The Nobel Prize for Physiology or Medicine, 1997.

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The Nobel Prize for Physiology or Medicine in 1997 has been awarded to Stanley B Prusiner of the University of California at San Francisco. The award has been made for a finding that calls into question the very basis of our understanding of what constitutes life. Prusiner has been acclaimed for his 'pioneering discovery of entirely new genera of disease-causing agents and the elucidation of the underlying principles of their mode of action'. The causative factor was identified initially in the case of a disease in sheep and goats known as scrapie. In the itchy (as opposed to drowsy) sub-type of scrapie, affected animals are in the grip of an uncontrollable itch and literally scrape themselves to death. Scrapie is believed to be caused by one member of a class of proteinaceous particles known, following the acronym coined by Prusiner, as *prions*. Prions can be thought of as being at the bottom of a hierarchy of infectious agents that include bacteria and viruses, albeit with an essential difference that we will discuss below. Prion diseases are characterised by extensive damage to the nervous system. Lack of coordination, progressively deteriorating mental abilities and insomnia can be among the symptoms seen in humans. All prion diseases lead to death; there are no cures as yet. The present essay attempts to place Prusiner's discovery in the context of modern biology. But to do that we need to go back into the past.

What one might call the principal thread of the story dates from about 45 years ago. A young medical anthropologist, D Carleton Gajdusek, decided to investigate the basis of a chronic illness called Kuru that had been reported among the Fore and other culturally-related groups of Papua New Guinea. Its onset was sudden and unrelated to any obvious causes. The symptoms were unsteady gait, tremor and involuntary trembling movements, basically a description of the word 'Kuru' in the Fore language. Kuru was found mostly in adult females and children of both sexes and only rarely in adult males. Affected individuals died after a period of 3 to 20 months. In a classic study combining elements of detective work, intuition and hard science, Gajdusek demonstrated fairly convincingly that the transmission of Kuru was correlated with mourning rites involving ritualistic cannibalism during which infected brains were cooked and eaten (that the brains were infected was unknown of course). It is a matter of controversy among anthropologists whether cannibalism has ever formed a part of human culture at all; the fact remains that as incidents of alleged cannibalism in New Guinea have vanished over the past three decades, so has Kuru. Scrapie and bovine spongiform encephalopathy (BSE or 'mad cow disease') are other Kuru-type illnesses, as is Creutzfeldt-Jakob disease (CJD) in humans. The recent epidemic of BSE in Britain is thought to have been triggered by

using the discarded remains of sheep, including brains, in the preparation of cattle feed.

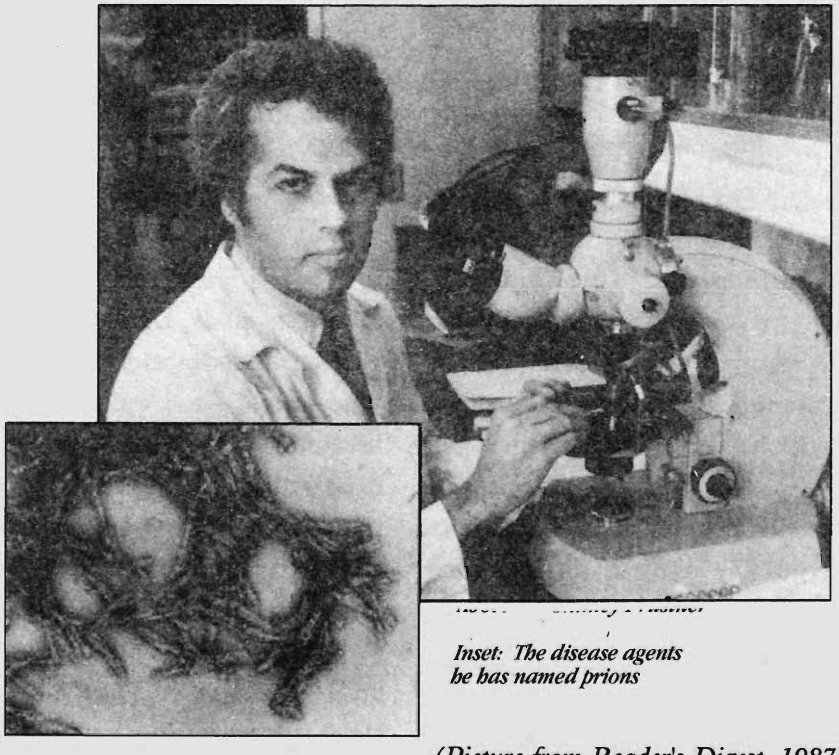
Because the time lag between getting infected and the onset of symptoms the period of incubation - was very high, estimated in some cases as many years, the link between the suspected act of infection and Kuru had been difficult to pin down to everyone's satisfaction. A crucial breakthrough occured in the laboratory study of prion diseases when a technique was developed to transmit scrapie to mice. Even so, the incubation time was about 6 months. The finding that scrapie could also be transferred to hamsters, where the disease could be induced in 60 days, made it more amenable to experimental investigations.

