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Calcium and Parkinson's disease

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Abstract

Parkinson's disease (PD) is the second most common neurodegenerative disease in the world. Its causes are poorly understood and there is no proven therapeutic strategy for slowing disease progression. The core motor symptoms of PD are caused by the death of dopaminergic neurons in the substantia nigra pars compacta (SNc). In these neurons, Ca^{2+}entry through plasma membrane Cav1 channels drives a sustained feed-forward stimulation of mitochondrial oxidative phosphorylation. Although this design helps prevent bioenergetic failure when activity needs to be sustained, it leads to basal mitochondrial oxidant stress. Over decades, this basal oxidant stress could compromise mitochondrial function and increase mitophagy, resulting in increased vulnerability to other proteostatic stressors, like elevated alpha synuclein expression. Because this feedforward mechanism is no longer demanded by our lifestyle, it could be dispensed with. Indeed, use of dihydropyridines – negative allosteric modulators of Cav1 Ca^{2+} channels – comes with little or no effect on brain function but is associated with decreased risk and progression of PD. An ongoing, NIH sponsored, Phase 3 clinical trial in North America is testing the ability of one member of the dihydropyridine class (isradipine) to slow PD progression in early stage patients. The review summarizes the rationale for the trial and outlines some unanswered questions.

Keywords

Substantia nigra; dopamine; electrophysiology; mitochondria; oxidant stress; two photon microscopy; endoplasmic reticulum

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What causes PD?

PD poses a major health care challenge worldwide, disabling millions. The cardinal symptoms of the disease are motoric: slowness of movement, rigidity and resting tremor [1,2]. Although there are palliative treatments, there is no proven strategy for slowing the progression of the disease. This hole in our therapeutic arsenal stems from an incomplete understanding of what causes the disease.

Most of what we know about PD etiology comes from two very different lines of investigation. The first line of study is built upon clinical observation and pathological analysis of human PD brains. Using this approach, the core motor symptoms have been traced to the loss of dopaminergic neurons in the substantia nigra pars compacta (SNc), a small region in the mesencephalon. These dopaminergic neurons innervate the basal ganglia, a subcortical structure involved in the control of movement and action selection. In early stage patients, boosting the production of dopamine (DA) by systemic administration of a brain penetrant DA precursor (levodopa) alleviates symptoms, establishing a clear causal connection between the loss of dopaminergic neurons and motoric symptoms[2].

More detailed study of the brains of PD patients has revealed two additional features of the disease that are important to understanding its causes. First, there is neurodegeneration in other parts of the brain aside from the SNc. Although reliable quantitative estimates of neuronal loss have not been systematically compiled, in advanced PD patients there is compelling evidence of neuronal loss in a handful of other structures, like the locus ceruleus (LC), pedunculopontine nucleus (PPN), intralaminar nuclei of the thalamus (ILT) and lateral hypothalamus (LH), in advanced PD patients [3]. Importantly, these other neurons do not use DA as a neurotransmitter, arguing that DA is not uniquely linked to degeneration. Second, there is evidence of proteostatic failure in several of the neuron types affected in PD. The principal piece of evidence for this conclusion is the observed accumulation of intracellular protein aggregates in the brains of PD patients. These were first described by Lewy [4] and called Lewy bodies when they appear in the somatic region of neurons or Lewy neurites when in an axon or dendrite. A major component of this Lewy pathology (LP) is phosphorylated alpha synuclein (αSYN). What is less clear is the relationship between LP and neurodegeneration. Although commonly observed in neurons affected by PD, the question of whether these aggregates represent a cause, as opposed to a marker, of cellular degeneration is not established.

The second major line of study that has informed us about PD mechanisms is built upon genetics [5]. About one in ten PD cases is familial; that is, the appearance of the disease can be traced to a genetic mutation. Both recessive and dominant mutations associated with PD have been identified [6]. The best characterized, recessive, loss-of-function mutations have been clearly linked to mitochondrial maintenance. *PINK1* and *PARKIN* mutations in non-neuronal cells lead to deficits in mitochondrial turnover or quality control[7]; whether this is also the case in neurons is controversial. Parkin also has been implicated in mitochondrial biogenesis through its negative regulation of the transcriptional repressor PARIS [8]. Mitochondrial biogenesis is promoted by the transcriptional co-activator PGC-1a and its target gene, NRF-1. In the absence of parkin, paris accumulates and leads to the progressive

loss of dopaminergic neurons in the SNc. Mutations in *DJ-1* also fall into this category; DJ-1 helps regulate cellular and mitochondrial oxidant defenses. All three of these genetic mutations are linked to mitochondrial function and increase the probability of an early onset form of PD [9,10]. Of note, LP is not an obligatory component of the pathological profile in many of the PD patients with these mutations.

Autosomal dominant mutations associated with PD have not yet been linked to any particular cellular pathway. Mutations or copy duplications/triplications of *SNCA* are associated with normal age-of-onset (~60 years old) PD. As *SNCA* codes for αSYN – a major component of LP – the proposition that elevated or misfolded αSYN drives PD pathogenesis has gained widespread acceptance. But precisely how this might happen is still unclear. One hypothesis is that oligomeric fibrils of αSYN behave like a prion, catalyzing the creation of new fibrils that spread from neuron to neuron leaving a trail of LP [11,12,13]. Although intriguing, it is not obvious how a process like this could create a relatively stereotyped pattern of pathology in PD patients without additional factors coming into play. Mutations in a number of other genes, including LRRK2, also are associated with PD [6,14,15] Considerable progress has been made toward understanding the function of the proteins encoded by these genes. While functions including autophagy and vesicular trafficking and exocytosis appear to be affected a coherent thesis regarding pathogenesis has not yet emerged. A persistent limitation in this line of study is that none of the genetic mutations associated with PD recapitulates the disease in mice, suggesting that additional factors contribute to the disease phenotype.

Although identification of specific genetic defects in familial cases of the disease offers important clues about pathogenesis, the vast majority of PD cases are idiopathic and lack a discernible genetic cause. Environmental toxins have been implicated in a few cases, but they cannot explain the vast majority. The factor most closely linked with PD onset is aging. The average age at the time of idiopathic PD diagnosis is around 60 years; PD risk increases steadily beyond that point [16,17]. Unfortunately, little data exists on the effects of aging in the brain that would lead to a better understanding of why this disease appears late in life and how it could be stopped.

What does the pattern of pathology in PD tell us?

Another clue about pathogenesis in PD is the regionally selective nature of the pathology. As mentioned above, the selective loss of SNc dopaminergic neurons is responsible for the core motor symptoms of the disease. What makes them vulnerable? There are three features of these neurons that may be mechanistically linked to their selective vulnerability: 1) the reliance upon DA as a neurotransmitter; 2) a highly branched and long axonal arbor; 3) a distinctive pacemaker phenotype that relies heavily upon Ca^{2+} entry through Cav1 channels and leads to basal mitochondrial oxidant stress.

Regarding the first two, DA has long been thought be a risk factor for SNc DA neurons [18,19,20]. The combination of elevated DA, cytosolic Ca^{2+} and αSYN could be a particularly toxic combination, especially in axon terminals [21,22,23]. Consistent with this idea, DA axon terminals are lost early in the development of PD, preceding the loss of DA

cell bodies [24]. Thus, cellular stresses associated with DA metabolism could put these cells at risk. However, three facts are difficult to reconcile with the DA hypothesis. One is that levodopa therapy does not seem to accelerate disease progression [25]; if DA was the toxin driving the disease, this would not be true. Another is that many non-DA neurons are affected in PD, either degenerating or manifesting LP. Third, DA neurons in the ventral tegmental area (VTA) are relatively protected in PD. Thus, while DA is associated with vulnerability of SNc neurons, there must be other factors in PD [3].

