

Prion Diseases: Fatal and greatly feared.

**A review of current knowledge, two and a half centuries after the 1st
description of scrapie.**

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01.10.10



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ABSTRACT

Prion diseases (or transmissible spongiform encephalopathies) are rare, infectious neurodegenerative disorders caused by the accumulation of aberrant prion proteins in the brain. TSEs can affect both animals and humans and can be transmitted with varying degrees of efficiency between species. Prion disease can be sporadic, genetic, or acquired. Testing in humans is not feasible, yet, alarmingly, vCJD has been shown to transmit through blood transfusions. Other patients have been iatrogenically infected from surgical instruments that were contaminated with prions. Prions are extremely difficult to inactivate and standard disinfection methods for surgical equipment are inadequate. There are at present time no effective treatments for TSEs in humans or in animals. The incubation time is generally long, but death usually occurs within months after the onset of clinical disease. The aim of this paper is to present an overview of the prion diseases, including medical, historical, legal, and economical perspectives.

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I. Introduction

Medical students at the University of Oslo are required to write a thesis paper on a topic of their own choosing. I have chosen prion diseases, a topic that has interested me since the first reported case of mad cow disease in the United Kingdom in 1996.

I am a native of Seattle, Washington. I returned for a visit to Seattle in December 2003 at the same time that the U.S. had its first case of mad cow disease. A dairy cow from a farm in Mabton, Washington was reported to have had mad cow disease, and the meat had been consumed by people in Washington state.

The Holstein cow had been imported from Alberta, Canada in 2001. The animal was slaughtered on Dec. 9, 2003. It had apparently become paralyzed while giving birth and was thus considered a “downer cow”, meaning that it was unable to walk at the time of slaughter¹. Because it was a downer cow, screening tests for bovine spongiform encephalopathy (BSE or mad cow disease) were taken and shipped to the United States Department of Agriculture’s laboratory in Iowa¹. The most high risk parts of the animal; the brain, spinal cord, and intestines, were sent to a rendering plant that turns animal tissues into soaps and other non-food products. However, slaughtering processes can still involve some risk of contamination of the “safe” parts of the animal by the infectious tissues. The meat for human consumption was processed at three different plants in Washington and Oregon, and subsequently shipped to wholesalers and retailers in Washington, Oregon, California, Idaho, Montana and Nevada². After the positive test results on Dec. 23, 2003, the farm in Mabton was quarantined and efforts were made to trace the potentially contaminated meat. USDA officials subsequently “depopulated” the Mabton herd, other Washington and Idaho herds associated with the infected animal, and the herd containing the sick Holstein’s calf. All of the euthanized animals, approximately 500 cattle, tested negative for BSE¹.

Businesses began pulling potentially infectious beef products from store shelves under a voluntary recall starting December 23rd. The total amount of meat recalled, consumed, or thrown away by consumers is uncertain, estimates range from 10000 pounds to 38000 pounds^{1,2}. USDA officials, fearing a drop in beef sales and exports, were quick to try to reassure consumers of food safety. The discovery of BSE in a Canadian cow earlier in 2003 had resulted in the US, Japan, Korea, Australia, and other countries issuing import bans on Canadian cattle, with catastrophic effects for the cattle industry in Canada. The United States Secretary of Agriculture Ann Veneman stated she was serving beef to her family for their Christmas dinner, and the town of Mabton had a free barbecue a few weeks after the mad cow discovery.^{1,3} Consumers who were concerned about food safety could employ extra precautions by buying only whole-muscle cuts of beef, rather than ground beef.³

The mad cow discovery caused a great deal of fear among Seattle residents who had potentially consumed contaminated beef products, myself included. One resident, a Jill Crowson, even went so far as to file a class-action (group, meaning others can join in) lawsuit against the grocery store chain QFC.² QFC had been warned of the meat contamination Dec. 23rd, yet didn’t remove the meat from the shelves until Dec. 24th.² The grocery store chain did not try to warn its

customers about the recalled beef until Dec. 27th, and at that time only with small, easily missed signs at the stores. The QFC stores have a customer rewards program, meaning that shoppers use a QFC store card each time they shop, effectively tracing all purchases. Crowson's argument was that QFC should have used their tracking data to warn consumers. Jill Crowson had purchased ground beef from QFC on Dec. 22nd and Dec. 23rd and had used it to prepare meals for her family.² In January, she contacted QFC and was informed that the beef had indeed been part of the batch of meat that was recalled.² This student has not been able to find the results of the Crowson versus QFC lawsuit, even after enlisting the help of a friend who is a lawyer. The probable conclusion is that Crowson and QFC reached a settlement before the case went to court.⁴ For Crowson and for others who may have consumed infected meat, living with the knowledge that they and their families may be bearers of this fatal disease is a heavy emotional burden.

Mad cow disease and the possibility of its transmission to humans has interested me since I first heard of it in the 1990s. I have a personal interest in mad cow disease, as I have close family living in the UK and in Seattle. My parents, sisters and I consumed beef products in Seattle at the time of the 2003 mad cow discovery. The uncertainty of knowing that one may be at risk for Creutzfeldt-Jakob disease is something we have to live with, as there is no screening test for it at present. With this paper, I hope to give a concise overview of what we know about prion diseases today.

II. Methods

I have searched PubMed Clinical queries using the Mesh terms/search terms Prion Diseases; Creutzfeldt-Jakob syndrome; Fatal Familial Insomnia; Kuru; Gerstmann-Straussler-Scheinker Disease; Bovine Spongiform Encephalopathy; Scrapie; Wasting Disease, Chronic; and Transmissible Mink Encephalopathy and gone through the systematic reviews listed. Limits imposed on the searches were for English and links to full text. I have also researched newspaper articles and websites for consumers. A lawyer who is a personal friend has performed searches on paid legal research sites for me related to the mad cow case in Seattle. I have used the websites of the Center for Disease Control in the USA, the Veterinary Institute in Norway, and United States Department of Agriculture Animal and Plant Health Inspection Service.

III. Human Prion Diseases

Prion is a combination of two words, infectious and protein. Prion diseases are inevitably fatal. Human prion diseases include Creutzfeldt-Jakob Disease (CJD), Gerstmann-Straussler-Scheinker syndrome (GSS), familial fatal insomnia (FFI), and kuru. CJD has four subclassifications, including sporadic (about 90% of all CJD cases occur sporadically), familial, iatrogenic, and variant. Familial CJD, GSS, and FFI are all genetic prion diseases with an autosomal dominant pattern of inheritance. Kuru, iatrogenic CJD, and variant CJD are all acquired forms of prion disease.^{5,6}

The prion gene (PRNP) encodes for the prion protein PrP^c. In prion disease, PrP^c undergoes post-translational modification to the disease-related protein PrP^{Sc}.⁷ The precise way in which PrP^{Sc} infects the brain is still unclear; it seems to be able to induce a conformational change in the normal prion protein PrP^c.⁵

A. Creutzfeldt-Jakob Disease

Background

Creutzfeldt-Jakob Disease (CJD) is a rare, rapidly progressive neurodegenerative disease caused by infectious prions. Prion diseases are inevitably fatal. H.G. Creutzfeldt is credited with the first clinicopathological description of the disease now known as CJD in 1920. In the following year another German neurologist, A. Jakob, described four cases of what we now find highly suggestive of CJD. Hence the name Creutzfeldt-Jakob disease.⁸

Variant CJD was first described in 1996 in the United Kingdom.⁹ It has been undeniably linked to the outbreak of mad cow disease, or Bovine Spongiform Encephalopathy.⁷ There are various clinical and pathological features which distinguish sporadic CJD from variant CJD. Sporadic CJD has occurred throughout the world, whereas variant CJD has occurred mainly in the UK and France.¹⁰

How does vCJD differ from sporadic CJD?

