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Evaluate how genetics and biochemistry impact Parkinson's disease and explore recent advances in its treatment

Abstract

The mechanisms and pathology that underly Parkinson's disease are extremely complex and have been subject to study since the discovery of the disease. A massive contributing factor to the lack of a current cure is the influence from both inherited genetics and environmental conditions, but greater understanding of the fundamental processes at play would provide a better foundation for development of new treatments. Two targets of current ongoing research are the alpha-synuclein and leucine-rich repeat kinase 2 proteins, each having distinct structures and characteristics that enable progression of the disease. Gaining a clearer picture of their biochemistry, along with constant advancements in the field of genetics, has paved the way to investigating innovative and alternative forms of therapy.

Introduction

Parkinson's disease (PD) is a progressive neurodegenerative disorder, second only to Alzheimer's in prevalence. Though it is most common globally in individuals over 80 years old (3% affected compared to 0.3% of the global population), there are a number of other potential risk factors and genetic predispositions to the disease. The disease is characterized physically by tremors, slowed movement known as bradykinesia and general abnormal motor function. In turn, this can impact an individual's balance, posture, speech and written communication (5). There is further potential for the disease to progress into Parkinson's disease dementia (PDD) if the cognitive functions of the brain also become damaged. The multiple forms of PD, in addition to other neurodegenerative diseases, are characterized by proteinaceous inclusions known as Lewy bodies (LBs) present in affected areas of the brain (6). With regards to Parkinson's disease, the dopaminergic neurons (DA) of the substantia nigra in the midbrain are the most notably affected. The damage and death of these neurons is the cause of the resulting loss of motor capability. Basal ganglia dysfunction leading to worsened coordination and motor learning, has also been associated with PD-linked neurodegeneration. Recent years have led to the discovery of some key proteins that appear to be incredibly influential to the progression of Parkinson's disease, with the hope that they can be utilized in the efforts to more effectively treat the disease.

Alpha-synuclein

1. Structure

The intracellular alpha-synuclein protein is primarily located in the presynaptic terminals and is comprised of an alpha helix domain, a non-amyloid component (NAC) and an acidic tail as illustrated in Fig. 1 (6). The alpha helix has distinct lipid-binding properties, allowing influence over lipid membranes and their interactions. The significance of the abundant a-synuclein proteins lies in their ability to misfold and aggregate. The NAC domain has the capability to form cross beta-sheets and includes a strongly hydrophobic sequence (4). This structure lends itself to conversion into oligomers, then further aggregation into fibrils, before forming the complete Lewy bodies deposited throughout the neuronal pathways (9).

Figure 1. Alpha-synuclein protein. The 140 kDa structure is comprised of three domains, an N terminal alpha helix, a non-amyloid component (NAC) and a C terminal acidic tail (Ajpolino, CC0, via Wikimedia Commons).

2. Role in PD

Although Parkinson's disease damages many neuronal pathways, there are specific areas where cell death is more acute: dopaminergic neurons (DA) in the midbrain, and the nucleus Basalis of Meynert (nBM). These areas are associated with the type of muscle movement affected by PD, though the reasoning behind why these areas appear to be targeted isn't clear (12). The method of neuron death is thought to involve the alpha-synuclein protein. When correctly functioning it plays a role in neurotransmitter release, vesicle docking and fusion (4). In the context of PD, misfolded a-synuclein is present as the primary component in Lewy bodies (LB), first described in 1912 (4), as seen in Figure 2. LBs are disruptive protein deposits found in the brain, and are strongly associated with neurodegenerative diseases including dementia and Parkinson's. The typical loss of motor - and later cognitive - function observed in individuals with PD may correlate to the location of a-synuclein, as its

pathology has been found in the structures responsible for these functions (6). Recent research suggest that the formation and presence of Lewy bodies alone is not sufficient to cause the extent of neuronal degeneration exhibited in PD. This has lead to suggestion that a-synuclein itself is of key importance. The process by which a-synuclein initiates neuron death is relatively unclear but is likely to feature the misfolded protein being transported between neurons (6). The overarching question here is what drives the protein to misfold initially? Disrupting the formation of LBs may not lead to a direct cure, but it would provide vital information for furthering the understanding of alpha-synuclein interactions.

