Molecular origins of Parkinson's Disease

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Neurodegeneration and Parkinson's Disease

Neurodegenerative diseases are a major cause of disability, suffering and health care costs [1]. Now that our population is aging [2] we will see an increase in the number of patients suffering from these diseases as age is the largest risk factor [3, 4]. Relatively common diseases are Alzheimer's Disease and Parkinson's Disease (PD) but more rare diseases include Amyotrophic Lateral Sclerosis, Creutzfeldt-Jakob Disease ('mad cow disease') and Huntington's Disease [5]. Despite having different molecular origins, disease progression and symptoms, these diseases are all chronic, progressive and thought to be caused by specific protein being present in an aberrant conformation (structure/shape): they are all protein misfolding diseases (PMDs) (see Chiti and Dobson, 2006 for a review [6]).

PD was first described by James Parkinson in his work of 1817 'An Essay on the Shaking Palsy'. In it he describes the history of the condition and several case studies [7]. He states how at that point in time the precise nature of the disease is not understood but 'it ought not to be considered as one against which there exists no countervailing remedy.' Two centuries later our knowledge on the nature of the disease has vastly increased with symptomatic treatments available but sadly no curative treatment is available as of yet.

PD is progressive and characterised by resting tremor, bradykinesia (slowness of movement) and rigidity and loss of postural reflexes (needed for maintaining posture, balance and fluidity of movement) and can be accompanied by a plethora of additional clinical features such as cognitive neurobehavioral abnormalities and sleep disorders (reviewed by Jankovic et al. 2008 [8]). After 20 years of suffering from PD, 80% of surviving patients show dementia [9]. Disease onset is generally between 55-75 years of age [8] but often under age 50 for certain hereditary forms of PD [10].

PD causes death of neuronal cells and is thus called a neurodegenerative disease [11, 12]. The disease is characterised by the presence of clusters of material inside the cell which are called Lewy Bodies. These clusters are mainly composed of the protein α-synuclein (AS) (reviewed by Wakabayashi et al. 2013 [13]). The current view is that these clusters may not be harmful to neurons but instead might be protective by trapping harmful molecular structures [14, 15].

Protein misfolding and aggregation

PMDs are characterised by the misfolding and aggregation of native protein (which is the state of the protein in which it is properly folded into its functional conformation) leading to either a gain of toxic function and/or loss of physiological function of the protein at hand [16].

Proteins are macromolecules made up of amino acids. Proteins are important structures for life as they perform many crucial functions inside as well as outside of cells. There are twenty two different naturally occuring amino acids, each with a different structure. The number of amino acids and the order in which they are linked together to form a protein determine the structure of the protein and thus its function. Hydrophobic ('water fearing') patches of protein are usually buried inside

the functional shape of the protein, whilst polar (charged) residues are exposed to make the protein soluble in water [17, 18]. However, misfolding into a non-native partially or globally unfolded state can cause exposure of hydrophobic patches which makes the protein less soluble and more prone to interact with other protein increasing the odds of forming a nucleus [19]: an aggregate of protein which is capable of misfolding and inducing aggregation of other proteins in solution [20]. Intrinsically disordered proteins (IDPs) are suggested to be more prone to aggregation as these do not natively fold, which would be protective [21]. One such IDP is AS; the protein implicated in development of PD. AS is localised at the synaptic terminal (the part of neurons where communication with other neurons takes place) where it is suggested to have a regulatory role in transmission of signals [22, 23]. In addition, the protein is also localised in the nucleus (the large central organelle of a cell where the DNA, amongst other things, is present) where it is found to co-localize with proteins that are involved in structuring DNA, suggesting a function of AS in regulation of gene expression [24, 25].

Protein aggregation is the formation of large aggregates of insoluble protein with a high proportion of what is called β-pleated sheet (a specific conformation of a sequence of amino acids), termed amyloid [26]. Some amyloid is functional such as amyloid formed from the Curlin protein in the bacterium *E. coli* where the protein is involved in biofilm formation (groups of bacteria clustering together on a surface) and binding to host proteins [27, 28]. However, some amyloid is not functional and instead forms fibrils/aggregates inside or outside of the cell.

For many years it was thought that amyloid fibrils (long strands of misfolded proteins linked together) were the end product of the cascade of misfolding and aggregation and that this structure caused neuronal cell death (discussed by Neve and Robakis, 1998 [29]). However, in the past few years this view has changed by findings that intermediate species, called oligomers, might be most harmful to neurons (reviewed by Kayed and Lasagna-Reeves, 2013 [30]). Tremendous effort has been put into unraveling the different processes involved in protein misfolding and aggregation.

The protein aggregation cascade

Currently, several different types of protein aggregates and mechanistic steps have been identified which are either on- or off pathway from soluble monomeric protein (a single molecule of protein in solution) to amyloid fibrils, outlined in Figure 1: Monomeric protein (structure 1) may interact to form dimers and multimers/oligomers (consisting of two or more protein molecules respectively). Oligomeric species can be either on-pathway to becoming amyloid [31] (structure 2) or off-pathway [32, 33] (structure 3) (discussed by Bemporad and Chiti, 2012 [34]) and these oligomeric structures have been implicated to be the molecular species causing cell death through disruption of membranes (which are components of the cell) [35–38]. The oligomeric structure may undergo a conformational change and form a nucleus (structure 4) giving the aggregate properties to propagate by addition of monomers to the growth ends. However, it has also been suggested a monomer could even be considered a nucleus if it acts as a template for growth of amyloid (discussed by Bemporad and Chiti, 2012 [34]). The structures formed by elongation of the nucleus are sometimes called pre- or proto-fibrils and are on-pathway to forming mature amyloid fibrils (structure 5) [39, 40] via a process called 'maturation' (omitted from the schematic illustration for simplicity) [41, 42].