The long, highly branched axon of SNc DA neurons is another potential risk factor. SNc DA neurons in the rodent have axons that branch profusely in the striatum and possess as many as 200,000 vesicular release sites [26]. It has been proposed that the bioenergetic demand of sustaining electrical excitability in a long unmyelinated axon like this puts SNc DA neurons at risk [27,28]. The aggregate energy consumption of a long axon is undoubtedly substantial. The question is whether this creates a bioenergetic stress, possibly related to active axonal transport functions. Recent work has provided some direct support for this hypothesis [29], by showing that *in vitro* mitochondrial oxidant stress is higher in SNc DA neuron axons than in the less branched axons of VTA DA neurons. Moreover, reducing the size of the arbor decreases this stress. It is also possible that this long axon poses a proteostatic burden on the somatic compartment [30]. Another possibility is that a highly branched axon is an efficient conduit for retrogradely transmitted pathogens, such as misfolded αSYN, simply because of the large number of synaptic uptake sites [31]. However, there are neurons with long, highly branched axons with as many release sites as those of SNc DA neurons, which do not succumb in PD [32]– arguing that a long axon alone is not a primary cause of neurodegeneration or LP in PD.

The third major hypothesis of selective vulnerability of SNc DA neurons is based upon physiological studies. SNc DA neurons are autonomous pacemakers, generating action potentials (spikes) at a relatively slow rate (2–10 Hz) in the absence of synaptic input [33,34,35,36,37,38,39]. The spikes in these neurons are broad, which maximizes Ca^{2+} entry and promotes slow rhythmic activity [40]. The autonomous activity is accompanied by slow oscillations in intracellular Ca^{2+} concentration that are caused by the opening of plasma membrane Cav1 Ca^{2+} channels and release of Ca^{2+} from endoplasmic reticulum (ER) and by the absence of strong buffering of intracellular Ca^{2+} [36,38,41,42,43,44,45,46]. Channels with a pore-forming Cav1.2 subunit and channels with a Cav1.3 subunit contribute to this oscillation in SNc DA neurons. The Cav1.3 channels are particularly interesting because they open at relatively hyperpolarized membrane potentials [47,48,49]. In fact, because of the depolarized operating range of SNc DA neurons, the Cav1.3 channels never close fully during the pacemaking cycle [36,39]; this and the expression of C-terminal splice variants of the Cav1.3 subunit, which minimize Ca^{2+}-dependent inactivation, result in a sustained Ca^{2+} influx [50,51,52] Recently it has also been shown that low threshold L-type Ca^{2+} channels, most likely Cav1.3 channels, contribute strongly to retrograde-propagation of spikes in dendrites, further increasing Ca^{2+} entry and modulating the synaptic gain upon stimulation and consequently the initiation of burst firing [53]. Underscoring the potential importance of this Ca^{2+} oscillation, there is a clear inverse correlation between the expression of the Ca^{2+} binding protein calbindin and risk of degeneration in PD patients [54,55]; this correlation is

also found in transgenic mice [56]. Hence, loss of localized intracellular control of Ca^{2+} oscillations may contribute to neuronal vulnerability to other stresses.

Initially, pacemaking activity was thought to be dependent upon Cav1 Ca^{2+} channels because dihydropyridine (DHP) inhibitors slowed or stopped pacemaking [38,44,57,58]. However, the DHP concentrations used in these experiments were orders of magnitude greater than the IC_{50} of recombinant channels [59,60], at these concentrations both Na^+ and K^+ channels were also antagonized [38,44,45,57,58,61]. Using Ca^{2+} imaging of dendrites to monitor DHP antagonism of L-type channels, it was shown that nanomolar or low micromolar DHP concentrations eliminated dendritic calcium oscillations (reflecting near complete antagonism of L-type channels) without altering pacemaking rate or regularity [45]. In the absence of Cav1 channels, pacemaking was sustained by a combination of voltage-dependent Na^+ and K^+ channels in conjunction with a voltage-independent leak channel [39,45]; hyperpolarization and cyclic nucleotide-activated cation (HCN) channels, which are prominent in SNc dopaminergic neurons [37], also contributed to the sustained pacemaking activity. These channels appear to reside in the soma, proximal dendrites and axon initial segment.

The slow cytosolic Ca^{2+} oscillations in SNc DA neurons sub-serve three complementary functions. First, even though they are not required for pacemaking, they help maintain the slow tonic spiking in these neurons [39,44,45,62]. Second, Ca^{2+} positively modulates the expression and activity of enzymes involved in DA synthesis, ensuring that there is a match between the supply and demand of the neurotransmitter [63,64,65]. Third, they promote Ca^{2+} entry into mitochondria at specialized junctions with the ER; mitochondrial Ca^{2+} entry stimulates oxidative phosphorylation (OXPHOS) and the production of Adenosine Triphosphate (ATP) [43,66,67]. In principle, this feed-forward control of OXPHOS helps to ensure that bioenergetic needs are met [68,69,70,71] and that intracellular ATP levels do not fall into a range that would trigger protective activation of K-ATP channels, which would cause cessation of on-going activity [72]. But this bioenergetic control system comes at a cost. First, stimulating OXPHOS in the absence of strong ATP demand (that is, most of the time) leads to mitochondrial hyperpolarization, retrograde electron flux through the electron transport chain and increased production of reactive oxygen species (ROS) and reactive nitrogen species (RNS) [29,66,73]. Although antioxidant defenses of mitochondria are substantial [74], they are imperfect [75]. ROS can damage proteins, lipids and DNA, particularly in mitochondria. Sustained mitochondrial oxidant stress could be a major factor underlying a decline in mitochondrial function in at-risk neurons in humans with age [76]. The progressive loss of mitochondrial function with time – as a consequence of accumulated oxidant damage – has been proposed to be a primary driver of aging-related decline in neuronal viability [77,78]. In either case, the ongoing mitochondrial oxidant stress in SNc DA neurons could lead to an increased rate of turnover of these organelles by mitophagy [74,79]. Aging-related decline in the ability to recycle effete mitochondria could lead to premature senescence and death of the cells. Morover an enhanced need to eliminate damaged mitochondria could compromise other proteostatic functions, including the degradation of misfolded proteins, like αSYN. If so, this might suggest that accumulation of αSYN is a consequence of mitochondrial damage in SNc cells. Another downside of a Ca^{2+}-mediated feed-forward system of bioenergetics supply is that it results a sustained elevation

in cytosolic Ca^{2+}. The bioenergetic expense of sequestering Ca^{2+} is substantial because pumping divalent Ca^{2+} against a steep electrochemical gradient (10^4 difference in concentration across the plasma membrane). requires active transport. These gradients are maintained by ATP-dependent pumps, whose sustained engagement creates a contuous bioenergetic burden for the cells [80,81]. Lastly, Ca^{2+} promotes αSYN aggregation [82,83,84,85], because the Ca^{2+}-dependent protease calpain cleaves off the carboxyl-terminus of αSYN, promoting aggregation [86]. This is further enhanced by oxidative stress [87] or DA [88]. In animal models, inhibiting calpain markedly reduces the neuronal toxicity of αSYN over-expression or mutation [89]. Interestingly, Ca^{2+} oscillations and incubation with αSYN fibrils creates a cytosolic oxidant stress that may combine with mitochondrial oxidant stress in SNc DA neurons [90]. To make matters worse, the Ca^{2+}-dependent protein phosphatase calcineurin promotes αSYN toxicity [22]. Thus, Ca^{2+} entry in SNc DA neurons is Janus faced; there is clearly an upside to it, but there also is a dark side.

