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LRRKing up the right trees? On figuring out the effects of mutant LRRK2 and other Parkinson's disease-related genes

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It has been 10 years and more since associations between specific genes and Parkinson's disease (PD) were discovered, and it is now assumed that mutations in such PD (risk) genes, probably in interaction with other factors, are a major cause for PD [1–4]. These PD risk genes include alpha-synuclein (SNCA), LRRK2, Parkin, PINK1 and others. Yet after a decade of intense research it is still unclear how most mutations in these genes contribute to the PD pathology. This is likely due to a number of reasons, including that some of these genes seem to encode complex molecules with multiple functions; that mutations may lead to toxic gain-of-function and/or loss-of-function defects; that mutated molecules may need to interact with one another or other influences to be effective; or that some of these molecules or their products may need to migrate from other brain structures or even from the periphery to the dopamine neurons that they are supposed to kill. These factors all complicate the analysis of the mechanisms of action of PD risk genes.

LRRK2

Perhaps most is known about the potential mechanisms of action of LRRK2 (e.g., [3,5–11]), which can serve as an example to highlight the many complexities encountered in the search of a function. With the first mutation discovered a decade ago in familial cases of PD [12–14], many mutations have since been described in the LRRK2 gene and several are considered pathogenic [3,5,6,9]. These are missense mutations. The most prevalent of these mutations, G2019S, was found in 1–2% of sporadic PD cases (in a Caucasian population) [15], but in up to 37% of familial PD cases in specific ethnic groups (Ashkenazi Jews, North African Arabs; c.f. [5]).

The LRRK2 gene has 51 exons and encodes a large (286 kDa) protein with several predicted functional domains. These include a MAPKKK-like kinase domain and a ROC GTPase domain, as well as COR, leucine-rich repeat (LRR), ankyrin, armadillo and WD40 domains [5–7]. Wildtype LRRK2 binds/affects a variety of proteins, including Parkin, HSP90, moesin, tubulin, as well as presynaptic proteins involved in vesicle trafficking such as NSF, syntaxin 1, actin and others [5,7,16]. The mutations occur throughout LRRK2 [5,6]. Several mutations, including G2019S, are in the kinase domain [5,7,17]; G2019S, for example, abnormally increases kinase activity in vitro [5,7] and in mouse models [17].

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Models

To study the function of the wildtype protein or one of these mutations, researchers have used a variety of approaches and expression systems, including targeted deletion (knockout), or overexpression of wildtype or mutant LRRK2 in bacterial systems, cell lines (e.g., HEK-293, Cos-7, SH-SY5Y), primary neuron cultures or invertebrate models (*C. elegans*, *Drosophila*) [3,5,8,9,11], as well as in mouse lines [3,5,18]. Hypotheses as to what could be wrong in these mutants are typically derived from the predicted protein functions (e.g., kinase, GTPase), or from what is now known to be amiss in PD (e.g., loss of dopamine neurons, mitochondrial vulnerability, inflammation, etc). These studies yielded a plethora of findings on potential LRRK2 (mal)functions, including effects on synaptic vesicle recycling, neurite morphology/outgrowth, autophagy, pro-inflammatory factors, susceptibility to oxidative stress, cell death via accumulation of alpha-synuclein, and others [5–9,17,19].

Yet it is not clear whether or to what extent these mutations make dopamine neurons sick in mammals. For one, in PD their association remains correlative. In animal models, degeneration of dopamine neurons after deletion of LRRK2 (e.g., [20]) or overexpression of LRRK2 or LRRK2-G2019S [21] was reported in some (but not all) *Drosophila* and other invertebrate models. However, transgenic mice display surprisingly subtle or no effects on the dopamine transmission [3,5,11,18]. Indeed, a lack of significant degeneration of dopamine neurons seems to be a common feature of most mouse models of PD risk genes [3].

For LRRK2, mouse lines with a variety of constructs (knock-out, knock-in, BAC) have been developed [3,5,11,18]. These include lines that lack the LRRK2 protein (whole or part), or overexpress human wildtype LRRK2, the LRRK2-G2019S mutation or a LRRK2 kinase-dead mutation. In some cases, neither of these lines showed changes in markers of dopamine neuron health, dopamine levels or dopaminergic drug-evoked behaviors (while displaying pronounced histopathological changes in the kidney and lung where LRRK2 is also highly expressed) [22,23]. In other cases, such transgenics did display modest alterations in dopamine function/release and/or related behaviors, but, with one exception [24], none showed dopamine neuron degeneration [25–31].