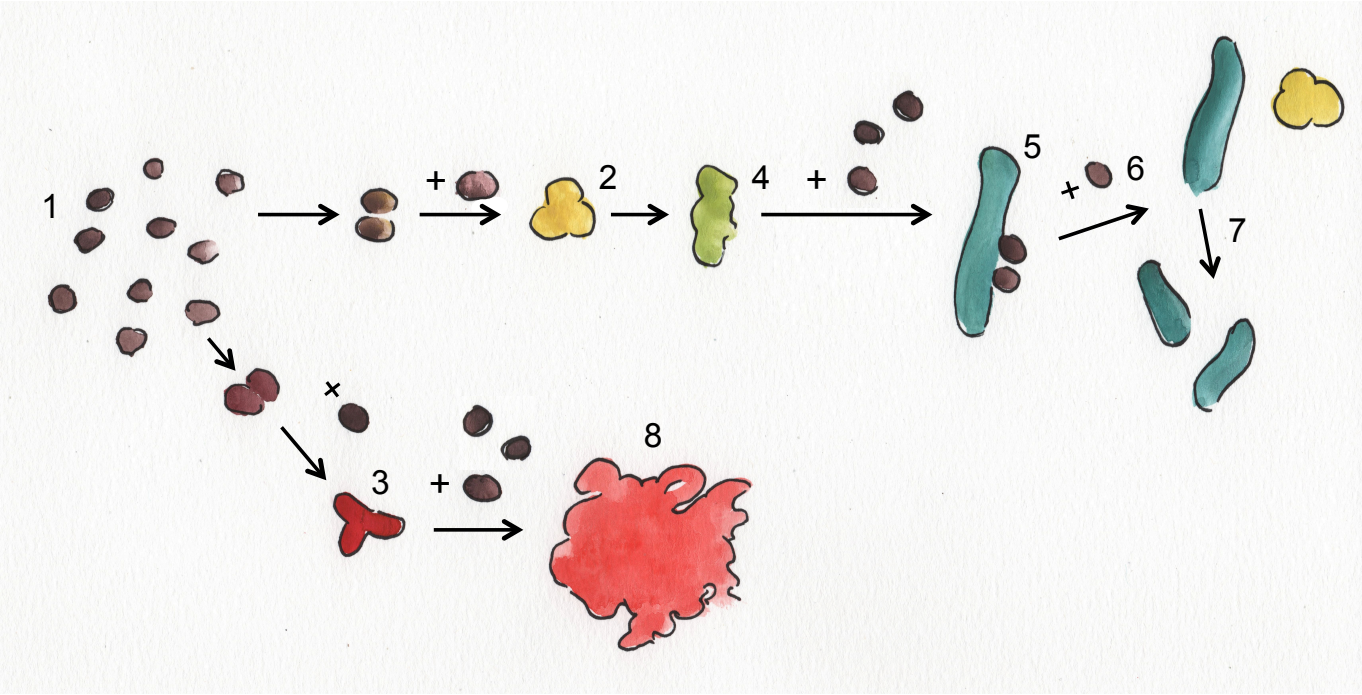

Figure 1: Amyloid formation takes place via several steps. Schematic representation of protein aggregation through several steps from soluble, functional monomeric protein (1) to oligomers (2 and 3), formation of a nucleus (4) and elongation of said nucleus into fibrils (amyloid) (5). Surface catalysed nucleation (6) (where soluble protein interacts with fibril surfaces to form oligomers or nuclei) and fibril fragmentation (7) might also take place and can greatly enhance the conversion rate of soluble protein into amyloid. Formation of amorphous aggregates (8) (which do not have a defined structure like amyloid does) can also occur depending on experimental conditions.

Under certain conditions, monomeric protein may interact with the fibril surface, resulting in formation of oligomers [38, 43] (at No. 6) that may go on to form amyloid fibrils. Fibrils may fragment (at No. 7) either naturally or induced by, for instance, disruption of the solution by stirring (in an experimental setting) which enhances the number of growth ends and by this, enhances initial extension (elongation) rate and toxicity to cells as shorter fibrils appear to be more harmful to cells [44, 45]. Amorphous aggregates (structure 8), which lack the defined structure that amyloid has, can also form depending on environmental conditions.

Final thoughts

PD, amongst other diseases, is accompanied by misfolding and aggregation of protein which normally has a function in the body. Several different molecular steps have been identified and efforts are made to find molecules that interfere with one or more of these steps aiming to halt or slow down development of PMDs once diagnosed [46, 47]. One caveat is that when patients start presenting with symptoms, too much damage might already have been done meaning that a full or even partial recovery might not be possible. In light of this, work is also carried out to refine diagnostic methods aiming to diagnose people before they have developed symptoms increasing the chances of succesful treatment. It is thus incredibly important to approach PMDs from different angles and on different levels if we are to offer the best possible treatment to people with these debilitating illnesses.

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