Sporadic CJD is primarily a disease of older people, with a median age at death of 68 years. Variant CJD affects younger people, with a median age at death of 28 years. Sporadic CJD tends to present as dementia, whereas vCJD presents with changes in behavior, psychiatric problems, and painful dysesthesias. Neurological signs appear early in sporadic CJD and later in variant CJD. EEG changes are often present in sporadic CJD, and often absent in variant CJD. The pulvinar sign on brain MRI imaging is present in over 75% of variant CJD cases, and absent in sporadic CJD. In variant CJD (but not in sporadic CJD) deposition of prion amyloid protein is found in lymphoid tissues.⁹

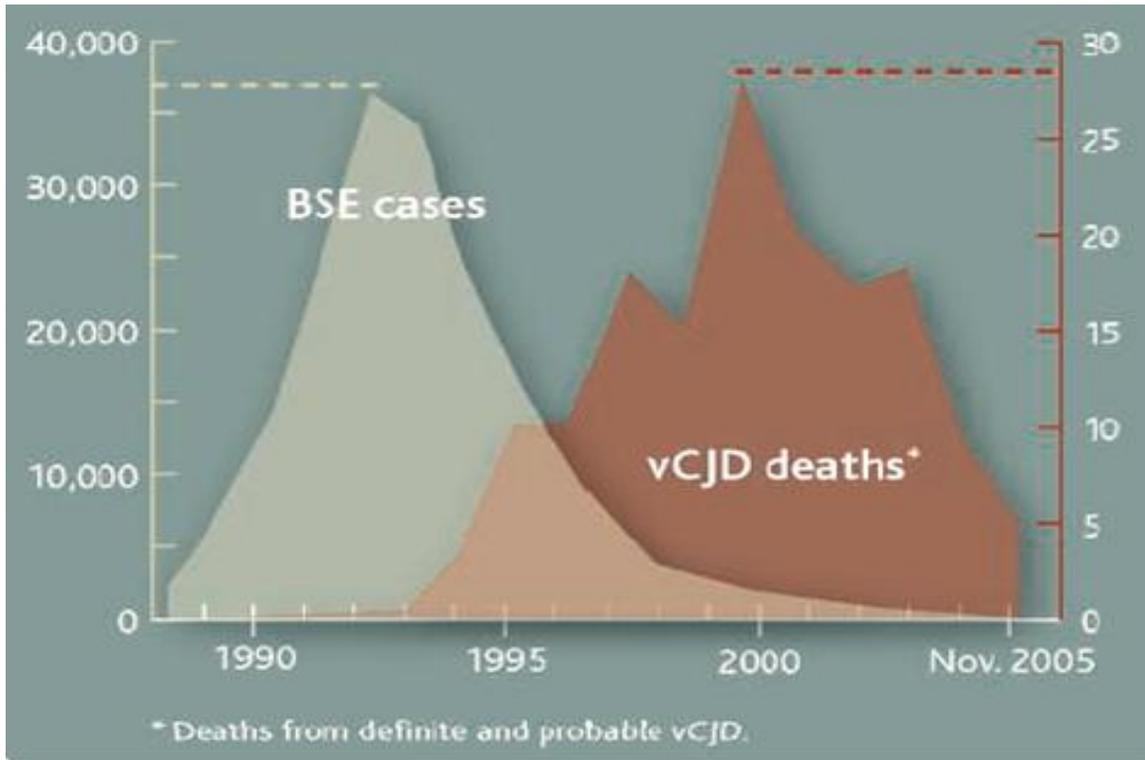
Occurrence and Transmission

CJD is rare, with only 1-2 cases per million people per year¹¹ (in Norway about 5 cases per year). Most cases of sporadic CJD occur after age 50. A cause for sporadic disease has not been found but it has been speculated that somatic mutation of the prion protein gene or spontaneous change to prion proteins in the brain may be involved.¹² All cases of familial prion disease are caused by germ line mutations in the prion protein gene.¹¹ Iatrogenic CJD is caused by the transfer of prions by transplantation, hormones derived from human cadavers, blood transfusion and or contaminated surgical instruments.¹³

Variant CJD is believed to be transmitted through the consumption of cattle products infected with Bovine Spongiform Encephalopathy (BSE).¹⁰ About 200 people in the world have been diagnosed with vCJD after the first ten cases were described in 1996¹⁴, most of these in the United Kingdom.¹⁵ A sharp decline in the number of new cases has occurred after the year 2000. The timing of the vCJD epidemic, which peaked some years after the maximum of the BSE outbreak, was consistent with the long incubation period for prion diseases. It is thought that the BSE epidemic was the result of feeding cattle with meat and bone meal contaminated with prions from a new type of prion disease that may have originated in sheep or another animal.¹⁶ The practice of using mechanically recovered meat represents a certain risk of contamination of meat with brain and spinal cord tissues.¹⁶

There is compelling evidence supporting the theory that variant CJD is a human form of BSE. An important piece of evidence rests on the fact that the glycoform pattern of PrP found in vCJD is similar to that of BSE (PrP exists in three different glycoforms: diglycosylated,

monoglycosylated and unglycosylated form; the diglycosylated form dominates in BSE and vCJD).¹⁷



Courtesy of Prof. Jan Mæhlen, UUS

vCJD has also been transmitted through blood transfusion from infected, asymptomatic blood donors.¹⁸ A study by Hewitt and Will identified three likely cases of transmission of vCJD through blood transfusions in the UK.¹⁸ Individuals who have lived in the UK for more than one year between 1980 and 1996 are permanently banned as blood donors in Norway, as are persons who have received blood transfusions in the UK after 1980.¹⁹ This is due to the risk of transmission of vCJD.

Symptoms and signs

Appleby *et al.*¹³ found that the most common presenting symptom for all CJD types is a cerebellar or gait disturbance. Dementia, cognitive decline, and memory impairment are also common at presentation. Visual or oculomotor disturbances are frequent, as are changes in behaviour and personality. Myoclonus is also frequently reported as the illness progresses(3). Appleby *et al.* conducted a meta-analysis¹³ to investigate whether the disease presentation varies between CJD types and with age. Variant CJD patients commonly presented with affective illnesses and other psychiatric symptoms, whereas sporadic CJD patients most often presented with neurological symptoms. vCJD patients also had more sensory complaints. Genetic forms of CJD tended to present with cerebellar/gait disturbances, dementia/memory impairment/higher cerebral dysfunction. Patients infected with CJD iatrogenically tended to present with delirium and confusion. This may be due to brain damage from dural graft procedures.¹³

In addition, Appleby *et al.* found that age can explain differences in the symptom presentation of CJD, regardless of the etiology: “Younger age was associated with a higher likelihood of presenting with cerebellar/gait disturbance, visual/oculomotor disturbance, sensory disorder, speech disturbance, seizures, vertigo/dizziness, headache, and apraxia... Additionally,

younger patients had higher presentation rates of affective illness, sleep disorder, and poor concentration”. Older people are more likely to have other brain disorders (not CJD) and have lower cognitive reserves. This may explain why they were more likely to present with delirium.¹³

In familial CJD, the age of onset is usually between 30-55 years of age.²⁰ The clinical picture resembles that of sCJD.