In contrast to these transgenic models, substantial degeneration of dopamine neurons could be achieved by virally-driven overexpression of pathogenic LRRK2 (G2019S) in dopamine neurons of rodents [32,33]. However, while providing a valuable tool for screening for neuroprotective drugs [17], it is not known whether these viral-based models mimic natural G2019S expression (i.e., whether overexpression is crucial for the pathogenicity in PD). Importantly, further work in these models showed that it is the kinase overactivity of the G2019S mutant that is detrimental ([32]; see also [7,17]), rather than the overexpression of this protein per se, which is a concern with overexpression models (i.e., there is the danger that overexpression of molecules nonspecifically hampers normal cellular functions - think of your car filled with tennis balls).

It is unclear why the transgenic approaches so far do not reproduce PD pathology. It is possible that manipulating individual functional domains or single genes is not sufficient. For example, other studies indicate that interactions between two (e.g., LRRK2 and Parkin [34]; LRRK2 and alpha-synuclein [27]; LRRK2 and RAB7L1 (PARK16 locus) [35]) or more [1,3] of the PD risk genes may be essential. Also, of course, PD is a late adult neurodegenerative disorder and, with the exception of the rodent models, the above discussed approaches can not recreate normal mammalian aging. Moreover, while some aging-related degeneration was seen in one mouse model [24], mice may also not live long enough to develop full PD pathology [3].

Anatomical Mismatch

There may be more to the puzzle. Surprisingly, LRRK2 mRNA is robustly expressed in several areas of the brain, but not in the substantia nigra dopamine neurons that are supposed to be affected by the mutations. In situ hybridization studies consistently showed that LRRK2 expression in dopamine neurons is low or below the threshold of detection [36–41] (Fig. 1). This is true for mice and rats (e.g., [36–38,40]), but also for humans [36,41]. While more sensitive RT-PCR studies did find some mRNA in the midbrain dopamine cell body regions, these levels were minimal compared to those in other brain areas [37,38].

Interestingly, in contrast, it is the main dopamine target areas, such as the striatum, that express the highest levels of LRRK2 mRNA, by far higher than the midbrain [36–43] (Fig. 1). In the striatum, LRRK2 is expressed in projection neurons and some interneurons [40,44]. High levels of LRRK2 mRNA expression are also present in the cortex (Fig. 1), cerebellum, hippocampus and several other brain regions [36–40,42,43]. [It should be noted that other PD risk genes, including alpha-synuclein (SNCA) [37], PINK1 [38] and PJ-1 [36], are also widely expressed throughout the brain, but robust expression for these is also present in dopamine neurons (e.g., [36–38]).]

The fact that LRRK2 is predominantly expressed in dopamine-receptive neurons such as those in the striatum let some investigators (e.g., [37,45]) propose a “retrograde disease mechanism”, in which aberrant LRRK2 activity in striatal neurons would affect nigrostriatal dopamine neurons retrogradely (e.g., via diminished trophic support) and lead to degeneration of these neurons. Alternatively, striatal LRRK2 products could conceivably be transported to the midbrain dopamine neurons anterogradely in striatonigral projection neurons. There is evidence for both mechanisms in the central nervous system. For example, the trophic factor BDNF is known to be synthesized in cortical neurons and in the midbrain dopamine neurons and then transported anterogradely to the striatum [46,47]. Conversely, retrograde signaling is important, for example, in the cerebellum for the survival of granule cells; these cells are dependent on factors expressed by their projection targets, the Purkinje cells [48]. The recent finding of a decrease in number of LRRK2-positive neurons in the striatum of PD patients ([45], but see [36]) would be consistent with a role for striatal LRRK2 in PD via either mechanism. However, with direct evidence for striatal LRRK2 products affecting dopamine neurons lacking, these proposals have yet to play a role in conceptualizations of LRRK2 action.