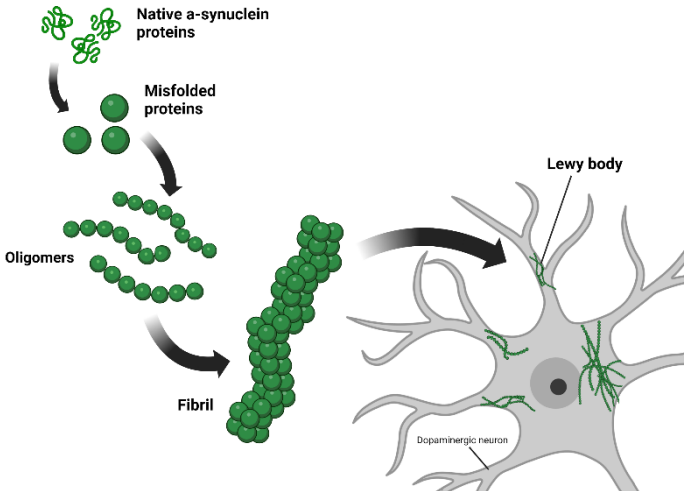

Figure 2. Misfolding and aggregation process of alpha-synuclein protein. The primary component of Lewy bodies are the aggregated fibrils of a-synuclein. The native protein can be triggered into misfolding from the influence of pre-folded fibrils before forming oligomers and later forming part of a Lewy body. Diagram adapted from (9) using Biorender.

3. Mutation

A large number of chromosome and point mutations have been identified with potential for causing PD-like symptoms. However, onset of the disease is often due to a combination of risk factors, so while useful, confirming a single source of mutation will not bring about a cure in itself. Despite this, a deeper understanding of the genetic components involved and their effects will open up the possibility of tailoring treatments and therapies specifically to patients. Mutant alpha-synuclein proteins are encoded from mutations carried on the SNCA gene. All known a-synuclein mutations are found within the N-terminal amphipathic (AMP) helical domain. These mutants have been associated specifically with onset of autosomal dominant PD. There is a further suggestion that the mitochondrial targeting signal sequence found repeated in this domain could be the cause of the mitochondrial dysfunction often observed in PD patients. (11). Experiments were conducted with a-synuclein proteins pre-

folded fibrils (PFFs) that were introduced to wild type neurons in an effort to investigate the transmission mode of a-synuclein misfolding. It was found that in the absence of any reagents, the PFFs were accepted by the primary neurons and able to influence misfolding of the endogenous a-synuclein. Further research discovered that a-synuclein extracted from the brains of individuals with Parkinson's disease was capable of inducing this same pathology in wild type neurons (6). This exposes a lot of key understanding about Lewy body formation and pathology, especially when seen as a potential target process for inhibition or mutagenic knock out of genes involved in this transmission.

Leucine-rich repeat kinase 2 (LRRK2)

1. **Structure**

LRRK2 is a large protein (286 kDa compared to the 140 kDa of alpha-synuclein) with multiple domains (Fig. 3). LRRK2 features protein-protein interaction domains, GTPase activity and a serine/threonine kinase (8). The abundant expression of LRRK2 throughout the brain and some peripheral organs, as well as the potential for phosphorylation activity on several of its domains, means it plays a role in multiple signaling pathways.

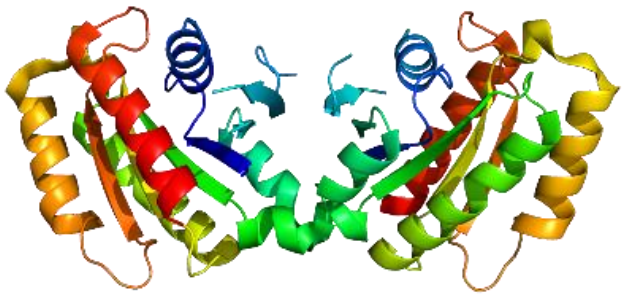

Figure 3. Leucine-rich repeat kinase 2 (LRRK2) protein. A large protein consisting of many domains including two catalytic: Ras of complex (Roc) proteins and a serine/threonine kinase (Pleiotrope, Public domain, via Wikimedia Commons).

2. **Role in PD**

Despite the seemingly wide array of functions LRRK2 is capable of, its activity can be divided into GTPase and kinase, due to its multiple domains. This dual functionality means the protein has a part in trafficking of vesicles and synaptic function, in addition to its presence in immune processes such as inflammation, phagocytosis and tissue repair (11). There is ongoing discussion surrounding the extent of the influence LRRK2 has on alpha-synuclein activity and their interaction itself, though it is known to be involved in the aggregation induction process between cells.

3. Mutation

The size of leucine-rich repeat kinase 2 and the number of its domains permit a wide range of possible mutations, over 100 genetic variants have been found thus far. The extent of biochemical activity LRRK2 partakes in allows a mutation to disrupt any number of processes. Some of the LRRK2 mutations are associated with several forms of Parkinson's disease, especially idiopathic and sporadic PD, and are the most common genetic cause of the disease. Most notably, LRRK2 is involved in the development of neurites. Neurites form axons or dendrites by extending the plasma membrane on developing neurons (14). The presence of LRRK2 mutants promoted disruption to this process causing neurite shortening, a source of neuropathic pain and contributing to neurodegeneration. The most common and one of the most significant pathogenic mutants of LRRK2 is the G2019S variant, featuring a substitution of glycine to serine. G2019S has been implicated in increased transmission of misfolded alpha-synuclein proteins between neurons, due to its function in lysosome regulation and secretory pathways as illustrated in Fig. 4 (7).

