

SPONGIFORM ENCEPHALOPATHY ADVISORY COMMITTEE

Minutes of the 73rd meeting held on 10 April 2002 at DEFRA,

Conference Room A, Whitehall Place West, London

- Members:**
- Professor P Smith (Chairman)**
 - Professor R Anderson (Items 1-4, 6, 9-10 only)**
 - Professor G Bulfield**
 - Professor C Bostock**
 - Professor R Carrell**
 - Dr D Cunningham**
 - Professor J Ironside**
 - Professor H Kimbell**
 - Mr P Jinman**
 - Professor C Masters**
- Technical Advisors:**
- Mr P Soul (DEFRA)**
 - Dr H Gates (DEFRA)**
 - Dr J Stephenson (DH)**
 - Ms A Conroy (FSA)**
- Observers:**
- Dr A Douglas (DARDNI)**
 - Dr M Simmons (NAW)**
 - Mrs L Shepherd (HSE)**
 - Dr N Coulson (DEFRA)**
 - Dr S Baxter (SEERAD)**
 - Dr A Allman (BBSRC)**
 - Dr K Finney (MRC)**
 - Dr D Matthews (VLA)**
 - Professor J Wilesmith (VLA)**
- Secretaries:**
- Dr M Bailey (DEFRA)**
 - Dr R Jecock (DH)**
 - Mr D Carruthers (FSA)**
- Secretariat:**
- Dr L Harbron (DEFRA)**
 - Mr H Needham (DEFRA)**
 - Dr A Leigh (DH)**
 - Mr M Hall (DH)**
 - Dr I Hill (FSA)**
- Also in attendance:**
- Dr D Reynolds (FSA)**
 - Dr L Tsang (MCA)- Item 3&4 only**
 - Dr H Ward (CJSSU)- Item 3&4 only**
 - Dr P Minor (NIBSC)- Item 3&4 only**
 - Dr S Cousens (LSHTM)- Item 3& 4 only**
 - Professor H Dalton (DEFRA) Morning only**
 - Dr Iram Malik (DH)**

Item 1- Chairman's introduction

1.1 The Chair welcomed Professor Grahame Bulfield following his appointment to SEAC. Professor Bulfield is the Director and Chief Executive of the Roslin Institute in Edinburgh, and brings expertise in genetics to the Committee.

1.2 Apologies for absence were received from Professor Collinge, Dr Safar, Professor Aguzzi, Professor McConnell and Mr Bradley. The Chair drew Members attention to the written comments submitted by Mr Bradley on several agenda items.

1.3 The Chair welcomed Professor Howard Dalton, DEFRA's new chief scientist, who was attending the meeting as an observer.

Item 2- Approval of Minutes (SEAC73/1)

2.1 Members considered the draft minutes from the previous meeting in February (SEAC 73/1). These had been published in draft form, subject to final agreement by Members.

2.2 The Chair noted that he had received correspondence from Professor Ebringer including a detailed critique of the relevant section of the minutes following SEAC's discussion of his work in February. Members considering the points raised by Professor Ebringer in detail, but agreed that the minutes were an accurate reflection of SEAC's views. The Committee agreed not to amend the record of discussions in light of Professor Ebringer's comments, other than to record that Professor Ebringer had not been present during some of the Committee's discussion of his work. It was agreed that the Chairman would write to Professor Ebringer informing him of the Committee's view.

Action: Chairman

2.3 Subject to the inclusion of the amendments above, Members agreed the minutes from February 2002. A final version would replace the draft minutes on the SEAC web-pages.

Action: Secretariat

Item 3- Vaccine History as part of CJD epidemiological investigations (SEAC 73/6A)

3.1 At the November 2001 meeting of SEAC there was discussion of two vCJD cases in Southampton who had received the same batch number of oral polio vaccine. The Committee concluded that this observation did not provide a reason to change the recent risk assessment by the Committee of Safety of Medicines regarding polio vaccines. The Committee had also requested an

update from the National CJD Surveillance Unit (NCJDSU) of work on epidemiological analysis of CJD cases and vaccine history.

3.2 Dr Ward, consultant epidemiologist, from the CJD Unit described the investigation used to compile a history of vaccines administered to vCJD cases. The vaccine history was compiled using reported history from relatives, GP records, school health records, health visitor records and health authority or health board records.

3.3 A total of 469 records of vaccination had been ascertained for the 69 vCJD cases whose vaccine histories had been fully investigated. Of these 469, 94 per cent had had the date recorded and 29 per cent had had the batch number recorded. The average number of vaccine doses received by each vCJD case was 7, with a range of 1 to 23. This compared to the experience of 77 controls who had 524 records of vaccination. Of the 524, 90 per cent had the date recorded and 41 per cent had the batch number recorded. The average number of vaccine doses received by each control was 7, with a range of 1 to 24.

