PRESS RELEASE

Research reveals the importance of various cofactors in determining the biological properties of different prion strains

The work advances understanding of the pathogenesis of prion strains and their ability to propagate in different tissues and hosts

Furthermore, the research proposes a mechanism potentially explaining the different locations of prion strains in the central nervous system and its periphery

Findings from the study on prion strains may be applicable to other neurodegenerative diseases such as Alzheimer's and Parkinson's

Joaquín Castilla, an Ikerbasque researcher in CIC bioGUNE, is the lead author of the article published in the journal Acta Neuropathologica

(Bilbao, 16 November 2017). A study led by CIC bioGUNE has confirmed the importance that various cofactors may have in determining the biological properties of different prion strains. These different biological factors (referred to as cofactors) may, despite not necessarily being an integral part of the infectious prion, restrict propagation of certain strains and thereby determine the characteristics of those strains conducive to being propagated by them.

The research, conducted in collaboration with the University of Santiago de Compostela, the University of Salamanca, the Central Veterinary Laboratory (Madrid) and the *Istituto Superiore di Sanità*/ Italian Higher Institute of Health, also proposes a mechanism potentially explaining the different locations of the diverse prion strains in the central nervous system and its periphery, which would depend on the cofactors available in each tissue and tissue region.

The work has been published in Acta Neuropathologica, one of the most prestigious journals in the field of pathogenesis of all types of neurological diseases. As Joaquin Castilla, Ikerbasque Research professor in CIC bioGUNE and research study leader, explains: "This work paves the way for a greater understanding of what to date has been a highly complex and little-understood phenomenon... namely, the existence of different strains – similar to the case of viruses – of the same misfolded protein. The idea that cofactors may govern or restrict the variability of prion strains is the starting point for studying the relevant cofactors in the natural processes of infection and, having identified them, for being able to predict the properties of the different strains such as their capacity for neuroinvasion and their facility for interspecies transmission".

The main difficulty encountered in this research project, which has been conducted over the last 5 years, has been in distinguishing between the different strains. In many cases,

despite having distinguishable biological properties, the strains are very similar, and distinguishing between them demands detailed knowledge of the experimental models used. Furthermore, generating and maintaining different prion strains of the same species is a highly complex process from the technical perspective due to potential cross contamination. For this reason, an expert laboratory in *invitro* propagation of prions has been required.

Causes of prion diseases

Prion diseases are caused by the aberrant folding or misfolding of the cellular prion protein (PrP^{C}) to a pathogenic isoform named PrP^{Sc}. The different conformations PrP^{Sc} may acquire means that prions may exist in different strains or conformational variations characterised by specific pathological properties (incubation periods, brain areas affected, symptomatology, etc.) likely encoded in its three-dimensional structure.

Up to now, exactly how these conformational variations appeared, and how the different biological factors (referred to as cofactors) impacted on the different conformations of PrP^{Sc} and determined their specific pathological properties, was unknown.

To understand how different cofactors modulate prion strain generation and selection, a system for in-vitro prion propagation known as Protein Misfolding Cyclic Amplification (PMCA) was used. This test tube system reproduces, in controlled conditions, the process of misfolding from PrP^{C} to PrP^{Sc} which occurs in humans and animals affected by these devastating diseases.

Given that the presence of all the cofactors found in the brain could give rise *in vitro* to a similar variety of strains to those observed in natural conditions, a diverse set of infectious recombinant prion strains (capable of inducing a prion disease in animal models) were generated by using bacterially expressed (recombinant) prion proteins in the presence of mouse brain homogenate not containing PrP^{C} and the use of PMCA. Having obtained different strains and a mix of infectious strains in the presence of brain homogenate and with distinguishable pathological properties, the propagation medium of these prions was then changed. The same recombinant proteins were used but supplemented, in this case, by one single cofactor (different molecules selected due to their interaction with this protein). It could then be observed how different cofactors are capable of driving the evolution of mixed prion strains towards specific and particular cofactor conformations.

As Dr. Castilla further points out: “Our results show that a variety of infectious recombinant prions can be generated *in vitro* and that their biological properties are dependent on the cofactors available during the propagation process. These observations have significant implications for understanding the pathogenesis of prion diseases and their ability to replicate in different tissues and hosts, which may depend on the relative abundance of different cofactors”.

Findings from research on prion strains may be applicable to other neurodegenerative diseases (for example, Alzheimer's and Parkinson's) for which misfolded proteins with slightly different conformations have been described.

About CIC bioGUNE

The Centre for Cooperative Research in Biosciences (CIC bioGUNE), located in the Bizkaia Technology Park, is a biomedical research organisation conducting cutting-edge research at the interface between structural, molecular and cell biology, with a particular focus on generating knowledge on the molecular bases of disease, for use in the development of new diagnostic methods and advanced therapies. CIC bioGUNE has been accredited as a "Severo Ochoa Centre of Excellence", the highest level of recognition for centres of excellence in Spain.