Diagnosis

Clinicians should be aware of the symptoms of CJD, although it is rare. Other, more common (and treatable) illnesses must be ruled out first, as the diagnosis of CJD will likely be devastating for the patient and his family. Symptoms suggestive of sporadic CJD include rapidly progressive dementia, cerebellar disturbances, and problems with vision. Myoclonus is also typical for sporadic CJD.⁷

An EEG test will in most sCJD patients show periodic sharp wave complexes. However, these waves can also be present in some other conditions. The presence of a protein called CSF 14-3-3 in the spinal fluid also supports the sCJD diagnosis. An MRI test will help to exclude other diagnoses. A characteristic signal change in the putamen and caudate is suggestive of sCJD.^{7,20}

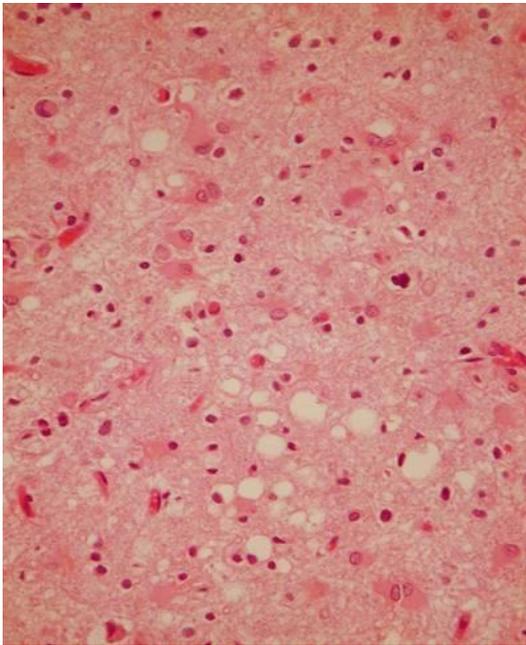
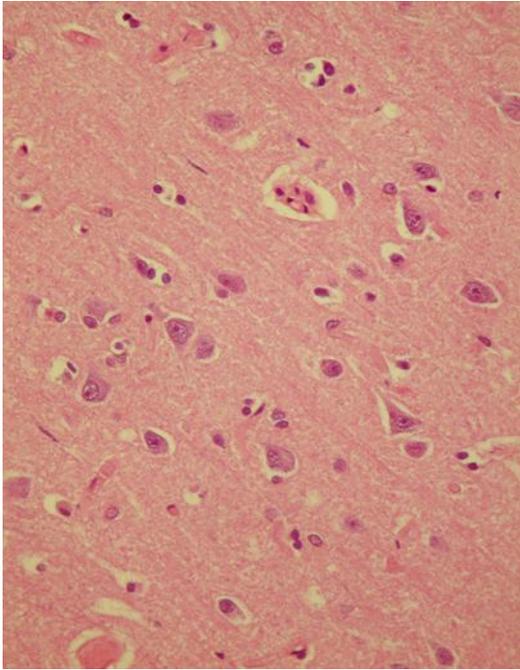
Inherited/familial CJD can be diagnosed by the family history, if known, and by a blood test screening for mutations in PRNP.⁷

Iatrogenic CJD is diagnosed by being aware of relevant risk factors, such as having had brain surgery, having received pituitary growth hormone or human dura mater grafts, or blood transfusions in the UK. EEG and MRI investigations may be similar to those of patients with sporadic CJD.⁷

For variant CJD, residence in UK during the BSE epidemic is a major risk factor. Some of the symptoms of vCJD can be confused with psychiatric disorders. Key to the diagnosis is the observation of ataxia and involuntary movements. There are important differences which distinguish vCJD from sCJD. The EEG does not show the periodic sharp wave complexes in vCJD, and the CSF 14-3-3 test is positive in only about half of the patients.⁷ MRI investigation can show a high signal in the posterior thalamus, known as the “pulvinar sign”.⁹

Important differential diagnoses include Alzheimer disease, dementia with Lewy bodies, and Huntington disease, among others.²⁰

Autopsy is used to confirm the diagnosis of prion disease. Neuropathological changes in the brain include spongiform degeneration, astrocytic gliosis, formation of amyloid plaques and loss of neurons.^{5,16}



The top picture is a normal brain. The bottom picture shows the “holes” in the brain, hence the name spongiform, and the lack of neurons characteristic of the TSEs. Pictures courtesy of Prof. Jan Mæhlen, Ullevål Universitetssykehus.

Treatment

There is at present no curative treatment for CJD; it is universally fatal. However, some treatments have shown promise in animal experiments.¹⁵ CJD is caused by the transformation of normal prion protein PrP^c to PrP^{Sc}. Treatments are based on removing or reducing the amount of PrP^c. Mice have been genetically manipulated so that they stop producing PrP^c after 10 weeks of age. When the gene-manipulated mice were exposed to scrapie, they remained healthy after 52 weeks, whereas control mice died within 12-18 weeks.¹⁵ RNA-interference can also be used to suppress expression of a gene. In a study by White et al, scrapie-infected mice were injected with a lentivirus with double-stranded RNA directly into the hippocampus. These mice lived longer than controls, and their post-mortem examinations revealed fewer degenerative brain changes. A third possibility for therapy is the use of antibodies, either through passive or active immunization.¹⁵ White et al. found that injecting mice with antibodies against PrP^c extended the lifespan of scrapie-infected mice from 200 to 500 days. Theoretically, the binding of the antibody to PrP^c may prevent its transformation to PrP^{Sc}. A medicine that would work against CJD would have to cross the blood-brain barrier or be directly applied into the brain. Pentosan polysulphate has been tested in mice with scrapie and in humans in a small study in the UK. In both the mice and the human studies, it was applied to the ventricles of the brain. The mice who received treatment had a longer incubation time before they showed signs of illness as compared to controls. The human study included only 7 patients, but showed promising results with each of the 7 patients attaining a higher survival time than the average for untreated patients.²¹ Pentosan polysulphate is primarily used to alleviate interstitial cystitis symptoms; its effect on CJD is attributed to binding to PrP^c and preventing polymerization of PrP^{Sc}.¹⁵

In a systematic review of treatments for human prion disease published in 2008, Stewart and colleagues¹² found that most studies of therapeutic interventions for prion disease were poorly designed or reported, and often included very few patients. Only one double-blind, randomized controlled trial was found, and showed that the drug Flupirtine could help to slow cognitive decline, but did not increase survival time. This systematic review was however published before the pentosan polysulphate study mentioned above. Various types of analgesic, anticonvulsant, antidepressant, antifungal, antimalarial, and antiviral drugs have been tested in the treatment of prion disease without effect. Many studies published are case reports of individual patients, thus they carry limited statistical weight.¹²