A common, at-risk neuronal phenotype in PD?

Do other at-risk neurons in PD exhibit a similar physiological phenotype? Although the data are sparse, the tentative answer appears to be yes. Neurons in the LC, dorsal motor nucleus of the vagus (DMV) and PPN also exhibit pacemaking activity and cytosolic Ca^{2+} oscillations [43,91,92,93,94]. Although not autonomous pacemakers, neurons in Raphe nuclei (RN) and in the nucleus basalis of Meynert (NBM) spike continuously during the waking state [95,96]. LC and DMV have also been demonstrated to express Cav1.3 channels and to exhibit elevated mitochondria oxidative stress [43,94]. In contrast, VTA DA neurons, which are relatively spared in PD, are also autonomous pacemakers but have been shown to have modest Cav1 channel currents [66,97,98], robust cytosolic Ca^{2+} buffering [56,99] and low mitochondrial oxidant stress. Additional studies are needed to determine whether there are other physiological phenotypes that contribute to vulnerability in PD.

The extent to which the axonal phenotype of SNc DA neurons is shared by other at-risk neurons is not clear at this point because the experiments to answer this question are difficult. However, it appears that neurons in the DMV, gigantocellular reticular nucleus, dorsal and median RN, LC, LH, PPN, basal forebrain and ILT all have large, diffuse axonal projections to varying degrees, distinguishing them from the vast majority of sensory or motor neurons in the brain, which typically have spatially focused, modestly branched axons that conform to topographic maps [100,101,102,103,104,105,106,107]

Does the unusual use of Ca^{2+} by at-risk neurons cause PD?

The proposition that Ca^{2+} entry through Cav1 channels and Ca^{2+}-dependent feed-forward control of mitochondrial metabolism sets up SNc DA neurons (and other at-risk neurons) to be knocked over by other stressors linked to PD (i.e. aging, genetic mutations) is plausible but untested. Unfortunately, there are no genetic animal models of PD with predictive validity. That said, inhibiting Cav1 channels diminishes the damage induced by 6-hydroxydopamine and 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) when given at doses that produce partial loss of SNc DA neurons [38,108,109,110]. Inhibiting Cav1 channels also protects SNc DA neurons from low doses of the environmental toxin rotenone

[38]. The rodent chemical models of PD have generally proven disappointing, as they lack LP and manifest a pattern of pathology that does not resemble that in PD, even though there is cell loss in SNc.. A major obstacle in the field continues to be a lack of genetic models that recapitulate the phenotype seen in PD. Lacking such models, it has been difficult to obtain clear mechanistic insight into the factors regulating cell loss, and to test potential therapeutic interventions designed to modify the damage-inducing pathways. Recently, the use of stereotaxically injected, αSYN fibril fragments has gained popularity as a model of PD [13,111,112,113,114]. Fibrils injection into the striatum has been reported to produce degeneration in SNc DA neurons following retrograde transport. However, to date, there has been no attempt to determine whether inhibition of Cav1 channels blunts the toxicity of αSYN fibrils in vivo. What has recently been shown is that over-expression of αSYN increases oxidant stress in SNc DA neurons (see [90])and, in so doing, down-regulates the function of K^+ (Kv4) channels that slow the spiking rate [115]. Thus, the convergence of LP and mitochondrial stress could force SNc DA neurons into a ‘death spiral’, where growing oxidant stress elicits an increase in firing rate that further augments bioenergetic demand

Despite of the limitations of the animal models, there are several observations from humans that are consistent with the proposition that Cav1 channels and Ca^{2+}-stimulated mitochondrial oxidant stress are drivers of neurodegeneration and LP in PD. First, as mentioned above, the expression of the Ca^{2+} buffering protein calbindin is inversely correlated with risk of degeneration in the mesencephalon [54,116]. Second, in the human mesecephalon, there are clear signs of sustained mitochondrial oxidant stress. In the SNc from PD patients, mtDNA deletions (like those produce by ROS) are common [117,118,119,120] and Complex I activity is diminished [121,122].

But the most compelling evidence for this theory comes from epidemiological studies of patients being treated for hypertension. DHPs are FDA approved, negative allosteric modulators of Cav1 Ca^{2+} channels [123]. They are not channel blockers, in that they do not plug the channel pore. DHPs instead interact with a site buried in the lipid bilayer and impede channel activation. This interaction is dependent upon membrane potential; during membrane depolarization, DHPs bind efficiently, whereas with membrane hyperpolarization, they hardly bind at all. This is important because it makes DHPs effective inhibitors of Cav1 channels only in cells that are relatively depolarized. This explains why, in spite of widespread expression of Cav1 channels in the brain, use of brain penetrant DHPs have few, if any, short-term cognitive, sensory or motor effects in humans. For decades, DHPs have been used in humans to treat hypertension. They lower blood pressure primarily by inhibiting Cav1 channels in vascular smooth muscle, causing them to relax and dilate blood vessels. More importantly for PD, use of DHPs to treat hypertension has consistently been linked to a substantial reduction in the risk of being diagnosed with PD (~30% reduction in risk)[124,125,126,127,128]. This is similar to the risk reduction linked to smoking tobaccco [129,130,131]. This reduction in risk was not related to simply lowering blood pressure as treatment with beta-adrenergic blockers or acetylcholinesterase inhibitors was not associated with reduced risk, in spite of the fact both are typically more effective treatments for hypertension. Treatment with DHPs also is associated with slowing of disease progression even if initiated after diagnosis [132].

Although consistent with the hypothesis, these epidemiological studies are retrospective, and a definitive test requires a prospective, randomized, placebo-controlled study. Phase II tolerability trial in early stage PD patients was conducted and found that the DHP isradipine is well tolerated at doses below 10 mg/day [133,134]. Isradipine was chosen for this study because of its excellent pharmacokinetics and nearly equal affinity for Cav1.2 and Cav1.3 channels [48,135,136]. In 2013, NIH funded a 5 year, randomized, placebo-controlled, double blind, Phase III multicenter study to determine whether a 3 year regimen of isradipine treatment could slow the progression of PD in recently diagnosed patients. The study has enrolled over 300 patients and is slated for completion in 2019.

Conclusions and Challenges

The causes of PD in humans remain unresolved, although growing understanding of the common characteristics of neurons affected by the disease has led to hypothesis-driven studies to determine the significance of these in the pathogenesis. One such theory is that the pattern of pathology in PD is dictated by an unusual neuronal phenotype. This phenotype includes a long highly branched axon and an unusual pacemaking phenotype that engages Cav1 Ca^{2+} channels in a feed-forward control of mitochondrial OXPHOS, leading to sustained mitochondrial oxidant stress. This oxidant stress, over time, could lead to progressive loss of mitochondrial function that results in bioenergetic deficits. Oxidant stress also could increase proteostatic dysfunction by damaging cellular proteins and increasing mitophagic flux, broadly compromising the ability to deal with protein dysfunction. In addition, the elevation in cytosolic Ca^{2+} concentration accompanying this feedforward mechanism could promote aggregation of αSYN, a major component of LP. Like Humpty Dumpty who by sitting precariously on a high wall becomes susceptible to falling and breaking into pieces, this phenotype could poise SNc dopaminergic neurons, and other neurons with a similar phenotype, up for a ‘fall’ when pushed by other stressors that normally could be tolerated, including genetic mutations, environmental toxins or reactive neurotransmitter (i.e., DA).