Interneuronal transfer of PD risk gene products has in the meantime been demonstrated for alpha-synuclein [49]. For example, early studies showed that host alpha-synuclein can infiltrate dopamine neurons grafted into the striatum and other cells [50–52]. It is now accepted that alpha-synuclein (Lewy) pathology distributes through the central nervous system (and may originate in the periphery), seeded by transiting alpha-synuclein [49]. Supporting this theory, recent work found that a single injection of alpha-synuclein into the striatum resulted in spread to the substantia nigra and then produced progressive dopamine neuron degeneration [53]. (It is also noteworthy that the dopamine cell loss induced by virally-driven overexpression of G2019S discussed above was achieved (and indeed worked best) with intrastriatal infusion of the viral construct [32,33]. The construct was expressed in striatal neurons and was transported to the dopamine neurons and expressed in these [32,33]. It is assumed that the latter process produced the damage.) Could it be that LRRK2 products similarly originate in dopamine-receptive neurons and then transfer to the dopamine neurons?

Such an effect could explain the puzzling finding of significant levels of LRRK2 protein in dopamine neurons, despite the scarcity of mRNA expression in these neurons (see above).

Thus, immunohistochemical studies in rodents, monkeys and humans showed that, in addition to high LRRK2 protein levels in the striatum, cortex, hippocampus and other areas, robust LRRK2 immunoreactivity was also present in midbrain dopamine neurons [40–42,44,54]. This mismatch between LRRK2 protein presence and LRRK2 gene expression is not unique for dopamine neurons. A similar mismatch between mRNA levels (low) and protein levels (high) was seen in thalamic nuclei [37,40,42,44] and in the external globus pallidus. For the latter nucleus, studies in monkeys [44] and mice [40] showed robust LRRK2 immunoreactivity in neuropil and perikarya, with little or no mRNA expression present (in rodents; [38–40]) (Fig. 1). Notably, for the striatum, the study in monkeys also localized significant LRRK2 immunoreactivity in glutamatergic afferent terminals [44], thus demonstrating either LRRK2 anterograde axonal transport (from cortex or thalamus) or interneuronal transfer (uptake) after striatal synthesis.

Future studies will have to determine whether the striatum (and other dopamine-receptive neurons) could be the source for LRRK2 products found in dopamine neurons. If this is indeed the case, then overexpression of LRRK2/mutations in dopamine neurons may not recreate the situation in PD pathology.

Why Dopamine Neurons?

Given the widespread expression of LRRK2 (and other PD risk genes) in the brain, it remains unexplained as to why dopamine neurons are more selectively affected in PD, if then mutant proteins such as LRRK2 are causally involved in their demise. Recent findings indicate that one contributor to the increased vulnerability of dopamine neurons compared with other neurons (and for substantia nigra dopamine neurons over ventral tegmental dopamine neurons, consistent with the PD pathology) is their physiological phenotype (e.g., firing characteristics, calcium management, etc.) and related increased metabolic demands/oxidative stress [55].

But there is likely more. Perhaps related to their oxidative risks, dopamine neurons indeed seem to have a life-long need for trophic support. Thus, it is well known that adult dopamine neurons are among a select group of neuron types that are sensitive to various neurotrophic factors [56] and feature receptors for several of these factors, including GDNF (Ret), BDNF, EGF (ErbB1), neuregulin (ErbB4), and NT-3 (c.f. [57]), indicating that under normal circumstances these neurons remain dependent on trophic molecules. There are also several examples for interactions between PD risk genes and trophic factors. Thus, recent findings indicate that Nurr1-dependent GDNF signaling in dopamine neurons can attenuate alpha-synuclein toxicity [58]. Moreover, certain BDNF variants were found to greatly increased the LRRK2-associated PD risk [59].

It thus appears that a complex mix of factors, including PD risk mutations, neuronal phenotype, trophic support and other/peripheral influences (e.g., environmental toxins, immune responses [2–4,19,60]) interact to determine the health of dopamine neurons.

Conclusions

Much has been learned on the potential effects of PD risk mutations over the last decade. Cellular expression systems for such mutations are indispensable to identify their impact on molecular signaling involved in cellular health, but these systems can not model aspects such as mammalian aging and neuronal networks that may also play a role. While possibly also too “short-term” for developing full PD pathology, present mouse models may still provide valuable insights into early PD disease processes. Nevertheless, improved mammalian models that mimic the natural (i.e., anatomically correct) expression patterns of

these mutations are needed to investigate the complex interactions at the neurosystems level that are likely also involved in the function of PD-associated mutations.