Prevention of iatrogenic prion disease

More than 400 patients have contracted CJD iatrogenically. CJD is proven to transmit through contaminated neurosurgical instruments, intracerebral encephalographic electrodes, human pituitary hormone, corneal transplant, and dura mater grafts. vCJD is also transmitted through blood transfusion, whereas there is no evidence that sCJD is transmitted via blood transfusion.²² Prions are notoriously difficult to inactivate, and many sterilization methods are ineffective. Important precautions should be taken when dealing with a high-risk patient in a surgical setting. Instruments should not be allowed to become dry after use, as drying makes prions more resistant to inactivation. Disposable instruments should be used whenever possible.²² The WHO recommendation for disinfection of surgical instruments is the use of 1 N NaOH or 20,000 parts per million active chlorine followed by sterilization.⁵⁹ Noncritical environmental surfaces contaminated with high-risk tissues should be cleaned with detergent and then bleach. Standard disinfection procedures should be used to clean environmental surfaces contaminated with low-risk tissues.⁵⁸ There have been theoretical concerns about the risk of

prion contamination of dental instruments, but there have not been any proven cases of CJD transmission through dental procedures. When treating patients with known CJD in a dental setting, it is recommended that dentists discard instruments (such as endodontic^a files) that are difficult to clean, and clean heat-resistant instruments according to WHO guidelines for prion disinfection.²³

A practical, affordable test for presymptomatic vCJD for blood and organ donors will hopefully become available in the future. However, an article by Duncan *et al*²⁴ discusses important ethical considerations in presymptomatic testing. As the disease is incurable, informing asymptomatic blood donors that they are vCJD carriers could be psychologically devastating.²⁴ This is well enough when it involves blood donation, which is something a person volunteers for, and can accept that testing is necessary due to the risk posed to the blood recipient, similarly to reasoning for HIV testing with blood donation. Pre- and post-test counselling would be necessary. However, Duncan argues that it would be beneficial for patients to be tested for vCJD before invasive surgeries with a high risk of disease transmission. This is ethically more difficult, as the patient is undergoing the surgery out of necessity for his own health, not performing an altruistic act as in blood donation. The benefit to the operating team of knowing the vCJD status must be weighed against the potentially devastating consequences of receiving a positive vCJD test for an asymptomatic patient. In this case, patients could choose to not receive the results of a vCJD test.²⁴

vCJD prevention as related to food safety is discussed below under BSE.

Prognosis

The prognosis for all types of prion disease is poor, as there is no cure. The median time from the onset of symptoms to death in sporadic CJD patients is 6 months (ranging from 1 month to 11 years).¹² In familial CJD, the mean duration of illness is also around 6 months.⁵ In vCJD, the disease duration is typically 14 months.⁵ In patients infected iatrogenically, the duration of disease is similar to that of vCJD.⁵ The ultimate cause of death is usually infection or heart or respiratory failure.²⁵

The genetic makeup of patients with vCJD may play a very important role in incubation time.⁵⁶ To date, most patients with symptomatic vCJD have been homozygous for methionine at codon 129 of the PrP gene.⁵⁶ In a study of kuru patients⁵⁶, Collinge et al. found that people with the heterozygous phenotype methionine/valine at codon 129 had incubation times up to 50 years. They postulate that “a human BSE epidemic may be multiphasic, and recent estimates of the size of the vCJD epidemic based on uniform genetic susceptibility could be substantial underestimations”. This theory seems to be supported by a 2009 case study in the *Lancet* describing a 30-year-old British man who developed vCJD in 2008, and, of great interest, was found to be heterozygous for PRNP codon 129.²⁶ It is thus possible that there could be a second wave of vCJD in the future. However, a recent large scale British study in which tonsil specimens from 63000 individuals who would have been exposed to BSE found a prevalence of zero for PrP^{Sc} in tonsillar tissue.²⁷ Thus, the risk for healthcare-associated transmission of vCJD can be regarded as extremely low.²⁷

^a Related to the pulp of the tooth, which also contains lymphatic tissue

B. Genetic prion diseases

The genetic prion diseases include familial CJD (fCJD), Gerstmann-Straussler-Schienenker syndrome (GSS), and familial fatal insomnia (FFI). These diseases are caused by a mutation of the prion protein gene (PRNP) on human chromosome 20.⁷

1. Familial CJD

Approximately 10% of CJD patients have the inherited form of the disease.¹⁵ The symptoms are similar to those of sporadic CJD as described above. The age of onset is between 30 and 55 years, although it can also present very late in life.²⁰ It is diagnosed based on genetic analysis and autopsy findings. The prognosis is similar to that of sCJD, with a mean disease duration period of 6 months.⁵

2. Gerstmann-Straussler-Scheinker Disease

Gerstmann-Straussler-Scheinker disease (GSS) can be caused by various mutations of the PRNP gene, most commonly the proline → leucine mutation at codon 102.²⁰

Occurrence

GSS is an extremely rare disease. It is inherited in an autosomal dominant pattern.⁶

Symptoms and signs

GSS begins around age 50. It begins with cerebellar ataxia and slowly progresses to dementia.⁵ There can also be spasticity, bradykinesia, and increased muscle tone.²⁰

Diagnosis

GSS can be diagnosed by genetic analysis and family history. The presence of multiple amyloid plaques in the brain distinguishes GSS from the other prion diseases.²⁰

Prognosis

There is no treatment for GSS. However, the disease progresses more slowly than the other prion diseases, with mean survival of 5-6 years from the time of the onset of symptoms.⁵

3. Familial fatal insomnia



Man with insomnia, photo credit emedicinehealth.

Familial fatal insomnia (FFI) is a genetic prion disease linked to a mis-sense mutation at codon 178 of the PRNP gene.²⁸ It follows an autosomal dominant pattern of inheritance.²⁹ Aspartic acid is replaced by asparagine at codon 178.²⁹ The same mutation can also be found in families with fCJD, but the CJD families will have valine expressed at codon 129 of the mutated gene, whereas the FFI sufferers have methionine at codon 129 of the mutated gene.²⁸ “Non-familial” FFI also exists in a sporadic form which is clinically similar.²⁸

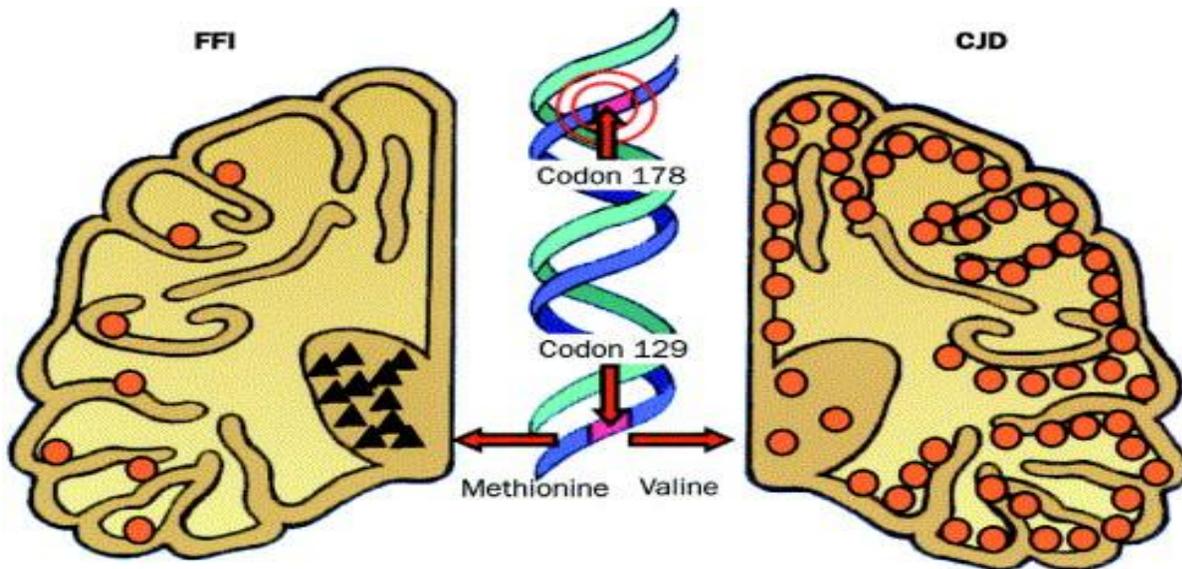


Figure from article by Montagna et al.²⁸, showing importance of codon 129 for disease phenotype