3.4 The Committee noted that as more vCJD cases occurred the probability that pairs of cases would share the same vaccine batch number increased. Two pairs of vCJD cases had been identified in which vaccines from the same batches had been administered. In addition to the pair of cases of vCJD who had received polio vaccines from the same batch, two cases of vCJD had received diphtheria/tetanus vaccine from the same batch of vaccine in 1995. NCJDSU's preliminary analysis had found no link between the use of this vaccine and the two cases of vCJD. (Information provided after the meeting indicated that there were approximately 377,000 doses of vaccine in this particular batch distributed throughout the United Kingdom). It was noted that 3 pairs of controls for different cases had also received vaccines from the same batches. One pair had received Hepatitis B vaccine in 1994, one pair had received Typhoid vaccine in 1993 and one pair had received Typhoid vaccine in 1991.

3.5 The recording of batch numbers improved at later dates. Twelve per cent of the vaccine doses with batch numbers available given to the vCJD cases had been given before 1980, 25 per cent had been between 1980 and 1989 and 64 per cent after 1990.

3.6 The Committee requested that more detailed information on epidemiological analysis of CJD cases and vaccine history be made available to SEAC's Epidemiology Sub-Group to review the information. Their analysis could then be presented to SEAC for further consideration.

3.7 The Committee reiterated their concern, expressed at the November 2001 meeting, at the low level of recording of batch numbers by GPs that made

further epidemiological investigation very difficult. This might also be particularly a problem for privately administered vaccines. The Committee requested that the Department of Health investigate record keeping in this regard.

Item 4- Proposed testing of MRC 5 cells- abnormal prion protein and infectivity (SEAC 73/6B)

4.1 At their November 2001 meeting SEAC suggested that it might be of value to conduct further scientific tests for abnormal prion protein and infectivity of the MRC 5 cell line (used in the production of the oral polio vaccine).

4.2 The Committee was informed that bovine serum is the major biological component used in cell culture medium. Batches of serum used by the National Institute of Biological Standards and Control (NIBSC) have been traced. At NIBSC full documentation back to the individual cow was available from 1993, and certificates of analysis documenting New Zealand sourcing from 1985. Records indicated that NIBSC MRC5 cells have not been in contact with UK bovine foetal calf serum over the last 20 years. Custom and practice before 1985 was to use New Zealand or US sourced serum because of its better growth supporting qualities. In addition, the United States Drug Administration regulations specified that importation of products required New Zealand or US sourced serum.

4.3 The Committee noted that it was intended to examine MRC5 cells, grown and stored at various passage levels at NIBSC, for abnormal prion protein (by immunoblot after proteinase K treatment) and infectivity. Protein assays might take 3 months, while infectivity assays would take at least two years. The work started at the end of March 2002.

4.4 The Committee noted that it was intended to attempt to infect MRC5 cells with vCJD/BSE preparations (from brain homogenate). Results were unlikely to be available for 12 months. Negative controls would include MRC5 cells which could never have been in contact with UK calf serum and positive controls e.g. neuroblastoma cell lines which were known to become infected with TSEs. The work would start in early April 2002.

4.5 Members requested that the results of these tests be made available to them when the studies were completed.

Item 5- NSP working group report (SEAC 73/2)

5.1 Following the SEAC meeting in November 2001, it was agreed to convene a working group to examine various aspects of the National Scrapie Plan (NSP). The working group met in March, and SEAC considered a draft report of the meeting. The Chair noted that the report had not yet been cleared by members of the sub-group. However, there was some urgency to carry forward aspects of the NSP, and hence SEAC were invited to consider the draft conclusions arising from the working group's discussions.

5.2 The working group had reiterated that the scientific basis of the NSP was sound. The group had also agreed that it was not appropriate to delay the NSP until outstanding questions on issues such as carrier status and mechanisms of TSE transmission had been formally resolved. A twin track approach was appropriate where research to inform policy runs alongside the implementation of the scheme.

Modelling projections on the genetic profile of the flock and outstanding research questions

5.3 The working group had received presentations on preliminary modelling work to assess the potential time scales before the effect of the NSP is seen in the genetic profile of the National flock. The group also received an update on research work relevant to the NSP objectives, including work to examine the possibility that scrapie resistant sheep are able to carry and transmit infection. The working group was then invited to outline areas where additional research would be of benefit to further inform NSP policy. The working group highlighted the following areas:

- 1) Although DEFRA were funding some research work to examine the possible loss of positive traits by selective breeding under the NSP, the WG agreed that there was a limited overall strategy to examine this issue.
- 2) More information on the basic demography of the UK sheep flock would be helpful. This would be particularly beneficial to modelling work to examine the likely impact over time of the NSP on the genetic profile of the national flock.
- 3) There should be further work to improve understanding of the mechanism of scrapie transmission
- 4) Ongoing work to develop transgenic mice carrying the sheep PrP gene was of substantial value. However there should be more focus on the generation of mice with resistant ovine alleles for use in experiments to examine possible carrier states.