References

1. Abeliovich A, Flint Beal M. Parkinsonism genes: culprits and clues. *J Neurochem.* 2006; 99:1062–72. [PubMed: 16836655]
2. Meredith GE, Sonsalla PK, Chesselet MF. Animal models of Parkinson's disease progression. *Acta Neuropathol.* 2008; 115:385–98. [PubMed: 18273623]
3. Dawson TM, Ko HS, Dawson VL. Genetic animal models of Parkinson's disease. *Neuron.* 2010; 66:646–61. [PubMed: 20547124]
4. Meredith, GE.; Totterdell, S. Parkinson's disease: Cross-talk between environmental factors and gene defects. In: Steiner, H.; Tseng, KY., editors. *Handbook of Basal Ganglia Structure and Function.* London: Academic Press/Elsevier; 2010. p. 593-606.
5. Melrose H. Update on the functional biology of Lrrk2. *Future Neurol.* 2008; 3:669–81. [PubMed: 19225574]
6. Gandhi PN, Chen SG, Wilson-Delfosse AL. Leucine-rich repeat kinase 2 (LRRK2): a key player in the pathogenesis of Parkinson's disease. *J Neurosci Res.* 2009; 87:1283–95. [PubMed: 19025767]
7. Greggio E, Cookson MR. Leucine-rich repeat kinase 2 mutations and Parkinson's disease: three questions. *ASN Neuro.* 2009; 1:13–24.
8. Yue Z. LRRK2 in Parkinson's disease: in vivo models and approaches for understanding pathogenic roles. *FEBS J.* 2009; 276:6445–54. [PubMed: 19804414]
9. Drolet RE, Sanders JM, Kern JT. Leucine-rich repeat kinase 2 (LRRK2) cellular biology: a review of recent advances in identifying physiological substrates and cellular functions. *J Neurogenet.* 2011; 25:140–51. [PubMed: 22077787]
10. Dächsel JC, Farrer MJ. LRRK2 and Parkinson disease. *Arch Neurol.* 2010; 67:542–7. [PubMed: 20457952]
11. Tsika E, Moore DJ. Mechanisms of LRRK2-mediated neurodegeneration. *Curr Neurol Neurosci Rep.* 2012; 12:251–60. [PubMed: 22441981]
12. Funayama M, Hasegawa K, Kowa H, Saito M, Tsuji S, Obata F. A new locus for Parkinson's disease (PARK8) maps to chromosome 12p11.2-q13. 1. *Ann Neurol.* 2002; 51:296–301. [PubMed: 11891824]
13. Paisán-Ruiz C, Jain S, Evans EW, Gilks WP, Simón J, van der Brug M, López de Munain A, Aparicio S, Gil AM, Khan N, Johnson J, Martinez JR, Nicholl D, Carrera IM, Pena AS, de Silva R, Lees A, Martí-Massó JF, Pérez-Tur J, Wood NW, Singleton AB. Cloning of the gene containing mutations that cause PARK8-linked Parkinson's disease. *Neuron.* 2004; 44:595–600. [PubMed: 15541308]
14. Zimprich A, Biskup S, Leitner P, Lichtner P, Farrer M, Lincoln S, Kachergus J, Hulihan M, Uitti RJ, Calne DB, Stoessl AJ, Pfeiffer RF, Patenge N, Carbajal IC, Vieregge P, Asmus F, Müller-Miyhok B, Dickson DW, Meitinger T, Strom TM, Wszolek ZK, Gasser T. Mutations in LRRK2 cause autosomal-dominant parkinsonism with pleomorphic pathology. *Neuron.* 2004; 44:601–7. [PubMed: 15541309]
15. Gilks WP, Abou-Sleiman PM, Gandhi S, Jain S, Singleton A, Lees AJ, Shaw K, Bhatia KP, Bonifati V, Quinn NP, Lynch J, Healy DG, Holton JL, Revesz T, Wood NW. A common LRRK2 mutation in idiopathic Parkinson's disease. *Lancet.* 2005; 365:415–6. [PubMed: 15680457]
16. Belluzzi E, Greggio E, Piccoli G. Presynaptic dysfunction in Parkinson's disease: a focus on LRRK2. *Biochem Soc Trans.* 2012; 40:1111–6. [PubMed: 22988874]
17. Lee BD, Dawson VL, Dawson TM. Leucine-rich repeat kinase 2 (LRRK2) as a potential therapeutic target in Parkinson's disease. *Trends Pharmacol Sci.* 2012; 33:365–73. [PubMed: 22578536]
18. Yue Z. Genetic mouse models for understanding LRRK2 biology, pathology and pre-clinical application. *Parkinsonism Relat Disord.* 2012; 18S1:S180–2. [PubMed: 22166428]
19. Greggio E, Civiero L, Bisaglia M, Bubacco L. Parkinson's disease and immune system: is the culprit LRRK2 in the periphery? *J Neuroinflammation.* 2012; 9:94. [PubMed: 22594666]