This ‘Humpty Dumpty’ hypothesis is supported by observations in humans, most compellingly epidemiological data linking use of DHP inhibitors of Cav1 channels to decreased risk of developing PD and slowed progression. The ongoing NIH sponsored Phase III clinical trial with the DHP isradipine will be the first prospective test of this theory of pathogenesis. One limitation in this trial is the outcome measure: the unified Parkinson’s disease rating scale (UPDRS). The UPDRS is comprehensive but noisy, as it is somewhat subjective, so with 56 centers involved in this trial, with several clinicians at each site rating patients, there will be unavoidable experimental variability that could obscure a modest effect. However there are not any validated, objective biomarkers that would provide a more uniform assessment of disease progression. Another limitation is that the trial is restricted to patients who are symptomatic. Unfortunately, symptomatic presentation in PD appears relatively late in the disease process when a significant fraction of their SNc dopaminergic neurons are already gone. As we have learned from many clinical trials of disease modifying interventions, efficacy is often optimized when treatment is initiated at very early stages. This is not possible in PD, so the hope is that relief of the mitochondrial stresses will extend survival of the remaining neurons. If isradipine treatment were to significantly slow disease

progression, this could have a profoundly positive impact on the quality and duration of life of these patients, in addition to the obvious benefits in terms of limiting health care costs.

There are many unanswered questions and challenges related to the general question of selective vulnerability and pathogenesis in PD. Answering these questions could lead to new neuroprotective strategies. From our perspective, these include:

- Why is age the number one risk factor, and what factors that change with aging contribute to neurodegeneration in PD?
- *Are neurons at risk in PD preferentially sensitive to the genetic mutations that have been linked to PD?* For the recessive mutations affecting mitochondrial function the connection seems clear. But it is not as evident for the dominant mutations in SNCA, LRRK2 and GBA, for example.
- *Are the long, branched axons of SNc DA neurons responsible for increased vulnerability?* Recent work in vitro has shown that SNc DA axons have greater engagement of Cav1 channels [23] and elevated mitochondrial oxidant stress than VTA DA axons [29], but it isn't clear how they are related and whether DA is a factor.
- *To what extent do other neurons at-risk in PD have the same anatomical and physiological phenotype as SNc DA neurons?* This has been addressed for LC and DMV neurons but the vast majority of at-risk neurons have not been studied in a way that would inform theories of pathogenesis.
- *Does the at-risk phenotype promote αSYN aggregation or propagation of αSYN pathology? Does LP preferentially kill some types of neuron and, if so, how?* The two major hallmarks of PD are neurodegeneration and LP. Yet, it is unclear whether the latter is a marker or a mediator of the former. As outlined above, it is possible that Ca^{2+} handling in at-risk neurons promotes LP, but this needs to be tested rigorously

References

1. Fahn S. Description of Parkinson's disease as a clinical syndrome. Ann N Y Acad Sci. 2003; 991:1–14.
2. Lees AJ, Hardy J, Revesz T. Parkinson's disease. Lancet. 2009; 373:2055–2066. [PubMed: 19524782]
3. Sulzer D, Surmeier DJ. Neuronal vulnerability, pathogenesis, and Parkinson's disease. Mov Disord. 2013; 28:41–50. [PubMed: 22791686]
4. Goedert M, Spillantini MG, Del Tredici K, Braak H. 100 years of Lewy pathology. Nat Rev Neurol. 2013; 9:13–24. [PubMed: 23183883]
5. Bras JM, Singleton A. Genetic susceptibility in Parkinson's disease. Biochim Biophys Acta. 2009; 1792:597–603. [PubMed: 19063963]
6. Kumar KR, Djarmati-Westenberger A, Grunewald A. Genetics of Parkinson's disease. Semin Neurol. 2011; 31:433–440. [PubMed: 22266881]
7. Pickrell AM, Youle RJ. The Roles of PINK1, Parkin, and Mitochondrial Fidelity in Parkinson's Disease. Neuron. 2015; 85:257–273. [PubMed: 25611507]

8. Shin JH, Ko HS, Kang H, Lee Y, Lee YI, Pletinkova O, Troconso JC, Dawson VL, Dawson TM. PARIS (ZNF746) repression of PGC-1alpha contributes to neurodegeneration in Parkinson's disease. Cell. 2011; 144:689–702. [PubMed: 21376232]
9. Exner N, Lutz AK, Haass C, Winklhofer KF. Mitochondrial dysfunction in Parkinson's disease: molecular mechanisms and pathophysiological consequences. EMBO J. 2012; 31:3038–3062. [PubMed: 22735187]
10. Cookson MR. Parkinsonism due to mutations in PINK1, parkin, and DJ-1 and oxidative stress and mitochondrial pathways. Cold Spring Harb Perspect Med. 2012; 2:a009415. [PubMed: 22951446]
11. Desplats P, Lee HJ, Bae EJ, Patrick C, Rockenstein E, Crews L, Spencer B, Masliah E, Lee SJ. Inclusion formation and neuronal cell death through neuron-to-neuron transmission of alpha-synuclein. Proc Natl Acad Sci U S A. 2009; 106:13010–13015. [PubMed: 19651612]
12. Steiner JA, Angot E, Brundin P. A deadly spread: cellular mechanisms of alpha-synuclein transfer. Cell Death Differ. 2011; 18:1425–1433. [PubMed: 21566660]
13. Luk KC, Kehm V, Carroll J, zhang B, O'Brien P, Trojanowski JQ, Lee VM. Pathological a-Synuclein Transmission Initiates Parkinson-like Neurodegeneration in Nontransgenic Mice. Science. 2012; 338:949–953. [PubMed: 23161999]
14. Schapira AH. Aetiopathogenesis of Parkinson's disease. J Neurol. 2011; 258:S307–310. [PubMed: 21560060]
15. Mullin S, Schapira A. The genetics of Parkinson's disease. Br Med Bull. 2015; 114:39–52. [PubMed: 25995343]
16. Rodriguez M, Rodriguez-Sabate C, Morales I, Sanchez A, Sabate M. Parkinson's disease as a result of aging. Aging Cell. 2015; 14:293–308. [PubMed: 25677794]
17. Collier TJ, Kanaan NM, Kordower JH. Ageing as a primary risk factor for Parkinson's disease: evidence from studies of non-human primates. Nat Rev Neurosci. 2011; 12:359–366. [PubMed: 21587290]
18. Sulzer D. Multiple hit hypotheses for dopamine neuron loss in Parkinson's disease. Trends Neurosci. 2007; 30:244–250. [PubMed: 17418429]
19. Bisaglia M, Filograna R, Beltramini M, Bubacco L. Are dopamine derivatives implicated in the pathogenesis of Parkinson's disease? Ageing Res Rev. 2014; 13:107–114. [PubMed: 24389159]
20. Zucca FA, Segura-Aguilar J, Ferrari E, Muñoz P, Paris I, Sulzer D, Sarna T, Casella L, Zecca L. Interactions of iron, dopamine and neuromelanin pathways in brain aging and Parkinson's disease. Progress in Neurobiology. 2015
21. Mosharov EV, Larsen KE, Kanter E, Phillips KA, Wilson K, Schmitz Y, Krantz DE, Kobayashi K, Edwards RH, Sulzer D. Interplay between cytosolic dopamine, calcium, and alpha-synuclein causes selective death of substantia nigra neurons. Neuron. 2009; 62:218–229. [PubMed: 19409267]
22. Caraveo G, Auluck PK, Whitesell L, Chung CY, Baru V, Mosharov EV, Yan X, Ben-Johny M, Soste M, Picotti P, Kim H, Caldwell KA, Caldwell GA, Sulzer D, Yue DT, Lindquist S. Calcineurin determines toxic versus beneficial responses to α-synuclein. Proc Natl Acad Sci U S A. 2014; 111:E3544–3552. [PubMed: 25122673]
23. Brimblecombe KR, Gracie CJ, Platt NJ, Cragg SJ. Gating of dopamine transmission by calcium and axonal N-, Q-, T- and L-type voltage-gated calcium channels differs between striatal domains. J Physiol. 2015; 593:929–946. [PubMed: 25533038]
24. Kordower JH, Olanow CW, Dodiya HB, Chu Y, Beach TG, Adler CH, Halliday GM, Bartus RT. Disease duration and the integrity of the nigrostriatal system in Parkinson's disease. Brain. 2013; 136:2419–2431. [PubMed: 23884810]
25. Fahn S. Does levodopa slow or hasten the rate of progression of Parkinson's disease? J Neurol. 2005; 252(Suppl 4):IV37–IV42. [PubMed: 16222436]
26. Matsuda W, Furuta T, Nakamura KC, Hioki H, Fujiyama F, Arai R, Kaneko T. Single nigrostriatal dopaminergic neurons form widely spread and highly dense axonal arborizations in the neostriatum. J Neurosci. 2009; 29:444–453. [PubMed: 19144844]
27. Bolam JP, Pissadaki EK. Living on the edge with too many mouths to feed: why dopamine neurons die. Mov Disord. 2012; 27:1478–1483. [PubMed: 23008164]