5.4 Members agreed that the working group had highlighted several key areas where research work would be beneficial to the NSP. However SEAC considered that the most important research question was work to examine the theoretical possibility that scrapie resistant animals may carry TSE infection. The assumption of a lack of scrapie infection in fully resistant animals underpinned the foundations on which the NSP was built. Members noted that a large body of research was underway. However work to examine possible carrier status should be explicitly stated as a primary research objective in relation to the NSP.

5.5 The working group had agreed that the limited experimental data on goats was of concern, particularly in regard to possible gene loci that may confer resistance to scrapie. SEAC Members noted that as a result of the apparent lack a scrapie resistant genotype in goats, currently there was no mechanism that allowed the selective breeding for scrapie resistance. Members agreed that work to investigate possible genetic resistance to scrapie in goats was an important research area. It was reported that DEFRA was currently in discussion with the EU to carry forward work on goats.

Action on Scrapie affected flocks

5.6 The working group had also considered possible action on scrapie affected flocks. They had concluded that:

- Taking action on identified sources of infection was valuable and hence action on scrapie affected flocks should be pursued.
- It was important to implement a policy that would be effective in removing potential sources of infection, encourage scrapie reporting and up-take in the scheme, but discourage ‘farming’ for scrapie. This was a very difficult balance to strike, and it was important that industry is fully consulted in order to bring them on board.
- Action should focus on genotyping all animals in an affected flock. NSP rules should be applied to restocking.
- Susceptible animals from scrapie affected flocks represented a small, but high-risk population as they were likely to also carry TSE infection. Hence additional measures to remove such animals from the food chain may be appropriate in order to reduce the theoretical risk to public health. However, it was important to maximise uptake to the scheme and additional control measures would only be of benefit if potential scheme participants were not inhibited from joining as a result.

- It may be appropriate to implement a less stringent course of action in some flocks where it was judged there was a low level of scrapie infection (e.g. if the scrapie infected animal was bought in or did not originate from the affected flock).

5.7 SEAC agreed that action to remove susceptible animals from scrapie infected flocks would reduce the theoretical risk to public health and concurred with the conclusions of the working group. SEAC re-iterated that it was vital that the farmers were not inhibited from reporting scrapie and hence it was important that the industry is consulted and was supportive of any proposed action.

Certified Flock scheme

5.8 The working group considered preliminary proposals on a certified flock scheme. This would allow farmers who had implemented breeding and management strategies to have their flocks certified if an appropriate level of genetic resistance to scrapie is reached. The working group concluded that:

- A certified flock scheme was scientifically justified, and would be a useful scheme to run alongside the NSP.
- It was important to produce pressure on susceptible genotypes. For that reason, it was not appropriate to provide a lower certification category for susceptible animals.

5.9 Members agreed that a certified flock scheme was consistent with the aims of the NSP and would be a useful parallel scheme. SEAC supported the working group's conclusions.

Safety of sheep entering the food chain if BSE was found in sheep

5.10 The FSA had asked the working group to consider previous SEAC advice from February 2001 (LINK) on the safety of sheep with certain genotypes entering the food chain if BSE was ever isolated from sheep. The working group concluded that:

- In line with previous SEAC advice, only animals carrying the ARR allele should enter the food chain.
- On a precautionary basis, the 12-month cut off previously advised by SEAC remained appropriate for ARR heterozygotes. However, in view of existing SRM regulations there was no justification for any age cut off in ARR homozygotes.
- In line with SEAC advice in 2001, only milk from ARR homozygote sheep can be considered as highly unlikely to contain the infectious agent. Further

experimental work was required before potential risks from small ruminant milk from goats and semi-resistant or susceptible sheep could be excluded.

5.11 SEAC concurred with the working group conclusions on animals that might be permitted to enter the food chain.

Item 6- Sheep Surveillance meeting report

6.1 At their previous meeting in February 2002, SEAC were given a brief outline of conclusions from a DEFRA working group that had met to consider surveillance for TSE infections in sheep. At this meeting, Members were asked to consider a more detailed report of the working group's conclusions on surveillance strategies to assess both the prevalence of scrapie infection in UK sheep and the possibility that BSE is present within the national flock.

6.2 The working group had recommended that any surveillance strategy should aim to detect a prevalence of TSEs in sheep of greater than 0.1%, and that an inverse sampling regime should be employed. Under this strategy, animals would be sampled until a specified number had tested positive. The working group recommended that testing should continue until 600 TSE positive animals had been identified. This would ensure that a large number of animals would be tested to assess the underlying prevalence of TSE infection in the national flock. The positive sheep would also supply a large number of TSE positive samples, which could be used to investigate the presence of a BSE-like strain.

