Leading article

Prion diseases

A major challenge for future research

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Prion diseases are rare forms of fatal and transmissible neurodegenerative conditions characterised by accumulation of protease-resistant forms of the prion protein (PrP), termed PrPres or PrP^{sc}, in the brain as insoluble aggregates of amyloid fibrils. In the recent past this group of diseases came to scientific, political and media attention because of a possible association of bovine spongiform encephalopathy to new variant Creutzfeldt-Jakob disease. Five human and six animal prion diseases have been described.This article focuses on the human prion diseases, and some animal prion diseases which are relevant to human disease.

Scrapie was the first prion disease to be described and as a clinical entity, was recognised in sheep in England as early as 1730. The first clue to its pathogenesis came in 1934 when Icelandic farmers found that scrapie could be transmitted by inoculation (1). About 20 phenotypically different strains of scrapie have been identified experimentally. It can affect sheep, goats and other animals. Diseased animals develop aggressive behaviour, restlessness, itching, tremor and ataxia, and death within 4 months (2).

Kuru, which was confined to Papua New Guinea, was first described in 1955. After extensive studies in 1960s and early 1970s Carleton Gajdusek demonstrated that Kuru was transmitted by ritualistic cannibalism practised by certain tribes (3), for which he was awarded the Nobel prize for medicine and physiology in 1976. The women and children were mostly affected, since they were commonly exposed to contaminated brain tissue during these rituals. The term 'Kuru' means 'shivering' or 'trembling' in the dialect of the Fore. The affected patients develop tremulousness together with other cerebellar symptoms such as dysarthria and ataxia. Emotional lability is so prominent that the disease is sometimes called 'laughing death'. Dementia is conspicuously absent. The disease progresses with multisystem involvement and death within 3 to 9 months. The illness gradually disappeared after the cessation of cannibalistic rituals in the region (2).

Creutzfeldt-Jakob disease (CJD) is the best known and commonest of the human prion diseases and occurs worldwide with an annual incidence of 0.5 to 1.5 cases per million, with no apparent geographical clustering (2). Although the majority of CJD cases occur sporadically, it can be transmitted genetically and iatrogenically. The genetic transmission is linked with the PrP gene, which resides within a single exon and encodes the product of 253 amino acids. To date 14 point mutations and 8 different length octa repeat insertions are known to be associated with genetic disease, which accounts for about 10 to 15% of all cases. The transmission is autosomal dominant.

The first evidence of iatrogenic transmission was found in 1974, when a cornea transplanted patient developed signs of CJD. Both the recipient and the donor had typical neuropathological features of CJD at autopsy. Later, there were more case reports of iatrogenic transmission of CJD following neurosurgery, stereotactic EEG electrode insertions, cadaveric dural homografts and human growth hormone and gonadotrophin injections (1).

According to the mode of transmission the clinical features vary slightly, but the main ones remain the same. The classical triad of CJD comprises rapidly progressive dementia, myoclonus and a characteristic EEG with 1 Hz regular periodic complexes. CJD can also present with cognitive decline, ataxia, visual disturbances, behavioural disturbances and a rapidly evolving neurological deficit resembling a stroke. Dementia is invariable at some stage of the illness and myoclonus is present in 80%. Eventually patients become bed-ridden, mute and unresponsive, and die in about 8 months (2).

Familial fatal insomnia (FFI) is due to a mutation in the gene encoding for prion protein. There are 9 families identified in the world carrying this mutation. FFI patients develop insomnia, dysautonomia and changes in circadian rhythms of hormone secretion. The duration of illness is 7 to 18 months. Gerstmann-Sraussler-Scheinker disease (GSS) is an autosomal dominant disease leading to progressive cerebellar ataxia, dementia and death. It is also associated with mutations of the PrP gene. There are about 50 families identified worldwide carrying this mutation.

Bovine spongiform encephalopathy (BSE)

BSE was first reported in Britain in 1986 and reached epidemic proportions in 1992, with a total number exceeding 170 000 cases (4). The cattle in Britain were fed with a preparation popularly known as meat and bone meal (MBM), made from the remains of slaughtered animals after rendering. As a cost reduction measure the strin-

gency of the rendering procedure was changed during 1970s and early 1980s, when reduced amounts of hydrocarbon solvents were used and processing temperatures were reduced. This may have resulted in survival and recycling of the infectious agent deriving from scrapie, resulting in the epidemic. A rapid decline of the epidemic after imposing a ruminant feed ban in 1988 was consistent with this hypothesis. The clinical course of BSE is typically about two months. The most commonly observed signs are apprehension, hyperaesthesia, ataxia, decreased milk yield and loss of condition (3).