Occurrence and transmission

FFI is a rare disease, with about 27 families known to have the disease in the world. It can be transmitted to animals.²⁸

Symptoms and signs

FFI typically begins around age 50, although the age of onset ranges from 20-62 years.²⁹ To begin with, the symptoms of the disease can be confused with stress or fatigue, as patients complain of difficulty sleeping. They may be very drowsy in the daytime. Double vision, diplopia, is common. Patients often experience hallucinations and appear to be in a dream-like state that can be confused with psychosis. Symptoms of sympathetic overactivation, such as tachycardia, hypertension, perspiration, and pyrexia occur.²⁹ Later, patients develop gait disturbances, myoclonus, dysarthria, dysphagia, and sphincter incontinence.²⁹ Interestingly, general intelligence is preserved in FFI, although patients do become progressively more confused.²⁸ At the end of life, they spend more and more time in a dream-like state.^{28,29}

Diagnosis

The disease is diagnosed on the basis of genetic analysis and family history. Sleep histograms show loss of deep-sleep and loss of slow-wave EEG activity. Another key finding is that body temperature, blood pressure, and heart rate are higher at rest than while awake.²⁸ Circadian rhythms are severely disrupted, including the secretions of various hormones important in regulating autonomic functioning.²⁸

Treatment

The only treatment for FFI today is merely palliative. Scientists have examined the use of various drugs to induce sleep, including benzodiazepines, sedatives, and GHB.²⁹ None of these has provided patients with a cure.

Prognosis

Patients typically live a mean of 18.4 months from the onset of insomnia until the time of death. FFI patients' children will have a 50% chance of inheriting the disease. Patients with the Met-Met genotype have a shorter survival time than patients with the Met-Val genotype.²⁸

C. Kuru

Background

Kuru is a prion disease caused by endocannibalism practices in Papua New Guinea. It has only been found among the Fore people and their close neighbors in Papua New Guinea.⁶⁰ Kuru was considered an epidemic, with more than 2700 cases from 1957-2004.⁵⁶ Women were more exposed than men, resulting in the “motherless nuclear family” becoming common among the Fore people.³⁰ Deaths among young women were so many that it became common practice to withhold the “bride price” until the bride had lived long enough to produce a child.³⁰ The Fore people believed the disease was caused by sorcerers.³⁰

Occurrence and transmission

Anthropologists Robert and Shirley Glasse were the first to suggest that cannibalism might be the route of transmission of the disease.³⁰ The Fore people likely began the practice of ritual consumption of dead relatives sometime in the late 19th or early 20th century. According to the oral history of the Fore people, people first started dying of kuru sometime between the 1920s and 1940s. All body parts except the gallbladder were consumed, and victims of kuru were considered favourable for consumption.³⁰ Adult women and small children of both sexes participated in the feasts, while boys over the age of approximately 6-8 years and adult men did not participate.⁵⁶ After the practice of cannibalism was banned by the Australian government in the late 1950s, the incidence of disease in young children dropped.⁶⁰ Gajdusek and colleagues proved that kuru was a transmissible disease when they succeeded in inoculating the chimpanzee Georgette with brain material from a human patient.³¹

The incubation period of kuru has a huge range, from 5 to possibly more than 50 years.⁵⁶ There have been no cases of Kuru in patients born after 1959.⁵⁶

Symptoms

Kuru often begins with a prodromal phase of headaches and pain.⁶⁰ This is followed by cerebellar ataxia, which progresses until the person is immobilized.⁶⁰ The cognitive processes can be fairly well-preserved.⁵⁶ Chronic emotional instability is common³², kuru has also been called “the laughing sickness”.³³ There are three stages in the progression of the disease: the ambulant, sedentary, and terminal stages.⁶⁰



The boy on the left, sick with kuru, is unable to stand without assistance. Courtesy of Prof. Jan Mæhlen, UUS.

Diagnosis

Kuru is a very distinctive disease and was usually easily recognized by the patients and their families.⁵⁶ Monitoring of the disease is performed by the Papua New Guinea Institute of Medical Research.⁵⁶ Histopathological changes on autopsy are most noticeable in the cerebellum, but are also found to a great extent in other parts of the brain and spinal cord.⁶⁰ Spongiform change, neuronal loss, and gliosis are typical.³²

Prognosis

Patients live an average of 12 months from the onset of symptoms until the time of death.⁶⁰ Collinge *et al.* have suggested that heterozygosity (Methionine-Valine) for the PRNP 129 genotype gives less susceptibility to kuru and a longer incubation time, ranging from 34-56 years, in those infected.⁵⁶ The ban (and enforcement of it) on cannibalism from the mid-1950s has given a unique opportunity to study the incubation period, as one to two people continued to develop kuru each year up until 2004.⁵⁶ Hopefully, the disease has now been eradicated and will merely remain a fascinating part of medical history.

IV. Animal prion diseases



Healthy cattle, picture from Annual Reports 2008 of Veterinærinstituttet



Downer cow, unable to stand, from Wikipedia.

A. BSE

Bovine spongiform encephalopathy (BSE) is a TSE which affects cattle.³⁴ BSE causes various clinical signs including changes in mental status, posture, and movement, and reduced milk yield.⁵⁷ Examination of the brain in sick cattle shows vacuolization and scrapie associated fibrils.⁵⁷

The first BSE case was observed in 1985 in the UK (Ducrot).⁵⁷ There have been over 180,000 cases of BSE in the UK, as well as a number of cases in the rest of Europe, Japan, Israel, Canada, and the USA.³⁴ The use of meat and bone meal (MBM) in cattle feed has been strongly implicated in the outbreak of the BSE epidemic. Changes in the early 1980s in the way MBM was produced are thought to have allowed the sheep scrapie prion protein to survive the cooking

process.¹⁶ The use of ruminant^b MBM for ruminant feed was forbidden in 1988 in the UK, and the use of specified risk materials/specified bovine offals^c was banned in 1990.⁵⁷ The use of MBM was forbidden in Norway in 1990.³⁴ The use of MBM in the production of feed for ruminants has been forbidden in the EU since 1994, and laws requiring the use of high temperature and pressure when producing MBM were passed.³⁴ EU regulations to prevent TSEs now include: surveillance of TSEs in animals, particularly cattle, sheep, and goats; removal and destruction of organs that are particularly infectious (specified risk materials); better slaughtering techniques in cattle to avoid contamination with brain matter; limited use of MBM; and regulations on import and export of animals or products that could be infectious.³⁴ The United States passed laws in 1997 and 2008, first prohibiting the use of mammalian protein in feed for ruminant animals, and subsequently prohibiting the use of brain and spinal cord tissues from cattle over 30 months of age in animal feed.³⁵

In Norway, a passive surveillance program for BSE was in place from 1990 through 2000, and active surveillance from 2000 onwards. Starting in 2001, an ELISA test to detect PrP^{Sc} was made available.³⁶ In the Norwegian program, clinically suspicious cattle, downer cattle over age 24 months, emergency slaughtered cattle over age 24 months, cattle over 24 months with abnormal ante-mortem findings, slaughtered cattle with unknown age or origin, and slaughtered imported cattle from any country are tested. In addition, 10,000 randomly selected healthy slaughtered cattle over age 30 months are examined yearly for BSE.³⁶ Norway is compliant with all EU/EEA regulations and in addition has some regulations that are even more stringent.³⁴ It is reasonable to conclude that there is negligible risk involved in the consumption of beef in Norway.