6.3 Using the inverse sampling regime, if an overall TSE prevalence of 0.1% of the population is assumed, 600,000 animals would need to be tested to generate 600 positive cases. This would be ten times more than currently specified under EU rules which dictate that the UK must test 60,000 sheep in abattoirs and 6000 fallen stock.

6.4 There was some discussion about the merits of conducting such a large surveillance study. Some Members considered a large abattoir study along the lines recommended by the working group was the most appropriate strategy as it would provide both an accurate estimate of overall TSE prevalence and supply positive animals which could be examined using appropriate tests for the presence of BSE-like strain.

6.5 However, others felt that it would be better to separate the two goals and use the existing EU Sheep surveillance requirement to gauge the overall TSE prevalence, and concentrate on clinically affected scrapie cases as a source of material to examine the presence of BSE. Studying reported scrapie cases, supplemented by archived TSE tissue such as the 1,400 scrapie samples at the

VLA would provide a larger sample size than would be generated under the current working group proposals.

6.6 Some Members considered that testing the existing tissue bank and reported cases of scrapie for evidence of BSE was a more appropriate use of resources. However, it was noted that the archive was collected over a period of at least ten years and hence it would be difficult to extrapolate results to the present day. The reporting of clinical scrapie was also unreliable because estimates suggest that only a minority of farmers report clinical suspects. This would inevitably bias the samples received for detailed testing. Without a random sample collected at the given point in time, it would be difficult to provide confidence bounds on the possible prevalence of BSE in the current flock given a negative result. A random sample would also allow a greater scope to further detailed analysis such as an assessment of geographical location.

6.7 Overall Members agreed that the primary objective was to test the maximum number of TSE sheep samples for evidence of BSE. The consensus was that clinical scrapie cases presented the most efficient source of material. Hence a representative sample of reported scrapie cases should be used in the first instance. However, this should not exclude other approaches.

6.8 Members agreed that gaining a further understanding of the overall prevalence of TSE infection was also vital. It was considered that the EU surveillance requirement of 66,000 sheep would provide a reasonable sample size with which to assess the current prevalence of TSE in the UK flock. It would also provide a baseline figure, which could be used in conjunction with information on the genetic profile of the flock to model scrapie and the possible level of BSE in the national flock. However, the EU sample requirements were not thought to be adequate to evaluate any subsequent changes in TSE prevalence as a result of the current action to reduce and eliminate scrapie under the National scrapie Plan.

6.9 Irrespective of the methods employed to gather data, Members reiterated that further information on sheep TSEs in the national flock was the highest priority. Members agreed that further work to ascertain if BSE was present in UK sheep and surveillance to examine the overall prevalence of TSE in the sheep flock should be implemented as soon as possible.

Item 7- Update on VLA research

Proposals for further evaluation of the differential diagnostic test for TSEs in sheep

7.1 The Committee was given a brief update on progress at the Veterinary

Laboratories Agency (VLA) to develop and validate a rapid biochemical test to discriminate BSE from scrapie in sheep. The VLA was currently assessing the discriminatory ability of the test using statistical analysis. This was not yet complete, but as a result of refinements to the test methodology, the raw data indicated sufficiently consistent performance to be optimistic that the test methods would be able to distinguish between experimental BSE and scrapie.

7.2 It was possible that the genetics of the infected sheep may affect the pattern seen on the western blot. This could affect the ability of the test to discriminate between different TSE strains. Hence the VLA had assessed variation in the patterns on the western blot from sheep with a range of genotypes. Currently, the pattern associated with experimental BSE remained consistent.

7.3 Experiments had also been carried out to assess if the natural breakdown of sample tissue between collection and testing (autolysis) affected the discriminatory ability of the test. Analysis suggested that although autolysis does effect the pattern seen on the western blot to some degree, the effects to date were relatively. However, it does reduce the effectiveness of the signal antibody to bind to the protein.

7.3 In light of the potential problems created by autolysis of tissue samples, it was considered that more effort should be made to ensure that tissues are collected and tested more rapidly following euthanasia. However it was noted that there were often practical difficulties and welfare issues involved with transporting scrapie suspect sheep to Veterinary Centres. Many scrapie suspects were also found dead on farm. However the VLA were working to encourage veterinarians to improve the quality of samples submitted.

Action: VLA

7.4 Because of the limited availability of tissue from sheep experimentally infected with BSE, it was the VLA's intention to use bovine BSE as the comparative control material as part of the initial screen for scrapie samples. VLA remained confident that this material was able to distinguish between BSE and scrapie during initial screening as the smallest molecular weight band was distinctively low from BSE-derived isolates. Experimental sheep BSE material would then be used as control material if further investigation was warranted.