28. Pissadaki EK, Bolam JP. The energy cost of action potential propagation in dopamine neurons: clues to susceptibility in Parkinson's disease. Front Comput Neurosci. 2013; 7:13. [PubMed: 23515615]
29. Pacelli C, Giguere N, Bourque MJ, Levesque M, Slack RS, Trudeau LE. Elevated Mitochondrial Bioenergetics and Axonal Arborization Size Are Key Contributors to the Vulnerability of Dopamine Neurons. Curr Biol. 2015; 25:2349–2360. [PubMed: 26320949]
30. Hunn BH, Cragg SJ, Bolam JP, Spillantini MG, Wade-Martins R. Impaired intracellular trafficking defines early Parkinson's disease. Trends Neurosci. 2015; 38:178–188. [PubMed: 25639775]
31. Venda LL, Cragg SJ, Buchman VL, Wade-Martins R. alpha-Synuclein and dopamine at the crossroads of Parkinson's disease. Trends Neurosci. 2010; 33:559–568. [PubMed: 20961626]
32. Zhou FM, Wilson CJ, Dani JA. Cholinergic interneuron characteristics and nicotinic properties in the striatum. J Neurobiol. 2002; 53:590–605. [PubMed: 12436423]
33. Grace AA, Bunney BS. Intracellular and extracellular electrophysiology of nigral dopaminergic neurons--1. Identification and characterization. Neuroscience. 1983; 10:301–315. [PubMed: 6633863]
34. Grace AA, Bunney BS. Intracellular and extracellular electrophysiology of nigral dopaminergic neurons--2. Action potential generating mechanisms and morphological correlates. Neuroscience. 1983; 10:317–331. [PubMed: 6633864]
35. Grace AA, Bunney BS. The control of firing pattern in nigral dopamine neurons: single spike firing. J Neurosci. 1984; 4:2866–2876. [PubMed: 6150070]
36. Wilson CJ, Callaway JC. Coupled oscillator model of the dopaminergic neuron of the substantia nigra. J Neurophysiol. 2000; 83:3084–3100. [PubMed: 10805703]
37. Neuhoff H, Neu A, Liss B, Roeper J. I(h) channels contribute to the different functional properties of identified dopaminergic subpopulations in the midbrain. J Neurosci. 2002; 22:1290–1302. [PubMed: 11850457]
38. Chan CS, Guzman JN, Ilijic E, Mercer JN, Rick C, Tkatch T, Meredith GE, Surmeier DJ. 'Rejuvenation' protects neurons in mouse models of Parkinson's disease. Nature. 2007; 447:1081–1086. [PubMed: 17558391]
39. Puopolo M, Raviola E, Bean BP. Roles of subthreshold calcium current and sodium current in spontaneous firing of mouse midbrain dopamine neurons. J Neurosci. 2007; 27:645–656. [PubMed: 17234596]
40. Bean BP. The action potential in mammalian central neurons. Nat Rev Neurosci. 2007; 8:451–465. [PubMed: 17514198]
41. Foehring RC, Zhang XF, Lee JC, Callaway JC. Endogenous calcium buffering capacity of substantia nigral dopamine neurons. J Neurophysiol. 2009; 102:2326–2333. [PubMed: 19675297]
42. Schwaller B. Cytosolic Ca2+ buffers. Cold Spring Harb Perspect Biol. 2010; 2:a004051. [PubMed: 20943758]
43. Sanchez-Padilla J, Guzman JN, Ilijic E, Kondapalli J, Galtieri DJ, Yang B, Schieber S, Oertel W, Wokosin D, Schumacker PT, Surmeier DJ. Mitochondrial oxidant stress in locus coeruleus is regulated by activity and nitric oxide synthase. Nat Neurosci. 2014; 17:832–840. [PubMed: 24816140]
44. Nedergaard S, Flatman JA, Engberg I. Nifedipine- and omega-conotoxin-sensitive Ca2+ conductances in guinea-pig substantia nigra pars compacta neurones. J Physiol. 1993; 466:727–747. [PubMed: 8410714]
45. Guzman JN, Sanchez-Padilla J, Chan CS, Surmeier DJ. Robust pacemaking in substantia nigra dopaminergic neurons. J Neurosci. 2009; 29:11011–11019. [PubMed: 19726659]
46. Morikawa H, Paladini CA. Dynamic regulation of midbrain dopamine neuron activity: intrinsic, synaptic, and plasticity mechanisms. Neuroscience. 2011; 198:95–111. [PubMed: 21872647]
47. Lipscombe D, Helton TD, Xu W. L-type calcium channels: the low down. J Neurophysiol. 2004; 92:2633–2641. [PubMed: 15486420]
48. Koschak A, Reimer D, Huber I, Grabner M, Glossmann H, Engel J, Striessnig J. alpha 1D (Cav1.3) subunits can form l-type Ca2+ channels activating at negative voltages. J Biol Chem. 2001; 276:22100–22106. [PubMed: 11285265]