Once the suspicion arose that these diseases could be transmitted, the hunt for the infectious agent, presumed to be an unusually slow-acting virus, was on. The hunt was motivated as much by an interest in the basic biology of the infection as for medical and veterinary reasons. Prusiner began his work in 1972 after he lost a patient to CJD. He chose the scrapie agent for his investigations. Those were the early heady days of molecular biology and a logical course was to isolate the DNA or RNA of the presumed virus. After ten years of work, he and his co-workers stunned the scientific world with the report that they had isolated a single, meaning pure, infectious agent from hamster brains and that the agent was a protein. Try as they might, they could detect neither DNA nor RNA in the infectious particle. The initial reactions to this assertion were almost uniformly sceptical. It was considered impossible to conceive of a living creature (even an honorary living creature such as a virus 'honorary' because it cannot reproduce on its own but needs the unwilling assistance of a host) that did not contain DNA (or RNA).

To understand why, one needs to pick up the second thread of the prion story. This too originates from research carried out in the 1950s, specifically from the finding by J D Watson and F H C Crick that the DNA molecule consists of two mutually complementary strands entwined into a double helical shape. Already at the time there were grounds for believing that hereditary traits of living organisms – for example, virulence in a bacterium – resided in some fashion in a class of molecules known as nucleic acids, DNA being the chief one. Two crucial implications followed from Watson and Crick's discovery. Firstly, it became clear how DNA could make copies of itself with a high degree of fidelity. Secondly, it was seen that DNA sequences could specify the information needed to synthesise both a 'messenger' or 'm' RNA and, via mRNA, proteins. (RNA is a polymer of nucleic acids and its components are closely related to those that form DNA. Proteins act as both the building blocks of the body and, in the form of enzymes, as its work horses.) Soon it was a

matter of text-book knowledge that given DNA and the correct environment, not only could organisms reproduce, but a newly fertilized egg could differentiate and give rise to an adult capable of reproduction in its turn. In short, the principles of both heredity and development seemed to reside in DNA. There is one exception to the rule: RNA substitutes for DNA in some viruses, for example in HIV, but then the virus has to make DNA from the RNA when it needs to reproduce. Spontaneous and unpredictable errors that occur when a DNA molecule is copied give rise to hereditarily transmissible variations in organisms. Such variations are the raw material on which evolution by natural selection acts. In this manner, as living creatures evolve, DNA evolves too. All this goes to explain why the conviction that DNA is more or less synonymous with life has become ingrained in biologists, and why Prusiner's finding appeared heretical.

Thanks to the work of many groups, what was inconceivable until quite recently is taken as possible today, though some



Inset: The disease agents he has named prions

(Picture from Reader's Digest, 1987)

scientists still think that infective prions are associated with traces of DNA or even with a very tiny virus. The consensus, however, is that the active agent in infectious prions very likely is a protein. More surprisingly, it is a modified form of a fairly ubiquitous host-encoded prion protein, PrP for short. To deepen the puzzle, the level of the mRNA that encodes PrP does not differ significantly between infected and uninfected brains. Why then do we all not come down with CJD (or scrapie, or BSE)? The answer seems to be that the protein can exist in two shapes or conformations. In its normal shape the protein is not harmful; indeed, it probably performs a mundane but essential function. But with a very low probability it can change its shape – quite how, nobody knows yet, into a form that is designated PrP-Sc (for Scrapie-causing prion protein). This can occur either spontaneously, when it is a rare event, or as a result of contact with existing Prp-Sc. The phenomenon is similar to what physicists call a phase-transition, and chemists, seeded polymerization. The process can accelerate: a seed of PrP-Sc can catalyse the transformation of normal PrP within the cell or, interestingly, within a test-tube. The resulting high concentration of PrP-Sc can make it aggregate and form tangles that resemble the translucent agglomerates of protein and polysaccharide known as amyloids (that happen to be one of the characteristics of a different disease, Alzheimer's disease). When that happens and the tangles spread, the functioning of the nerve cell and eventually that of the brain is impaired.

Many observations support the model just sketched, in particular the suggestion that a normal, host-encoded gene is an essential element of the pathology. Genetically engineered mice that do not express PrP cannot be infected by scrapie. Curiously, these mice appear normal in all respects. Inoculating PrP-deficient mice with mouse scrapie neither leads to disease symptoms nor causes the production of transmissible prions. The immune system does not respond to scrapie infection. But mice (for example) can be immunized against hamster PrP; when that happens they produce antibodies that react with hamster PrP, not with mouse PrP. It appears that the widespread presence of one's own PrP in the body causes potentially reactive immune cells to be eliminated during the development of the immune system. A company called – appropriately enough – Prionics has recently announced the discovery of a highly specific antibody to PrP-Sc. If confirmed, this would open the way to potential treatment.

The infectivity of prions would seem to be explained: we have a model for how they can make copies of themselves. But can they also exhibit intrinsic variations, give rise to different strains of scrapie, that is? It turns out that they can, most likely on account of mutational changes occuring in

the DNA that encodes them. There is a genetically inherited form of human prion disease, known as Gerstmann-Straussler-Scheinker syndrome (GSS). GSS is accompanied by a single amino acid change in the normal PrP. When the corresponding change is introduced into the mouse PrP gene, it leads to spontanous neurodegenerative disease with symptoms resembling the human disease rather than mouse scrapie, and without the production of infectious particles that can transmit the disease. Fascinatingly, it appears that two heritable traits in yeast that exhibit unusual (non-Mendelian) patterns of inheritance may be transmitted solely by proteins and without the participation of any nucleic acid. If confirmed, this would open up the field to more intensive laboratory investigation. If it is found one day that prion-like entities can also vary on their own, we will have a bonafide candidate for a form of life that, in principle, could be totally independent of nucleic acid chemistry.

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