7.5 The VLA hope to have sub-passaged BSE material (i.e. material from an infected sheep that had been inoculated with experimental sheep BSE) in the near future. Although scarce, this would provide important control material with which to compare scrapie isolates that display non-typical patterns using the test. This was important in view of the likelihood that if BSE was present in the flock, it was likely to have been passed through several generations of

sheep. Hence sub-passaged control material may mimic the sub-passaged BSE agent strain if it exists in the field.

7.6 Given the improved definition on the western blot, the VLA were in a position to re-examine their archive of 1400 natural scrapie cases using the refined test. However it was important that the test could be interpreted with confidence prior to examining the archive as in many cases the sample material was in short supply and repeat testing was not possible. SEAC noted that currently there was a some pressure on the VLA archive staff, but agreed that the testing of sheep brains to define agent strain was a high priority and every effort should be directed at making archive tissue available for this work.

Action: VLA

7.7 The VLA also intends to implement a study to evaluate the performance of various molecular approaches to discriminate between BSE and scrapie in sheep. Members agreed that it would be useful to compare other differential diagnostic tests to those developed at the VLA. However some concern was expressed about attempting to evaluate too many approaches in parallel.

Item 8- Prion in mouse skeletal muscle

8.1 Members considered a paper published in Proceedings of the National Academy of Science USA, vol. 99, 3812-3817 (March 19, 2002) by P.J. Bosque *et al* entitled 'Prions in skeletal muscle'. This paper reports on work with mice inoculated with mouse- (or hamster-) passaged scrapie. Wild-type mice and cattle both showed expression of normal prion in skeletal muscle at a level of 5-10% of that in brain, when analysed by Western blotting. In addition wild-type mice that had reached clinical disease following intracerebral inoculation with scrapie were analysed for expression of scrapie prion (PrP^{Sc}). Based on an incubation time assay in transgenic mice over expressing the mouse prion, infectivity levels in muscle were 1/10,000 of that found in brain. Confirmation of these findings by Western blotting showed that (with one exception) PrP^{Sc} was confined to the hind limb muscle at approximately 1/1000 of that in brain (no PrP^{Sc} was detected in head and neck muscle, forelimb and back muscles). Further experiments were carried out with transgenic mice to confirm that the accumulation of PrP^{Sc} in skeletal muscle was specific to the muscle and not an experimental artefact.

8.2 Members were given an update on ongoing research in which muscle samples from bovines infected with BSE are being bioassayed, without a species barrier, in cattle. No cattle being used in the assay have shown any signs of disease to date, some 42 – 65 months, post inoculation with muscle. There was also discussion as to whether the measured muscle infectivity in mice by Bosque *et al* might derive from nervous tissue within the muscle, since PrP^{Sc} has been detected in the peripheral nervous system (PNS) of sheep and

rodent models infected with scrapie. However all experiments on bovine PNS have given negative results.

8.3 Members considered that the work reported in this paper could not directly be applied to BSE in cattle, scrapie in sheep or vCJD in humans. This, therefore, does not change the risk assessment with respect to the likelihood of BSE infectivity in cattle with BSE or of infectivity in sheep should BSE be found to affect sheep, or the risks of vCJD associated with the reuse of surgical instruments. Nevertheless the Committee reiterated their previous advice that decontamination of surgical instruments is key in minimising any potential for person to person spread of vCJD. In relation to bovine BSE, the Committee concluded that the findings did not alter the assessment of the risk to humans of consumption of beef.

Item 9- vCJD update

9.1 The Committee conducted its regular review of epidemiological information on vCJD. The Committee was informed that the total number of definite or probable vCJD cases in the UK stood at 118 (89 confirmed cases, 18 who had not had post mortems, 3 dead awaiting post mortem and 8 still alive). There were 6 cases in France where the youngest case was 17 years old at onset, 1 in the Republic of Ireland and 1 in Italy. The Committee considered that the relatively high incidence of vCJD cases in France compared to the relatively low level of reported BSE cases in France could be due to the considerable amount of British culled cows (carcasses) exported to France before the export ban.

9.2 The Committee noted that there were 64 males and 54 females cases in the UK but this was not a statistically significant difference. The mean age at death for both genders was 29 years and at onset was 28 years. These figures were very similar to the mean ages of death and onset in France and Ireland. It remained the position that all of the cases tested for their prion protein (PrP) genotype, 99 in total, were Methionine/Methionine at codon 129 of the PrP gene (37 per cent of the UK population being Methionine/Methionine).

9.3 The Committee noted an analysis from the Public Health Laboratory Service (January 1994 – December 2001), which showed that the increase in the number of vCJD cases since 1995 continued to be significant, on average increasing at a rate of 21 per cent per year for onsets and 23 per cent per year for deaths. This analysis was available on the National CJD Surveillance Unit website: www.cjd.ed.ac.uk. The Committee emphasised that it was too early to forecast longer-term trends of the disease with any certainty.