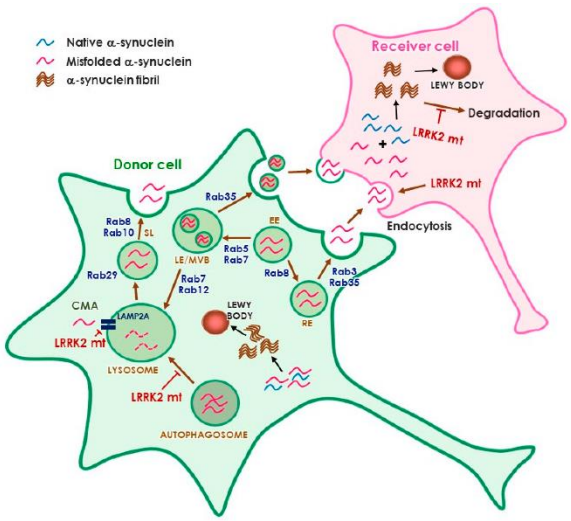

Figure 4. The suggested role of LRRK2 in the a-synuclein transmission pathway. Due to the part LRRK2 plays in endosomal regulation, the transport of endogenous misfolded a-synuclein via lysosomes is mediated by LRRK2 interaction (14).

Treatments

Due to the complexity of Parkinson's disease pathology and neurodegenerative disorders generally, there is currently no direct cure for the disease. Treatments and therapies in use mainly aim to alleviate some of the symptoms of PD, since the root cause of the disease is still largely unknown.

1. Targeting a-synuclein

Though only a contributing factor to the onset and progression of Parkinson's disease, reducing or preventing the damage done to dopaminergic neurons by a-synuclein would be a significant step forward. As a-synuclein is endogenous, specific attack of the protein itself proves difficult. To overcome this, the main routes being investigated are the production, misfolding, aggregation and transmission of the protein. Despite the exact secretion and uptake mechanism not being fully understood, there are known receptors involved in the spread of a-synuclein across cells. Lymphocyte activation gene 3 (LAG-3) is an immune cell-surface receptor that poses a potential therapeutic avenue (4). It's function lies in T cell activation and the proceeding response actions. Due to its role in maintaining immune homeostasis, it is understandable that LAG-3 has been associated with increased risk of PD. There is suggestion that the initial aggregation of a-synuclein could be due to disfunction of this receptor (2).

2. Use of stem cells

One of the most promising routes of research focuses on personalized treatments and the application of stem cells. The hope is to target localized neuron degeneration, to slow or stop progression of the disease. One of the most attractive features of the use of stem cells is the potential lack of immune rejection from the individual, as the procedure will be patient-specific (12). As the disease causes massive degeneration of dopaminergic (DA) neurons, experiments have taken place using human foetal brain tissue transplantation, more specifically mesencephalic tissue due to the high density of DA neurons. Donor tissue was grafted into the brains of Parkinson's patients, with hopes to replace lost neurons and potential control progression of the disease and its symptoms. Obstacles were reached due to difficulty in standardization, low tissue availability, ethical dilemmas and rejection of the transplantations (10). Significant advancements in the field of stem cell development and manipulation have been made over recent years; these show promise for avoiding use of human foetal tissue altogether in the form of somatic nuclear transfer and induced pluripotent stem cells (3).

3. Cell reprogramming

A limitation of using human stem cells, even when generated rather than donated, is the pluripotent stage of differentiation that harbours the possibility of forming tumours. This tumourigenicity can have devastating clinical implications (1). Interest in an alternative solution has mounted: direct conversion of somatic cells into induced dopaminergic neurons (iDNs). The process involves controlling the cell fate of somatic fibroblasts by diverting their lineage to produce functional DA cells, and is referred to as direct lineage reprogramming (DLP). Currently the low efficiency rate of reprogramming bars the immediate use of iDNs in

PD therapies, though efforts to enhance the conversions are ongoing. One avenue with potential is the addition of nanomaterials as promotors in transdifferentiation. Specifically, the introduction of modified gold nanoporous materials (AuNpRs) has been investigated and found to enhance fibroblast conversion to iDNs by alleviating the level of oxidative stress caused by the process (13). The rate at which developments continue to be made in the field of cell programming and epigenetics is exciting, and elicits hope for the future of Parkinson's disease treatment.

Conclusion

One thing known for certain is that Parkinson's disease remains an extremely complex neurodegenerative disease, that is often life-changing for the individual and those around them. Whether familial or idiopathic PD, the interplay of genetics, biochemistry and environmental factors does not provide an easy path to a cure. The sheer number of genetic variants present of a-synuclein and leucine-rich repeat kinase 2 alone is a challenge to many proposed treatments. However, the constantly progressing routes of PD therapy are inspiring, and if applied in conjunction with current palliative care, the aim to improve quality of life due to Parkinson's disease is achievable.

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