20. Lee SB, Kim W, Lee S, Chung J. Loss of LRRK2/PARK8 induces degeneration of dopaminergic neurons in *Drosophila*. *Biochem Biophys Res Commun*. 2007; 358:534–9. [PubMed: 17498648]
21. Liu Z, Wang X, Yu Y, Li X, Wang T, Jiang H, Ren Q, Jiao Y, Sawa A, Moran T, Ross CA, Montell C, Smith WW. A *Drosophila* model for LRRK2-linked parkinsonism. *Proc Natl Acad Sci USA*. 2008; 105:2693–8. [PubMed: 18258746]
22. Herzig MC, Kolly C, Persohn E, Theil D, Schweizer T, Hafner T, Stemmelen C, Troxler TJ, Schmid P, Danner S, Schnell CR, Mueller M, Kinzel B, Grevot A, Bolognani F, Stirn M, Kuhn RR, Kaupmann K, van der Putten PH, Rovelli G, Shimshek DR. LRRK2 protein levels are determined by kinase function and are crucial for kidney and lung homeostasis in mice. *Hum Mol Genet*. 2011; 20:4209–23. [PubMed: 21828077]
23. Herzig MC, Bidinosti M, Schweizer T, Hafner T, Stemmelen C, Weiss A, Danner S, Vidotto N, Stauffer D, Barske C, Mayer F, Schmid P, Rovelli G, van der Putten PH, Shimshek DR. High LRRK2 levels fail to induce or exacerbate neuronal alpha-synucleinopathy in mouse brain. *PLoS One*. 2012; 7:e36581. [PubMed: 22615783]
24. Ramonet D, Daher JP, Lin BM, Stafa K, Kim J, Banerjee R, Westerlund M, Pletnikova O, Glauser L, Yang L, Liu Y, Swing DA, Beal MF, Troncoso JC, McCaffery JM, Jenkins NA, Copeland NG, Galter D, Thomas B, Lee MK, Dawson TM, Dawson VL, Moore DJ. Dopaminergic neuronal loss, reduced neurite complexity and autophagic abnormalities in transgenic mice expressing G2019S mutant LRRK2. *PLoS One*. 2011; 6:e18568. [PubMed: 21494637]
25. Wang L, Xie C, Greggio E, Parisiadou L, Shim H, Sun L, Chandran J, Lin X, Lai C, Yang WJ, Moore DJ, Dawson TM, Dawson VL, Chiosis G, Cookson MR, Cai H. The chaperone activity of heat shock protein 90 is critical for maintaining the stability of leucine-rich repeat kinase 2. *J Neurosci*. 2008; 28:3384–91. [PubMed: 18367605]
26. Li Y, Liu W, Oo TF, Wang L, Tang Y, Jackson-Lewis V, Zhou C, Geghman K, Bogdanov M, Przedborski S, Beal MF, Burke RE, Li C. Mutant LRRK2(R1441G) BAC transgenic mice recapitulate cardinal features of Parkinson's disease. *Nat Neurosci*. 2009; 12:826–8. [PubMed: 19503083]
27. Lin X, Parisiadou L, Gu XL, Wang L, Shim H, Sun L, Xie C, Long CX, Yang WJ, Ding J, Chen ZZ, Gallant PE, Tao-Cheng JH, Rudow G, Troncoso JC, Liu Z, Li Z, Cai H. Leucine-rich repeat kinase 2 regulates the progression of neuropathology induced by Parkinson's-disease-related mutant alpha-synuclein. *Neuron*. 2009; 64:807–27. [PubMed: 20064389]
28. Tong Y, Pisani A, Martella G, Karouani M, Yamaguchi H, Pothos EN, Shen J. R1441C mutation in LRRK2 impairs dopaminergic neurotransmission in mice. *Proc Natl Acad Sci USA*. 2009; 106:14622–7. [PubMed: 19667187]
29. Li X, Patel JC, Wang J, Avshalumov MV, Nicholson C, Buxbaum JD, Elder GA, Rice ME, Yue Z. Enhanced striatal dopamine transmission and motor performance with LRRK2 overexpression in mice is eliminated by familial Parkinson's disease mutation G2019S. *J Neurosci*. 2010; 30:1788–97. [PubMed: 20130188]
30. Melrose HL, Dächsel JC, Behrouz B, Lincoln SJ, Yue M, Hinkle KM, Kent CB, Korvatska E, Taylor JP, Witten L, Liang YQ, Beevers JE, Boules M, Dugger BN, Serna VA, Gaukhman A, Yu X, Castaneda-Casey M, Braithwaite AT, Ogholikhan S, Yu N, Bass D, Tyndall G, Schellenberg GD, Dickson DW, Janus C, Farrer MJ. Impaired dopaminergic neurotransmission and microtubule-associated protein tau alterations in human LRRK2 transgenic mice. *Neurobiol Dis*. 2010; 40:503–17. [PubMed: 20659558]
31. Hinkle KM, Yue M, Behrouz B, Dächsel JC, Lincoln SJ, Bowles EE, Beevers JE, Dugger B, Winner B, Prots I, Kent CB, Nishioka K, Lin WL, Dickson DW, Janus CJ, Farrer MJ, Melrose HL. LRRK2 knockout mice have an intact dopaminergic system but display alterations in exploratory and motor co-ordination behaviors. *Mol Neurodegener*. 2012; 7:25. [PubMed: 22647713]
32. Lee BD, Shin JH, VanKampen J, Petrucelli L, West AB, Ko HS, Lee YI, Maguire-Zeiss KA, Bowers WJ, Federoff HJ, Dawson VL, Dawson TM. Inhibitors of leucine-rich repeat kinase-2 protect against models of Parkinson's disease. *Nat Med*. 2010; 16:998–1000. [PubMed: 20729864]
33. Dusanochet J, Kochubey O, Stafa K, Young SM, Zufferey R, Moore DJ, Schneider BL, Aebischer P. A rat model of progressive nigral neurodegeneration induced by the Parkinson's disease-associated G2019S mutation in LRRK2. *J Neurosci*. 2011; 31:907–12. [PubMed: 21248115]