Item 10- Committee business

Open meetings update

10.1 The Secretariat updated Members on plans for the SEAC meeting in September, when the Committee would begin to hold its meetings in public. Possible venues were currently being assessed.

Action: Secretariat

Item 11- Departmental Research

11.1 This item was deferred until the next meeting.

Item 12- Intra-species recycling

12.1 This item was deferred until the next meeting. However Members requested that the information presented on intraspecies recycling in terms of the sourcing and potential uses of material was collated into a table for ease of reference.

Action: Secretariat

SEAC Secretariat April 2002

List of scientific papers supplied to SEAC Members by the Secretariat between 07 February and 10 April 2002

Report “Confusion over experiment on BSE in sheep” published in The Veterinary Record, 2001, Vol 149, page 502.

Paper by F Heppner, I Arrighi, U Kalinke, and A Aguzzi, “Immunity against prions?” published in Trends in Molecular Medicine, 2001, Vol 7, pages 477-79.

Paper by R A Armstrong, P L Lantos and N J Cairns “ Correlations between the Clustering Patterns of the Pathological Changes in Sporadic Creutzfeldt-Jakob Disease” published in Neuroscience Research Communications, 2001, Vol 29, pages 89-98.

Paper by H Funke – Kaiser, S Theis, T Berhouzi, A Thomas, K Scheuch, F S Zollman, M Paterka, M Paul, H D Orzechowski, “Functional characterization

of the human prion protein promoter in neuronal and endothelial cells” published in the Journal of Molecular Medicine 2001, Vol 79, pages 529-535.

Article “Research gives hope for CJD drugs” published in Chemistry in Britain, 2001, Volume 37, page 9.

Paper by F Heppner, C Musahl, I Arrighi, M A Klein, T Rulicke, B Oesch, R M Zinkernagel, U Kalinke, A Aguzzi, “Prevention of Scrapie Pathogenesis by Transgenic Expression of Anti-Prion Protein Antibodies”, published in Science, 2001, Vol 294, pages 178-182.

Article “Japan’s beef scandal” published in Nature, Vol 413, page 333.

Article “Protocol For UK CJD trial due soon” published in Scrip World Pharmaceutical News 2001, page 6.

Paper by Michael Balter “Uncertainties Plague Projections of vCJD toll” published in Science, 2001 Vol 294, pages 770-1.

Article “ Health Department expands plan combating BSE, TSE” published in Journal of the American Veterinary Medical Association, 2001, Vol 219 page 889.

Scientific Paper by F L Heppner, M Printz and A Aguzzi, “Pathogenesis of prion diseases: possible implications of microglial cells”, published in Progress in Brain Research, 2001, Vol 132, page 737-750.

Scientific Paper by J W Ironside, D Seilhean, M W Head and J J Hauw, “ Investigation of Prion Diseases”, published in Current Topics in Pathology, 2001, Vol 95, pages 179-205.

Scientific Abstract by J Ironside, “Human Prion Diseases:genetic and transmissible disorders” published in Journal of Medical Genetics, 2001, Vol 38, page PS30.

Scientific Report “National Scrapie Plan to be accelerated” published in the Veterinary Record, 2001, Volume 149, page 434.

Scientific Reports, “Defra considers worst case scenario in its contingency plan for BSE in sheep” and “ FSA’s advice on the theoretical risk”, published in the Veterinary Record, 2001, Vol 149, pages 402-3.

Report “BSE confirmed in Japan” published in the Veterinary Record, 2001, Vol 149, page 371.

Paper by R Chiesa and D Harris, “Prion Disease: What is Neurotoxic Molecule?” published in *Neurobiology of Disease*, 2001, Vol 8, pages 743-763.

Paper by C Drogemuller, T Leeb and O Distl, “PrP genotype frequencies in German breeding sheep and the potential to breed for resistance to scrapie” published in the *Veterinary Record* 2001, Vol 149, pages 349-352.

Paper by P Calvo, B Gouritin, I Brigger, C Lasmezas, J-P Deslys, A Williams, J P Andreux, D Dormont and P Couvreur, “PEGylated polycyanocrylate nanoparticles as vector for drug delivery in prion disease” published in the *Journal of Neuroscience Methods*, 2001, Vol 111, pages 151-155.

Paper by B Van Everbroeck, E A Croes, P Pals, B Dermaut, G Jansen, C M van Duijn, M Cruts, C Van Broekhoven, J-J Martin and P Cras, “Influence of the prion protein and the apolipoprotein E genotype on the Creutzfeldt-Jakob Disease phenotype”, published in *Neuroscience Letters*, 2001, Vol 313, pages 69-72.