49. Xu W, Lipscombe D. Neuronal Ca(V)1.3alpha(1) L-type channels activate at relatively hyperpolarized membrane potentials and are incompletely inhibited by dihydropyridines. J Neurosci. 2001; 21:5944–5951. [PubMed: 11487617]
50. Singh A, Gebhart M, Fritsch R, Sinnegger-Brauns MJ, Poggiani C, Hoda JC, Engel J, Romanin C, Striessnig J, Koschak A. Modulation of voltage- and Ca2+-dependent gating of CaV1.3 L-type calcium channels by alternative splicing of a C-terminal regulatory domain. J Biol Chem. 2008; 283:20733–20744. [PubMed: 18482979]
51. Bock G, Gebhart M, Scharinger A, Jangsangthong W, Busquet P, Poggiani C, Sartori S, Mangoni ME, Sinnegger-Brauns MJ, Herzig S, Striessnig J, Koschak A. Functional properties of a newly identified C-terminal splice variant of Cav1.3 L-type Ca2+ channels. J Biol Chem. 2011; 286:42736–42748. [PubMed: 21998310]
52. Tan BZ, Jiang F, Tan MY, Yu D, Huang H, Shen Y, Soong TW. Functional characterization of alternative splicing in the C terminus of L-type CaV1.3 channels. J Biol Chem. 2011; 286:42725–42735. [PubMed: 21998309]
53. Hage TA, Khaliq ZM. Tonic firing rate controls dendritic Ca2+ signaling and synaptic gain in substantia nigra dopamine neurons. J Neurosci. 2015; 35:5823–5836. [PubMed: 25855191]
54. German DCMK, Sonsalla PK, Brooks BA. Midbrain dopaminergic cell loss in Parkinson's disease and MPTP-induced parkinsonism: sparing of calbindin-D28k-containing cells. Ann N Y Acad Sci. 1992; 648:42–62. [PubMed: 1353337]
55. Yamada TMP, Baimbridge KG, McGeer EG. Relative sparing in Parkinson's disease of substantia nigra dopamine neurons containing calbindin-D28K. Brain Res. 1990; 526:303–307. [PubMed: 2257487]
56. Gaspar P, Ben Jelloun N, Febvret A. Sparing of the dopaminergic neurons containing calbindin-D28k and of the dopaminergic mesocortical projections in weaver mutant mice. Neuroscience. 1994; 61:293–305. [PubMed: 7969910]
57. Mercuri NB, Bonci A, Calabresi P, Stratta F, Stefani A, Bernardi G. Effects of dihydropyridine calcium antagonists on rat midbrain dopaminergic neurones. Br J Pharmacol. 1994; 113:831–838. [PubMed: 7858874]
58. Ping HX, Shepard PD. Apamin-sensitive Ca(2+)-activated K+ channels regulate pacemaker activity in nigral dopamine neurons. Neuroreport. 1996; 7:809–814. [PubMed: 8733751]
59. Bean BP. Nitrendipine block of cardiac calcium channels: high-affinity binding to the inactivated state. Proc Natl Acad Sci U S A. 1984; 81:6388–6392. [PubMed: 6093100]
60. Striessnig J, Koschak A, Sinnegger-Brauns MJ, Hetzenauer A, Nguyen NK, Busquet P, Pelster G, Singewald N. Role of voltage-gated L-type Ca2+ channel isoforms for brain function. Biochem Soc Trans. 2006; 34:903–909. [PubMed: 17052224]
61. Shepard PD, Stump D. Nifedipine blocks apamin-induced bursting activity in nigral dopamine-containing neurons. Brain Res. 1999; 817:104–109. [PubMed: 9889338]
62. Putzier I, Kullmann PH, Horn JP, Levitan ES. Cav1.3 channel voltage dependence, not Ca2+ selectivity, drives pacemaker activity and amplifies bursts in nigral dopamine neurons. J Neurosci. 2009; 29:15414–15419. [PubMed: 20007466]
63. Morgenroth, VHr, Boadle-Biber, M., Roth, RH. Tyrosine hydroxylase: activation by nerve stimulation. Proc Natl Acad Sci U S A. 1974; 71:4283–4287. [PubMed: 4155067]
64. Aumann T, Horne M. Activity-dependent regulation of the dopamine phenotype in substantia nigra neurons. J Neurochem. 2012; 121:497–515. [PubMed: 22356203]
65. Aumann TD, Egan K, Lim J, Boon WC, Bye CR, Chua HK, Baban N, Parish CL, Bobrovskaya L, Dickson P, Horne MK. Neuronal activity regulates expression of tyrosine hydroxylase in adult mouse substantia nigra pars compacta neurons. J Neurochem. 2011; 116:646–658. [PubMed: 21166807]
66. Guzman JN, Sanchez-Padilla J, Wokosin D, Kondapalli J, Ilijic E, Schumacker PT, Surmeier DJ. Oxidant stress evoked by pacemaking in dopaminergic neurons is attenuated by DJ-1. Nature. 2010; 468:696–700. [PubMed: 21068725]
67. Surmeier DJ, Schumacker PT. Calcium, bioenergetics, and neuronal vulnerability in Parkinson's disease. J Biol Chem. 2013; 288:10736–10741. [PubMed: 23086948]

68. Balaban RS. Domestication of the cardiac mitochondrion for energy conversion. J Mol Cell Cardiol. 2009; 46:832–841. [PubMed: 19265699]
69. Budd SL, Nicholls DG. Mitochondria in the life and death of neurons. Essays Biochem. 1998; 33:43–52. [PubMed: 10488440]
70. Nicholls DG, Budd SL. Neuronal excitotoxicity: the role of mitochondria. BioFactors. 1998; 8:287–299. [PubMed: 9914831]
71. Denton RM. Regulation of mitochondrial dehydrogenases by calcium ions. Biochim Biophys Acta. 2009; 1787:1309–1316. [PubMed: 19413950]
72. Dragicevic E, Schiemann J, Liss B. Dopamine midbrain neurons in health and Parkinson's disease: emerging roles of voltage-gated calcium channels and ATP-sensitive potassium channels. Neuroscience. 2015; 284:798–814. [PubMed: 25450964]
73. Votyakova TV, Reynolds IJ. DeltaPsim-Dependent and -independent production of reactive oxygen species by rat brain mitochondria. J Neurochem. 2001; 79:266–277. [PubMed: 11677254]
74. Scheibye-Knudsen M, Fang EF, Croteau DL, Wilson DM 3rd, Bohr VA. Protecting the mitochondrial powerhouse. Trends Cell Biol. 2014
75. Nicholls DG, Budd SL. Mitochondria and neuronal survival. Physiol Rev. 2000; 80:315–360. [PubMed: 10617771]
76. Reeve A, Simcox E, Turnbull D. Ageing and Parkinson's disease: why is advancing age the biggest risk factor? Ageing Res Rev. 2014; 14:19–30. [PubMed: 24503004]
77. Wallace DC. A mitochondrial paradigm of metabolic and degenerative diseases, aging, and cancer: a dawn for evolutionary medicine. Annu Rev Genet. 2005; 39:359–407. [PubMed: 16285865]
78. Raffaello A, Rizzuto R. Mitochondrial longevity pathways. Biochim Biophys Acta. 2011; 1813:260–268. [PubMed: 20950653]
79. Mammucari C, Rizzuto R. Signaling pathways in mitochondrial dysfunction and aging. Mech Ageing Dev. 2010; 131:536–543. [PubMed: 20655326]
80. Chan CS, Gertler TS, Surmeier DJ. Calcium homeostasis, selective vulnerability and Parkinson's disease. Trends Neurosci. 2009; 32:249–256. [PubMed: 19307031]
81. Berridge MJ, Lipp P, Bootman MD. The versatility and universality of calcium signalling. Nat Rev Mol Cell Biol. 2000; 1:11–21. [PubMed: 11413485]
82. Rcom-H'cheo-Gauthier A, Goodwin J, Pountney DL. Interactions between calcium and alpha-synuclein in neurodegeneration. Biomolecules. 2014; 4:795–811. [PubMed: 25256602]
83. Nielsen MS, Vorum H, Lindersson E, Jensen PH. Ca2+ binding to alpha-synuclein regulates ligand binding and oligomerization. J Biol Chem. 2001; 276:22680–22684. [PubMed: 11312271]
84. Nath S, Goodwin J, Engelborghs Y, Pountney DL. Raised calcium promotes alpha-synuclein aggregate formation. Mol Cell Neurosci. 2011; 46:516–526. [PubMed: 21145971]
85. Follett J, Darlow B, Wong MB, Goodwin J, Pountney DL. Potassium depolarization and raised calcium induces alpha-synuclein aggregates. Neurotox Res. 2013; 23:378–392. [PubMed: 23250862]
86. Dufty BM, Warner LR, Hou ST, Jiang SX, Gomez-Isla T, Leenhouts KM, Oxford JT, Feany MB, Masliah E, Rohn TT. Calpain-cleavage of alpha-synuclein: connecting proteolytic processing to disease-linked aggregation. Am J Pathol. 2007; 170:1725–1738. [PubMed: 17456777]
87. Goodwin J, Nath S, Engelborghs Y, Pountney DL. Raised calcium and oxidative stress cooperatively promote alpha-synuclein aggregate formation. Neurochem Int. 2013; 62:703–711. [PubMed: 23159813]
88. Jain MK, Bhat R. Modulation of human alpha-synuclein aggregation by a combined effect of calcium and dopamine. Neurobiol Dis. 2014; 63:115–128. [PubMed: 24269918]
89. Diepenbroek M, Casadei N, Esmer H, Saido TC, Takano J, Kahle PJ, Nixon RA, Rao MV, Melki R, Pieri L, Helling S, Marcus K, Krueger R, Masliah E, Riess O, Nuber S. Overexpression of the calpain-specific inhibitor calpastatin reduces human alpha-Synuclein processing, aggregation and synaptic impairment in [A30P]alphaSyn transgenic mice. Hum Mol Genet. 2014; 23:3975–3989. [PubMed: 24619358]
90. Dryanovski DI, Guzman JN, Xie Z, Galteri DJ, Volpicelli-Daley LA, Lee VM, Miller RJ, Schumacker PT, Surmeier DJ. Calcium entry and alpha-synuclein inclusions elevate dendritic