Although there are some vocal dissidents, it is now generally agreed that BSE is transmitted to people through the oral route and causes variant CJD. In a 1996 *Lancet* article, Will *et al.* described 10 new cases of a new variant of CJD, all affecting younger people.¹⁴ They suggested that these patients, whose illnesses began earliest in 1994, had become ill due to the consumption of contaminated beef. This was consistent with an incubation period of 5-10 years, with the mid-late 1980s likely being the peak of the BSE exposure to the UK population. SBO were excluded in human food after November 1989, so the exposure probably was much less after this point. The British government had maintained that beef was safe to eat, despite the BSE outbreak. In a memorable moment in 1990, Agriculture Minister John Gummer was photographed together with his daughter eating hamburgers in an attempt to reassure the public of the safety of British beef.³⁷ These reassurances now rang false and consumers panicked.



^b Ruminant: An animal that chews the cud

^c Specified bovine offals: the term used in the UK to denote tissues that can be infected with the agent of bovine spongiform encephalopathy (BSE), namely brain and spinal cord, spinal ganglia, retina, and terminal small intestine

John Gummer and 4 year-old Cordelia. BBC news.

The BSE epidemic and ensuing vCJD scare had a drastic impact on the British cattle industry. At the peak of the epidemic, in 1992, approximately 1% of British cattle were infected with BSE. However, it was not until 1996, when scientists discovered the link between vCJD and BSE, that the industry really suffered. Cattle prices fell by 25%, consumer consumption of beef fell 26%, and the export market for beef was virtually eliminated.³⁸ The EU banned internal trading in UK beef, and the ban was not lifted until 1999. In the year following the outbreak of the BSE/vCJD epidemic, the total estimated economic losses due to BSE in the UK were estimated to be \$980 million.³⁹

Is it safe to eat beef? Countries have varying levels of surveillance for BSE and varying regulations regarding which tissues are deemed SRM. Importantly, many countries have never even had documented cases of BSE. There is no test to identify SRM on a beef carcass.⁴⁰ Even with the removal of specified risk materials, conventional slaughtering practices can still potentially allow contamination of meat with brain material. The use of captive bolt stunners, which damage the brain while the cow's heart is still beating, can potentially allow infectious brain material to enter the blood. Pneumatic captive bolt stunning, in which air is injected into the brain, has been banned in the U.S. and many other countries.⁴⁰ Other risky practices include the insertion of a rod through the stunner to destroy the brain stem and spinal cord, which prevents leg kicking and benefits the safety of abattoir workers^d. In Norway, the use of captive bolt stunners has been modified, so that the bolt does not penetrate to the brain. The safest methods of stunning appear to be either penetrative captive bolt stunning without pithing or nonpenetrative captive bolt stunning.⁴⁰ The floor, equipment, and abattoir^e personnel can also potentially be contaminated with CNS tissue; it is particularly of concern that workers could carry CNS tissue with them outside of the facilities and contaminate other areas⁴⁰. Other problematic areas in conventional cattle slaughter are the removal of the head, typically done with a knife, and the splitting of the carcass along the spine⁴¹. Carcass splitting represents a risk both for dissemination of CNS tissues to the carcass and for contamination of the saw and subsequent carcasses. Bowling *et al.* have performed multiple studies showing that washing done on the saw in between carcasses is often not sufficient to remove all CNS tissue⁴⁰. Alternative methods for removing meat products from the carcass without splitting have been researched, but the benefits must be weighed against additional costs and inconvenience.

B. Scrapie

Scrapie was the first TSE known to man. Some authors claim scrapie reports have dated as early as the 15th century. The first description of scrapie may have been as early as 1732, but this publication is not found. However, a 1772 publication refers to scrapie being reported over the last 40 years, which would be consistent with the first paper in 1732. One of the first texts on scrapie, written in the year 1750, translated from German, describes the symptoms as follows: "...scrapie, an illness recognizable in the recumbency of the animal. It nibbles at its claws and feet and scratches its back against posts. It ceases to prosper, loses appetite and finally gets weary. They drag themselves for a long time, are consumed more and more, and in the end, they are doomed to death". This early description of the symptoms remains accurate today.⁴² Other symptoms of scrapie are apprehensiveness or uneasiness during handling or feeding, tremors of

^d This practice is known as "pithing".

^e slaughterhouse

the head or neck, and isolation from the flock.⁴³ In the past, scrapie has been known by various descriptive names: cuddie trot, la maladie trotteurs, prurigo lumbar, rickets, rubbing disease, scratchie, shakings, shrugginess, tempermanner, and trotting disease.⁴² The incubation time is from 2-5 years, and the survival time after the onset of symptoms is from 1-6 months.⁴³

Even in olden times, farmers had an understanding that scrapie was infectious, and in the 19th century it was commonly believed that scrapie was spread by sexual intercourse.⁴² Others believed scrapie was a spontaneous disease, and some believed it to be a hereditary disease, since lambs of infected ewes often contracted the disease.⁴² Cuille and Chelle demonstrated in 1936 that scrapie could be transmitted to sheep and goats via inoculation with brain and spinal cord material. In another interesting experiment in the 1930s, J.R. Greig showed that a healthy flock of sheep could be infected with scrapie by grazing in the same field that a sick flock of sheep had grazed on.⁴² We now know that prions can effectively bind to soil and remain infectious for years.⁴⁴ One study showed that a scrapie-infected sheephouse and pasture remained infectious for 16 years, despite having been decontaminated. Scrapie prions are shed in urine, feces, saliva, blood, and birthing matter. There is at present no effective and practical method for detecting or measuring the presence of prions in the environment. Disposal of infected carcasses may be an environmental problem; in the BSE outbreak in the UK 6000 cattle carcasses were put in landfills, possibly posing a risk of infection.⁴⁴ The most effective method of disposal of prion-contaminated material is incineration at 1000 degrees Celsius, however, this is not always practically possible, as it is inconvenient to incinerate large amounts of contaminated soil, vegetation, fences, paddocks, etc.⁴⁴

An unusual type of scrapie was discovered in Norwegian sheep in 1998. The genotypes of the affected sheep are consistent with resistance to the classical form of scrapie. The distinguishing clinical feature of the Nor98 scrapie is ataxia.⁴⁵ The sheep also experience anxiety and loss of body condition. They do not generally seem to experience the pruritus associated with classical scrapie. The brains also have a different appearance and distribution of the PrP^{Sc} protein than in classical scrapie. In classical scrapie, the dorsal motor nucleus of the vagus is the most affected structure. This structure is not affected in Nor98 scrapie. Sheep with Nor98 scrapie tend to be older. It is possible that atypical/Nor98 scrapie is a sporadic, non-infectious disease, similar to sporadic CJD. It often occurs as single cases within a flock, whereas classical scrapie tends to affect multiple sheep within a flock.⁴⁵

