO guidance⁴ (annex B). A DH and NBS ad hoc expert committee on vCJD and tissues had collated information on clinical use of transplanted tissues (annex A).
51. Factors which may affect the level of risk from infection by transplantation were discussed, including the type of tissue, batching or pooling, number and mass transplanted, age of donor and recipient and the site of transplant. However lack of scientific data on the potential levels of infectivity in the various tissues made a quantitative risk assessment difficult. The scientific uncertainty could be addressed in part by using a linear dose-response model, as for blood infectivity, in which a dose of 2ID₅₀s or above was taken as sufficient to cause certain (or at least highly probable) infection. Given estimates for the mass of tissues transplanted, one could then calculate the infectivity per gram needed to reach this threshold. Sample calculations for corneal and sclera grafts were presented. These suggested that doses at or above this threshold could be reached with infectivities below the limit of sensitivity of current assays. As a consequence, negative results could not be taken to indicate that risks of transmission would be low.
52. Members were asked if more information was available on the use of tissue, as it may be possible to prioritise the assessment of each tissue in order of potential risk.
53. The committee asked if the approach aimed to assess the risk of infection by iatrogenic transmission in order to predict the size of a secondary epidemic of vCJD or if the aim was to minimise the potential for transmission by identifying tissues which would require risk management.

⁴ WHO guidelines on transmissible spongiform encephalopathies in relation to biological and pharmaceutical products

Members were informed that the assessment would be used for both purposes, and it was agreed to clarify this in the paper. One key use for the work would be to facilitate the development of specific measures to reduce the risk of infection on an individual basis, taking into consideration factors such as patient age.

54. Members commented that the number of donations to recipient was likely to be one key factor in assessing a risk of iatrogenic transmission. Members asked whether it would be possible to reduce risks of CJD transmission from tissues in a similar way to that previously done for HIV infected blood (i.e. did not pool samples from more than one donor). The committee was informed that in most examples of tissues used for transplantation, pooled donations were not used.
55. The committee agreed that the scientific uncertainty relating to tissue infectivity and the limitations imposed by assay sensitivity meant that it was difficult to conduct a risk assessment. Traceable systems, such as the one which identified the vCJD blood transfusion case, were acknowledged as an importance surveillance tool for identifying iatrogenic transmission.
56. The Chair summarised the committee view that the approach adopted in the paper was reasonable and it could not offer an alternative methodology. In response to a tabled question, members commented that an assessment, which focussed on the mononuclear cell count in a tissue, would be over-simplistic and that the origin of the tissue and site of transplant were also critical. More than one parameter would need to be considered to assess the tissue-specific risk.