mitochondrial oxidant stress in dopaminergic neurons. J Neurosci. 2013; 33:10154–10164. [PubMed: 23761910]

91. Williams JT, North RA, Shefner SA, Nishi S, Egan TM. Membrane properties of rat locus coeruleus neurones. Neuroscience. 1984; 13:137–156. [PubMed: 6493483]
92. Mo ZL, Katafuchi T, Muratani H, Hori T. Effects of vasopressin and angiotensin II on neurones in the rat dorsal motor nucleus of the vagus, in vitro. J Physiol. 1992; 458:561–577. [PubMed: 1302279]
93. Takakusaki K, Kitai ST. Ionic mechanisms involved in the spontaneous firing of tegmental pedunculopontine nucleus neurons of the rat. Neuroscience. 1997; 78:771–794. [PubMed: 9153657]
94. Goldberg JA, Guzman JN, Estep CM, Ilijic E, Kondapalli J, Sanchez-Padilla J, Surmeier DJ. Calcium entry induces mitochondrial oxidant stress in vagal neurons at risk in Parkinson's disease. Nat Neurosci. 2012; 15:1414–1421. [PubMed: 22941107]
95. McGinty DJ, Harper RM. Dorsal raphe neurons: depression of firing during sleep in cats. Brain Res. 1976; 101:569–575. [PubMed: 1244990]
96. Hedrick T, Waters J. Physiological properties of cholinergic and non-cholinergic magnocellular neurons in acute slices from adult mouse nucleus basalis. PLoS One. 2010; 5:e11046. [PubMed: 20548784]
97. Khaliq ZM, Bean BP. Pacemaking in dopaminergic ventral tegmental area neurons: depolarizing drive from background and voltage-dependent sodium conductances. J Neurosci. 2010; 30:7401–7413. [PubMed: 20505107]
98. Liu Y, Harding M, Pittman A, Dore J, Striessnig J, Rajadhyaksha A, Chen X. Cav1.2 and Cav1.3 L-type calcium channels regulate dopaminergic firing activity in the mouse ventral tegmental area. J Neurophysiol. 2014; 112:1119–1130. [PubMed: 24848473]
99. Liang C-L, Sinton CM, German DC. Midbrain dopaminergic neurons in the mouse: co-localization with Calbindin-D28K and calretinin. Neuroscience. 1996; 75:523–533. [PubMed: 8931015]
100. Martinez-Gonzalez C, Bolam JP, Mena-Segovia J. Topographical organization of the pedunculopontine nucleus. Front Neuroanat. 2011; 5:22. [PubMed: 21503154]
101. Aston-Jones G, Waterhouse B. Locus coeruleus: From global projection system to adaptive regulation of behavior. Brain Res. 2016; 1645:75–78. [PubMed: 26969408]
102. Pfaff DW, Martin EM, Faber D. Origins of arousal: roles for medullary reticular neurons. Trends Neurosci. 2012; 35:468–476. [PubMed: 22626543]
103. Hornung J-P. The human raphe nuclei and the serotonergic system. Journal of Chemical Neuroanatomy. 2003; 26:331–343. [PubMed: 14729135]
104. Ratcliffe EM. Molecular development of the extrinsic sensory innervation of the gastrointestinal tract. Auton Neurosci. 2011; 161:1–5. [PubMed: 21147045]
105. Baufreton J, Kirkham E, Atherton JF, Menard A, Magill PJ, Bolam JP, Bevan MD. Sparse but selective and potent synaptic transmission from the globus pallidus to the subthalamic nucleus. J Neurophysiol. 2009; 102:532–545. [PubMed: 19458148]
106. Hu B, Yang N, Qiao QC, Hu ZA, Zhang J. Roles of the orexin system in central motor control. Neurosci Biobehav Rev. 2015; 49:43–54. [PubMed: 25511388]
107. Liu S, Borgland SL. Regulation of the mesolimbic dopamine circuit by feeding peptides. Neuroscience. 2015; 289:19–42. [PubMed: 25583635]
108. Ilijic E, Guzman JN, Surmeier DJ. The L-type channel antagonist isradipine is neuroprotective in a mouse model of Parkinson's disease. Neurobiol Dis. 2011; 43:364–371. [PubMed: 21515375]
109. Kupsch A, Gerlach M, Pupeter SC, Sautter J, Dirr A, Arnold G, Opitz W, Przuntek H, Riederer P, Oertel WH. Pretreatment with nimodipine prevents MPTP-induced neurotoxicity at the nigral, but not at the striatal level in mice. Neuroreport. 1995; 6:621–625. [PubMed: 7605913]
110. Kupsch A, Sautter J, Schwarz J, Riederer P, Gerlach M, Oertel WH. 1-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine-induced neurotoxicity in non-human primates is antagonized by pretreatment with nimodipine at the nigral, but not at the striatal level. Brain Res. 1996; 741:185–196. [PubMed: 9001722]