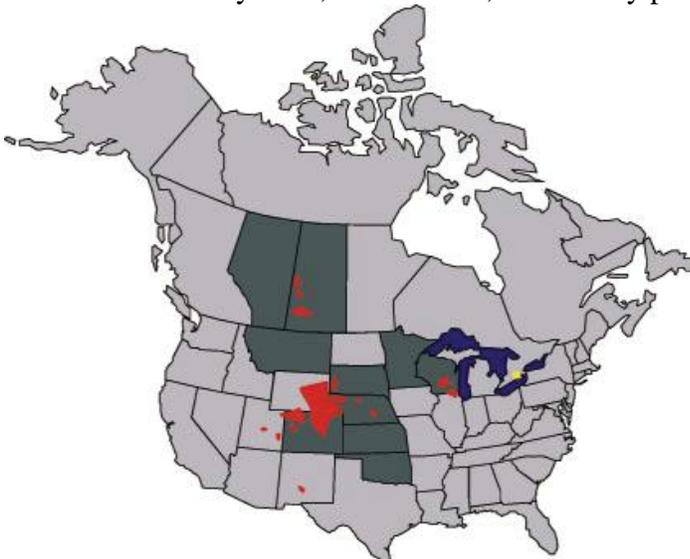
Classical scrapie is found all over the world with the exception of Australia and New Zealand. Goats can also become infected. Nor98 scrapie has been found in most countries in western Europe.⁴⁶ The Norwegian surveillance program for scrapie includes examination of all animals with clinical signs of scrapie, examination of slaughtered fallen stock over 18 months, examination of randomly selected slaughtered healthy sheep over 18 months, and examination of goats which have died or been killed on the farm⁴⁷. Today, as in the past, the only remedy for farmers with scrapie-infected sheep is to cull all affected animals.⁴² Genetic selection for scrapie-resistant sheep has also been explored as an alternative.

C. Chronic Wasting Disease



Emaciated animal with CWD, photo credit Dr. Elizabeth Williams, University of Wyoming

Chronic wasting disease (CWD) is a TSE affecting mule deer, white-tailed deer, Rocky Mountain elk, and Shira's moose. It is found in North America and South Korea. CWD is notably different from BSE and scrapie in that it occurs in free-ranging animals as well as in domestic herds. Pathologists Williams and Young were the first to describe CWD in captive mule deer in a 1980 publication⁴⁸. It is thought that CWD originated in mule deer and subsequently spread to elk and white-tailed deer.⁵⁵ While moose can be infected with CWD, they tend to live solitary lives, not in herds, which may protect them somewhat from infection.⁴⁸



Historic distribution of CWD from 1977-2004. Dark gray areas for CWD in farmed cervids, red for CWD in free-ranging cervids. From article by CJ Sigurdson.⁴⁸

CWD is likely horizontally transmitted among cervid^f animals. CWD can be spread through direct contact among animals, but the mechanism is unknown.⁵⁵ Prions can bind to soil particles and remain infectious for long periods of time.⁵⁵ Environmental contamination is thus a risk for CWD. Prion infectivity in CWD is found in the CNS, pancreas, adrenal gland, blood, and skeletal muscle.⁴⁸ Sheep and cattle grazing on areas shared with CWD-infected cervids may be exposed to CWD through the oral route, but there appears to be a strong species barrier. Even when cattle are exposed to CWD by intracerebral inoculation, less than half develop the disease. Hunters and others who consume venison may also be exposed to CWD, but there has been no epidemic of human TSE infection in the geographic areas endemic for CWD.⁴⁸ State authorities advise hunters to avoid harvesting cervids that appear ill and to take precautions when processing the meat.⁵⁵

How can farmers, hunters, gamekeepers and veterinarians recognize CWD? E.S. Williams described the manifestations of CWD in detail, noting that animals infected with CWD can exhibit symptoms resembling many other diseases, including hemorrhagic disease, meningeal worm infection, and locoweed intoxication.⁵⁵ Progressive CWD disease causes weight loss and behavioral changes, such as isolation from the herd, over weeks or months. Animals may salivate excessively due to difficulty in swallowing. They may also exhibit head tremors, ataxia, and an altered stance.⁵⁵ Changes in weight and body condition can be misinterpreted as illness, when they in fact represent seasonal variation in the animal's body mass. From the time of onset of clinical disease, death usually occurs within 4 months. Free-ranging cervids are more likely to have a shorter survival time than captive cervids, due to a reduced ability to find food and water and reduced capacity to avoid predators and vehicles.⁵⁵

Diagnosis of CWD is done by immunohistochemistry, which is considered to be the gold standard. Samples are taken from the animal's retropharyngeal lymph nodes or obex and are analyzed at U.S.D.A. approved laboratories.⁴⁸ Histopathological examination of brains of animals with CWD reveals bilateral, symmetrical spongiform degeneration and amyloid plaques.⁵⁵

What is being done about CWD? CWD affects farmed and wild cervids and has a negative economic impact on the hunting industry and on sales of farmed cervids and cervid products.⁴⁹ State and provincial agencies in the U.S. and Canada are working to identify sick animals, quarantine and depopulate herds with sick animals, and to provide compensation to affected farmers. There are also programs in place to certify healthy herds, in which there has been no CWD after 5 years of monitoring. Monitoring CWD in wild cervids is a greater challenge. Efforts include surveillance, guidelines for hunters, and reduction of the cervid population.⁵⁵ It is unlikely that complete eradication of CWD in free-ranging cervids will be possible, but hopefully control within domestic herds can be achieved.

D. Transmissible Mink Encephalopathy

Transmissible mink encephalopathy (TME) is a rare TSE affecting ranched mink. The first cases were reported in 1947 on a Wisconsin farm. The disease has since been documented in other U.S. states, Canada, East Germany, Finland, and the former Soviet Union.⁵⁰ The last outbreak was in Wisconsin in 1985.⁵¹

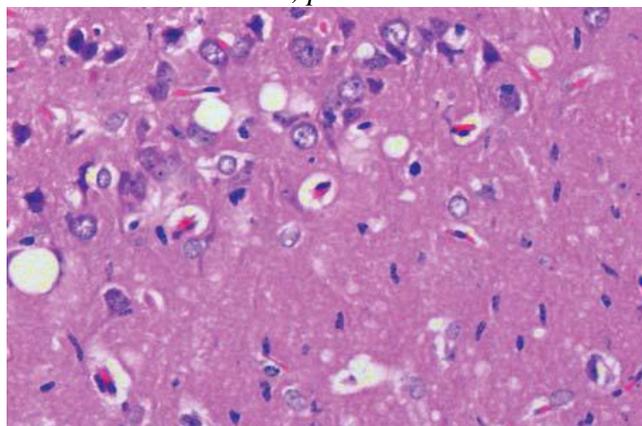
^f Hoofed mammals of the family Cervidae