Paper by T J Hagenaars, N M Ferguson, C A Donnelly and R M Anderson, “Persistence patterns of scrapie in a sheep flock” published in *Society for Veterinary Epidemiology and Preventive Medicine*, 2001, Vol 127, pages 157-167.

Paper “Codon 129 polymorphism of the *PRNP* gene in normal Polish population and in Creutzfeldt – Jakob disease, and the search for new mutations in *PRNP* gene”, published in *ACTA Neurobiologiae Experimentalis*, 2001, Vol 61, pages 151-6.

Paper by D Collie, R J Sellar, M Zeidler, A C F Colchester, R Knight, and R G Will, “MRI of Creutzfeldt-Jakob Disease: Imaging Features and Recommended MRI Protocol”, published in *Clinical Radiology*, 2001, Vol 56, pages 726-739.

Paper by C Gabus, S Auxilien, C Pechoux, D Dormont, W Swietnicki, Manuel Morillas, W Surewicz, P Nandi and J-L Darlix, “The Prion Protein has DNA Strand Transfer Properties Similar to Retroviral Nucleocapsid Protein”, published in the *Journal of Molecular biology*, 2002, Vol 307, pages 1011-1021.

Report by C Crozet, A Bencsik, F Flamant, S Lezmi, J Samarut and T Barron, “Florid plaques in ovine PrP transgenic mice infected with an experimental ovine BSE”, published in *Embo-Reports*, 2001, Vol 2, pages 952-956.

Paper by E A Croes, G H Jansen, A W Lemstra, C J M Frijns, W A van Gool, C M van Duijn, “The first two patients with dura mater associated Creutzfeldt-

Jakob disease in the Netherlands, published in Journal of Neurology, 2001, Volume 2, pages 952-956.

Paper by Paul Brown, “Creutzfeldt – Jakob Disease: Blood Infectivity and Screening Tests”, published in Seminars in Hematology, 2001, Vol 38, pages 2-6.

Article by D A Collie, “The role of MRI in the diagnosis of Sporadic and Variant Creutzfeldt-Jakob Disease” published in Clinical Radiology, 2001, Vol 84, pages 143-146

Paper (translated from German) “Bovine Spongiform Encephalopathy (BSE) in Cattle and its Transmissibility to Humans”, published in Bundesgesundheitsbl-Gesundheitsforsch-Gesundheitsschutz, 2001, Vol 44, pages 421-431.

Paper by J I Kourie, “Mechanisms of prion – induced modifications in membrane transport properties: Implications for signal transduction and neurotoxicity” , published in Chemico Biological Interactions, 2001, Vol 138, pages 1-26.

Paper by J D Foster, D W Parnham, N Hunter and M Bruce, “Distribution of the prion protein in sheep terminally affected with BSE following experimental oral transmission” published in Journal of General Virology, 2001, Vol 82, pages 2319-2326.

Article “Japan to test a million cattle after discovery of first case of BSE”, published in the British Medical Journal, 2001, Vol 323, page 713.

Paper by N Mabbot and M E Bruce, “The immunobiology of TSE diseases”, published in Journal of General Virology, 2001, Vol 82, pages 2307-2318.

Paper by R Love “Antibodies effective against scrapie infection, Report European researchers”, published in The Lancet, 2001, Vol 358, page 816.

Paper by A Ladogana, S Almonti, R Petraroli, E Giaccagliani, C Ciarmatori, Q G Lui, S Bevivino, F Squiteri and M Pocchiari, “Mutation of the *PRNP* gene at Codon 211 in Familial Creutzfeldt-Jakob Disease”, published in American Journal of American Genetics, 2001, Vol 103, Pages 133-137.

Letter by J M Scudamore, “EU requirement to survey for BSE in fallen stock:consequences for post mortem examination”, published in The Veterinary Record, 2001, Vol 149, page 367.

Paper by N F McLennan, K A Rennison, J E Bell and J W Ironside, " *In situ* hybridization analysis of PrP mRNA in human CNS tissues", published in *Neuropathology and Applied Neuropathology*, 2001, Vol 27, pages 373-383.

Paper by R Race, A Raines, G B Raymond, B Caughey and B Chesebro, "Long-Term Subclinical Carrier State Precedes Scrapie Replication and Adaptation in a Resistant Species: Analogies to Bovine Spongiform Encephalopathy and Variant Creutzfeldt – Jakob Disease in Humans", published in *Journal of Virology*, 2001, Vol 75, pages 10106-10112.

Paper by P Hopp, M J Ulvund and J Jarp, " A case-control study on scrapie in Norwegian sheep flocks", published in *Preventive Veterinary Medicine*, 2001, Vol 51, pages 183-198.

Abstract by C Pomfrett, "Analysis for the Presence of Degenerative Brain Disease" published in *Official Gazette of the United States patent and trademark office*", 2001, Vol 1245, page 2888.