111. Luk KC, Kehm VM, Zhang B, O'Brien P, Trojanowski JQ, Lee VM. Intracerebral inoculation of pathological alpha-synuclein initiates a rapidly progressive neurodegenerative alpha-synucleinopathy in mice. J Exp Med. 2012; 209:975–986. [PubMed: 22508839]
112. Jones DR, Delenclos M, Baine AT, DeTure M, Murray ME, Dickson DW, McLean PJ. Transmission of Soluble and Insoluble α-Synuclein to Mice. J Neuropathol Exp Neurol. 2015; 74:1158–1169. [PubMed: 26574670]
113. Masuda-Suzukake M, Nonaka T, Hosokawa M, Oikawa T, Arai T, Akiyama H, Mann DM, Hasegawa M. Prion-like spreading of pathological alpha-synuclein in brain. Brain. 2013; 136:1128–1138. [PubMed: 23466394]
114. Paumier KL, Luk KC, Manfredsson FP, Kanaan NM, Lipton JW, Collier TJ, Steece-Collier K, Kemp CJ, Celano S, Schulz E, Sandoval IM, Fleming S, Dirr E, Polinski NK, Trojanowski JQ, Lee VM, Sortwell CE. Intrastriatal injection of pre-formed mouse alpha-synuclein fibrils into rats triggers alpha-synuclein pathology and bilateral nigrostriatal degeneration. Neurobiol Dis. 2015; 82:185–199. [PubMed: 26093169]
115. Subramaniam M, Althof D, Gispert S, Schwenk J, Auburger G, Kulik A, Fakler B, Roeper J. Mutant alpha-synuclein enhances firing frequencies in dopamine substantia nigra neurons by oxidative impairment of A-type potassium channels. J Neurosci. 2014; 34:13586–13599. [PubMed: 25297088]
116. Graybiel AM, Ohta K, Roffler-Tarlov S. Patterns of cell and fiber vulnerability in the mesostriatal system of the mutant mouse weaver. I. Gradients and compartments. J Neurosci. 1990; 10:720–733. [PubMed: 1690789]
117. Bender A, Krishnan KJ, Morris CM, Taylor GA, Reeve AK, Perry RH, Jaros E, Hersheson JS, Betts J, Klopstock T, Taylor RW, Turnbull DM. High levels of mitochondrial DNA deletions in substantia nigra neurons in aging and Parkinson disease. Nat Genet. 2006; 38:515–517. [PubMed: 16604074]
118. Kraytsberg Y, Kudryavtseva E, McKee AC, Geula C, Kowall NW, Khrapko K. Mitochondrial DNA deletions are abundant and cause functional impairment in aged human substantia nigra neurons. Nat Genet. 2006; 38:518–520. [PubMed: 16604072]
119. Muller SK, Bender A, Laub C, Hogen T, Schlaudraff F, Liss B, Klopstock T, Elstner M. Lewy body pathology is associated with mitochondrial DNA damage in Parkinson's disease. Neurobiol Aging. 2013; 34:2231–2233. [PubMed: 23566333]
120. Sanders LH, McCoy J, Hu X, Mastroberardino PG, Dickinson BC, Chang CJ, Chu CT, Van Houten B, Greenamyre JT. Mitochondrial DNA damage: molecular marker of vulnerable nigral neurons in Parkinson's disease. Neurobiol Dis. 2014; 70:214–223. [PubMed: 24981012]
121. Schapira AH, Cooper JM, Dexter D, Clark JB, Jenner P, Marsden CD. Mitochondrial complex I deficiency in Parkinson's disease. J Neurochem. 1990; 54:823–827. [PubMed: 2154550]
122. Mann VM, Cooper JM, Daniel SE, Srai K, Jenner P, Marsden CD, Schapira AH. Complex I, iron, and ferritin in Parkinson's disease substantia nigra. Ann Neurol. 1994; 36:876–881. [PubMed: 7998774]
123. Epstein BJ, Vogel K, Palmer BF. Dihydropyridine calcium channel antagonists in the management of hypertension. Drugs. 2007; 67:1309–1327. [PubMed: 17547473]
124. Becker C, Jick SS, Meier CR. Use of antihypertensives and the risk of Parkinson disease. Neurology. 2008; 70:1438–1444. [PubMed: 18256367]
125. Ritz B, Rhodes SL, Qian L, Schernhammer E, Olsen JH, Friis S. L-type calcium channel blockers and Parkinson disease in Denmark. Ann Neurol. 2010; 67:600–606. [PubMed: 20437557]
126. Pasternak B, Svanstrom H, Nielsen NM, Fugger L, Melbye M, Hviid A. Use of calcium channel blockers and Parkinson's disease. Am J Epidemiol. 2012; 175:627–635. [PubMed: 22387374]
127. Lee YC, Lin CH, Wu RM, Lin JW, Chang CH, Lai MS. Antihypertensive agents and risk of Parkinson's disease: a nationwide cohort study. PLoS One. 2014; 9:e98961. [PubMed: 24910980]
128. Gudala K, Kanukula R, Bansal D. Reduced Risk of Parkinson's Disease in Users of Calcium Channel Blockers: A Meta-Analysis. Int J Chronic Dis. 2015; 2015:697404. [PubMed: 26464872]

129. Chen H, Huang X, Guo X, Mailman RB, Park Y, Kamel F, Umbach DM, Xu Q, Hollenbeck A, Schatzkin A, Blair A. Smoking duration, intensity, and risk of Parkinson disease. Neurology. 2010; 74:878–884. [PubMed: 20220126]

130. Ritz B, Ascherio A, Checkoway H, Marder KS, Nelson LM, Rocca WA, Ross GW, Strickland D, Van Den Eeden SK, Gorell J. Pooled analysis of tobacco use and risk of Parkinson disease. Arch Neurol. 2007; 64:990–997. [PubMed: 17620489]

131. Li X, Li W, Liu G, Shen X, Tang Y. Association between cigarette smoking and Parkinson's disease: A meta-analysis. Arch Gerontol Geriatr. 2015; 61:510–516. [PubMed: 26272284]

132. Marras C, Gruneir A, Rochon P, Wang X, Anderson G, Brotchie J, Bell CM, Fox S, Austin PC. Dihydropyridine calcium channel blockers and the progression of parkinsonism. Ann Neurol. 2012; 71:362–369. [PubMed: 22451203]

133. Simuni T, Borushko E, Avram MJ, Miskevics S, Martel A, Zadikoff C, Videnovic A, Weaver FM, Williams K, Surmeier DJ. Tolerability of isradipine in early Parkinson's disease: a pilot dose escalation study. Mov Disord. 2010; 25:2863–2866. [PubMed: 20818667]

134. Group PS. Phase II safety, tolerability, and dose selection study of isradipine as a potential disease-modifying intervention in early Parkinson's disease (STEADY-PD). Mov Disord. 2013; 28:1823–1831. [PubMed: 24123224]

135. Fitton A, Benfield P. Isradipine. A review of its pharmacodynamic and pharmacokinetic properties, and therapeutic use in cardiovascular disease. Drugs. 1990; 40:31–74.

136. Scholze A, Plant TD, Dolphin AC, Nürnberg B. Functional expression and characterization of a voltage-gated CaV1.3 (alpha1D) calcium channel subunit from an insulin-secreting cell line. Mol Endocrinol. 2001; 15:1211–2121. [PubMed: 11435619]

Highlights

- Parkinson's disease (PD) is the second most common neurodegenerative disease
- Core PD motor symptoms are caused by the death of pacemaking dopaminergic neurons
- These neurons use Cav1 Ca^{2+} channels to support pacemaking and bioenergetics
- Resulting high cytosolic Ca^{2+} and mitochondrial stress could drive degeneration
- Inhibitors of Cav1 channels may decrease PD progression