TME is thought to be acquired orally when mink eat feed contaminated with prions, possibly from a rare strain of BSE prion called L-type. In the first outbreak of TME in the U.S., all the affected farms had been using feed from the same producer, which strongly points to an oral route of transmission.⁵¹ Wisconsin, where most of the U.S. TME cases have been, is a dairy state, and aged cattle are frequently used in mink feed.⁵¹ In one case, a rancher made his own feed, composed of commercial feed and meat from sick or downer cattle.⁵¹ TME can also be experimentally transmitted from mink to cattle.⁵¹ When it occurs, TME tends to affect most or all adult animals within a ranch, but kits^g seem to be spared, even when they are nursing from infected dams^h.⁵⁰ Mink-to-mink transmission is possible, but unlikely, as there is usually only one mink per cage and the minks have no direct contact with each other.⁵⁰ The incubation period of TME is 7-12 months.⁵²

The symptoms of TME include changes in normal grooming behavior, soiling of the nest, difficulty swallowing and feeding, and arching of the tail over the back. Eventually, affected minks become uncoordinated, with jerky motions. The minks ultimately become somnolent and unresponsive before death occurs. Survival time from the onset of symptoms is only 2-6 weeks.^{51,52} Histopathological examination reveals spongiform change, astrocytic gliosis, and amyloid plaques.⁵¹ Western blotting or immunohistochemistry can detect accumulation of prions in the CNS.⁵⁰



Sick mink with TME, photo credit PP Liberski



Spongiform change in TME, photo credit PP Liberski

^g Baby mink

^h Females

Ranch owners are now aware of the danger of feeding infected meat from sheep or cattle to minks, and the last outbreak of TME in the U.S. was in 1985.⁵² Hopefully, there will be no further TME infections in the future other than those induced in scientific laboratories.

E. BSE related diseases

The BSE-related diseases include feline spongiform encephalopathy of zoological and domestic cats (FSE) and TSE in zoological ruminants and non-human primates.⁵² Feline spongiform encephalopathy was first reported in the U.K. in 1990.⁵³ Similar cases in domestic cats were reported in Northern Ireland, Norway, and Liechtenstein. The infected cats, all of whom were over 2 years of age, exhibited changes in behaviour, ataxia, hypermetria, and tactile and auditory hyperesthesia.⁵² The number of new infections dropped significantly following the U.K. ban on the use of specified bovine offals in 1990, and there has only been one FSE case in a cat born after the BSO ban. All of the non-domestic cats with FSE lived in zoos in Great Britain⁵²; it is assumed that the consumption of BSE-infected cattle carcasses was the mode of transmission. The geographical pattern of FSE, with nearly all of the cases located in Great Britain, as well as the rapid decline in new infections following the SBO ban, strongly points to a close association with the BSE epidemic. Mice inoculated with brain material from FSE-infected cats have the same incubation period and histological appearance as mice inoculated with BSE⁵⁴.

A variety of other exotic zoo animals residing in the U.K., all in the family Bovidae¹, were also infected with TSE. Most of the animals had been fed meat and bone meal.⁵³ A spongiform encephalopathy has also been described in non-human primates in France in the 1990s; it is likely that these animals consumed feed contaminated by infected British beef⁵².

V. Discussion

Prion diseases are progressive, fatal, neurodegenerative diseases caused by the accumulation of misfolded prion proteins in the brain.⁶ Prion disease affects animals and people, and can be transmitted between species.⁵ They can occur both in wild and in domesticated animals.⁵² Infection can result from the consumption of contaminated feed, from cannibalism, from exposure to prions in soil, or from medical procedures.^{7,42,55,56,57} Some forms of prion disease are genetic, and follow an autosomal dominant pattern.²⁰ Genetics play an important role in prion disease, with certain genotypes being more resistant to infection and having longer incubation times if infection does occur.⁵⁶ The symptoms and presentation of prion disease vary greatly; prion diseases can present as dementia, psychosis, sleep disturbances, or behavioural changes.¹³ The incubation times are generally long, but the duration of illness, once it is apparent, is short. Sadly, the treatments available for human prion diseases today can only be described as experimental or palliative at best.¹⁵ Even in case histories presented as “success stories”, patients usually have achieved just a few months more survival time than expected without treatment.¹² Finding a treatment for prion diseases is a great challenge. Since prion diseases are very rare, there are practical difficulties in recruiting enough patients to conduct a statistically sound study. Diagnosis can also be difficult, and some patients may not receive an accurate diagnosis until the time of autopsy.⁷ Additionally, although we know that the aberrant prion protein causes disease, we still do not even understand the function of the normal prion protein PrP^c.⁵

¹ Bovidae: cloven-hoofed mammals, including, but not limited to, bison, oryx, nyala, kudu, and eland.

Prevention of iatrogenic CJD is a medical issue of great importance. The risk of contracting CJD from a medical or dental procedure is extremely low, especially in countries with a low prevalence of vCJD, but the consequences of iatrogenic transmission are devastating for the unfortunate patients affected.²⁴ The risk can be reduced by using disposable instruments and proper cleaning of re-usable instruments.⁵⁸ Although it would be ideal to disinfect all surgical instruments so that one would be certain that all prions are removed, this is time-consuming and would cause significant wear on the instruments.⁵⁹ It is conceivable that doctors may test patients who are undergoing brain surgery in the future for CJD, once such a test becomes available.²⁴

Scientists and government officials have succeeded in eradicating some prion diseases, notably kuru⁶⁰ (which was caused by cannibalism) and transmissible mink encephalopathy⁵¹. The BSE epidemic remained largely contained to Great Britain due to strict sanctions placed on export of British cattle products and restrictions on the use of specified risk materials in food for animals and people.⁵⁷ More stringent regulations for slaughtering of cattle also have reduced the risk of contamination of meat with brain tissue, but this risk cannot be regarded as completely eliminated.⁴⁰ Researchers continue to investigate alternative, less risky, methods of slaughtering cattle but, especially for countries with a low prevalence of BSE, the benefits may not justify higher costs and time expenditure. BSE is particularly worrisome as it is shown to transmit to humans¹⁴, yet other animal prion diseases, such as scrapie and CWD, have not been shown to transmit to humans, despite both oral exposure and exposure to blood during slaughtering.^{61,55}

For animals infected with TSEs, the only “treatment” available in the foreseeable future will continue to be slaughter. The economic effects of TSEs in animals can be disastrous, as we saw during the BSE epidemic in Britain.³⁸ The threat of economic ruin will hopefully keep governments, farmers, and slaughterhouses motivated to strictly follow and improve guidelines for acceptable animal feed products and slaughtering methods.

Many mysteries remain in the rare but fascinating world of prion diseases. Even diseases we might think we know well, such as scrapie, continue to surprise and present themselves in new forms, as in the recent outbreak of Scrapie Nor98.⁴⁵ Genetics play an important role in prion diseases, and gene therapy may eventually be a focal point of treatment.¹⁵ In the coming years, it will be particularly interesting to see whether or not there will be a second wave of vCJD infection in the UK among patients who are heterozygous for PRNP codon 129. Researchers will aim to develop methods of effective prion disease detection in humans²⁴ and search for effective treatments. The function of the healthy prion protein, the nature of the infectious agent PrP^{Sc}, the manner in which peripherally administered prions travel to and invade the brain, and the mechanism behind the resulting spongiform degeneration are all topics which are not fully understood and will undoubtedly be the subject of further studies.⁶²

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