Article by B Matthews, "BSE/TSE Risks Associated with Active Pharmaceutical Ingredients and Starting Materials: The Situation in Europe and the Global Implications for Healthcare Manufacturers" published in *Journal of Pharmaceutical Science and Technology*, 2001, Vol 55, pagew 295-328.

Abstract by S B Prusiner and J G Safar, "Removal of Prions from Blood, Plasma and Other Liquids" published in *Official Gazette of the United States Patent and Trademark Office*, 2001, Vol 1245, page 4277.

Letter by M Tagaya, "BSE fostered by cosiness and lack of independent advice", published in *Nature*, 2001, Vol 414, page 147.

Paper by G A Venters, "New variant Creutzfeldt-Jakob disease: the epidemic that never was" , published in the *British Medical Journal*, 2001, Vol 323, pages 858-861.

Paper by P Valenti, A Cozzio, N Nishida, D P Wolfer, S Sakaguchi and H P Lipp, " Similar target, different effects: late-onset ataxia and spatial learning in prion protein – deficient mouse lines", published in *Neurogenetics*, 2001, Vol 3, pages 173-4.

Paper by J Kulczycki, W Lojkowska and K Niedzielska, "Epidemiological studies on Creutzfeldt – Jakob disease in Poland" published in *Folia Neuropathologica*, 2001, Vol 39, pages 175-9.

Letter by A Hoek, R D Eglin, C Van Coller, M Ali, and L Hoinville, “Scrapie: improving notification” published in The Veterinary Record, 2001, Vol 148, page 216.

Paper by P Libereski, D Guiroy, E S Williams, A Walis, H Budka, “Deposition patterns of disease-associated prion protein in captive mule deer brains with chronic wasting disease” published in Acta Neuropathologica, 2001, Vol 102, pages 496-500.

Paper by M Doherr, D Heim, R Fatzer, C H Cohen, M Vandavelde and A Zurbriggen, “Targeted screening of high-risk cattle populations for BSE to augment mandatory Reporting of clinical suspects”, published in Preventive Veterinary Medicine, 2001, Vol 51 , pages 3-16.

Paper by P Ostlund, H Lindegren, C Pettersson and K Bedecs, “Up –regulation of Functionally Impaired Insulin-like Growth Factor-1 Receptor in Scrapie-Infected Neuroblastoma Cells” published in the Journal of Biological Chemistry, 2001, Vol 276, pages 36110-36115.

Paper by T Pan, M Colucci, B-S Wong, R Li, T Liu, R Petersen, S Chen, P Gambetti and M-S Sy “Novel Differences between Two Human Prion Strains Revealed by Two – Dimensional Gel Electrophoresis*” published in the Journal of Biological Chemistry, 2001, Vol 276, pages 37284-37288.

Paper by B Schroder, B Franz, P Hempfling, M Selbert, T Jurgen, H Kretzshmar, M Bodemer, S Poser, I Zerr, “Polymorphisms within the prion-like protein gene (*Prnd*) and their implications in the human prion disease, Alzheimer’s disease and other neurological disorders”, published in Human Genetics, 2001, Vol 109, pages 319-325.

Paper by V Vitagliani and G D’Errico, “A simple kinetic model to describe the progression of prion disease” published in Physical-Chemistry-Chemical-Physics, 2001, Vol 3, pages 4547-4550.

Paper by J G Vostal, K Holada and J Simak, “Expression of Cellular Prion Protein on Blood Cells: Potential Functions in Cell Physiology and Pathophysiology of Transmissible Spongiform Encephalopathy Diseases”, published in Transfusion Medicine Reviews, 2001, Vol 15, pages 268-281.

Paper, (translated from German), by E Kolb, “Are there links between BSE and transmissible mink encephalopathy? Did BSE exist even earlier than 1985? (An overview)” published in Tierärztliche Umschau, 2001, Vo 56, pages 507-11

Paper, (translated from German), by G Hilderbrandt, “BSE-risk concerning the consumption of meat and milk”, published in Bundesgesundheitsblatt – Gesundheitsforsch – Gesundheitsschutz, 2001, Vol 44 pages 437-449.

Paper by B Van Everbroeck, P Pals, S Quoilin, J-J Martin and M Cras, “The many faces of human prion diseases in Belgium and the world”, published in Acta Neurologica Belgica, 2001, Vol 101, pages 81-87.

Paper by S Taylor, K Green, I Smith and C Peers, “Prion protein fragment 106-126 potentiates catecholamine secretion from PC-12 cells”, published in American Journal of Physiology Cell Physiology, 2001, Vol 281, pages C1850-1857.

Paper by C Thielen, F Melot, O Jolois, F Leclercq, R Tsunoda, Y Frobert, E Heinen, N Antoine, “Isolation of bovine follicular dendritic cells allow the demonstration of a particular cellular prion protein.