New variant Creutzfeldt-Jakob disease (nvCJD)

In 1990 surveillance of CJD was recommenced in the UK from concern that the agent responsible for the epidemic of BSE might transmit to humans. The rare occurrence of CJD in two teenagers in 1995 was followed in the next few months by the identification of 8 others. All 10 patients shared a distinct and previously unseen clinicopathological phenotype, that was named new variant Creutzfeldt-Jakob disease. The striking early features of nvCJD are sensory and psychiatric disturbances, which are unusual in sporadic CJD. The sensory symptoms included foot pain, cold feet, hemi-dysaesthesia and paraesthesiae. Most patients consulted psychiatrists for apathy, fleeting delusions and psychosis (1). Later they developed frank neurological features such as ataxia, global cognitive impairment, involuntary movements and urinary incontinence, with death within one year. There is a mass of experimental data derived from biochemical, biophysical and genetic studies indicating that infective agents in BSE and nvCJD are identical (5).

Pathogenesis

In many aspects prion diseases' biology is unorthodox. Perhaps the most fundamental paradox is posed by the co-existence of inherited, sporadic and infective forms of the disease (6). In 1967 a radical theory was put forward suggesting that the infective agent could be a self-replicating protein (7). Subsequent experiments showed that scrapie infectivity was associated with a protease resistant protein, and in 1982 the term prion was introduced for the hypothetical proteinaceous infectious particle. In 1984 the PrP gene was identified (termed PRNP), and subsequently it was shown that if we delete the PrP gene from mice (PrP knocked-out mice) they are not susceptible to prion diseases. Prion protein is now known to be a normal outer cellular membrane glycoprotein expressed in most cell types, especially in nervous tissue. Its exact function is unknown, but it appears to be necessary for long term survival of Purkinje neurons, regulation of circadian rhythm and for normal synaptic function. Prion disease is believed to occur when the normal protease sensitive cellular form (PrP^{c}) is transformed into an abnormal protease resistant isoform (PrP^{sc}). The differences between the two forms are solely conformational, with PrP^{sc} possessing a high beta-sheet content (8). The abnormal isoform PrP^{sc}, once produced, acts as a template for conversion of PrP^{c} to PrP^{sc}, setting a chain reaction in motion for producing more PrP^{sc} (9). The mechanism whereby the PrP^{sc} causes tissue damage is still unclear, and may partly be due to unavailability of the normal isoform. It has been suggested that in the hereditary forms, mutated sequence of PrP fragments may have increased neurotoxic and altered fibrillogenic activity, with a tendency to fold into pathological PrP^{sc}. In sporadic disease the initial PrP needed to seed the production of PrP^{sc} occurs as a rare spontaneous event, perhaps due to a somatic mutation of PrP gene in one or more cells. In infective disease the inoculated PrP^{sc} initiates a chain reaction of PrP^{c} conversion to PrP^{sc} in the host (10).

There are several other factors that determine susceptibility. Species barrier plays a major role. There is greater resistance to transferring the infection across species than within the same species. Evidence also suggests that structure homology between the donor and the host, especially in the middle third of the PrP molecule, is critical (10). Human PrP gene too plays a central role in conferring susceptibility through methionine-valine polymorphism at codon 129. It is interesting that all cases tested so far with nvCJD have methionine-methionine genotype at codon 129. Recent studies suggest that, unlike humans, bovines only code for methionine at an equivalent site to codon 129, which may explain why M-M genotype is susceptible to nvCJD. Stanley Prusiner received the Nobel prize for medicine and physiology for his pioneering research and proposing the prion theory. If this is confirmed prions become unique infectious pathogens, devoid of nucleic acid (11).

Although the prion theory gained increasing popularity over the past 10 to 15 years there are researchers who believe that the transmissible agent is an "unconventional virus". Others believe that the agent is a virino – a small informational molecule associated with a host protein. The only feature to support these hypotheses is the existence of several strains, especially in scrapie, where as many as 20 strains have been identified. Recent evidence suggests that PrP can retain strain information by adopting different conformational states without the presence of DNA (12).

Pathology

The unique feature in prion pathology is the absence of specific immune response. Indeed the infectious agent is essentially composed of a protein with a primary structure identical to host protein, making the immune system naturally tolerant. Lymphoid organs are strongly implicated in the early peripheral steps of the disease. Paradoxically,

too, immunodeficient animals, more susceptible to other infections, appear to be partially or completely resistant to experimental infection.

The microscopic hallmarks of prion diseases are spongiform change, neuronal loss, reactive astrocytosis and accumulation of a disease associated isoform of PrP in the nervous system (13). The PrP accumulates as large fibrillary plaques and small clusters best visualised by immunocytochemistry. The pattern of pathological changes varies. In CJD changes are predominant in cerebral cortex, in kuru in the cerebellum and in nvCJD in both cerebral and cerebellar cortex. In FFI the changes are predominant in the thalamus.

Risk of transmission and safety precautions

Despite many intimidating reports in the media it is important to note that prion diseases are not contagious in the usual sense. To date there have been no proven instances of CJD contacted occupationally. Precautions should be taken against iatrogenic transmission (12). There is experimentally proven potential yet minimal risk of transmission by blood products (14). It is essential to bear in mind that prions are resistant to standard sterilisation procedures, and to follow the guidelines recommended in autopsy procedures and tissue handling. Evaluation of the modes of infection and the disease associations of this novel strain of infectious agent is a major challenge for future research.

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