34. Smith WW, Pei Z, Jiang H, Moore DJ, Liang Y, West AB, Dawson VL, Dawson TM, Ross CA. Leucine-rich repeat kinase 2 (LRRK2) interacts with parkin, and mutant LRRK2 induces neuronal degeneration. *Proc Natl Acad Sci USA*. 2005; 102:18676–81. [PubMed: 16352719]
35. MacLeod DA, Rhinn H, Kuwahara T, Zolin A, Di Paolo G, McCabe BD, Marder KS, Honig LS, Clark LN, Small SA, Abeliovich A. RAB7L1 interacts with LRRK2 to modify intraneuronal protein sorting and Parkinson's disease risk. *Neuron*. 2013; 77:425–39. [PubMed: 23395371]
36. Galter D, Westerlund M, Carmine A, Lindqvist E, Sydow O, Olson L. LRRK2 expression linked to dopamine-innervated areas. *Ann Neurol*. 2006; 59:714–9. [PubMed: 16532471]
37. Melrose H, Lincoln S, Tyndall G, Dickson D, Farrer M. Anatomical localization of leucine-rich repeat kinase 2 in mouse brain. *Neuroscience*. 2006; 139:791–4. [PubMed: 16504409]
38. Taymans JM, Van den Haute C, Baekelandt V. Distribution of PINK1 and LRRK2 in rat and mouse brain. *J Neurochem*. 2006; 98:951–61. [PubMed: 16771836]
39. Simón-Sánchez J, Herranz-Pérez V, Olucha-Bordonau F, Pérez-Tur J. LRRK2 is expressed in areas affected by Parkinson's disease in the adult mouse brain. *Eur J Neurosci*. 2006; 23:659–66. [PubMed: 16487147]
40. Higashi S, Moore DJ, Colebrooke RE, Biskup S, Dawson VL, Arai H, Dawson TM, Emson PC. Expression and localization of Parkinson's disease-associated leucine-rich repeat kinase 2 in the mouse brain. *J Neurochem*. 2007; 100:368–81. [PubMed: 17101029]
41. Higashi S, Biskup S, West AB, Trinkaus D, Dawson VL, Faull RL, Waldevogel HJ, Arai H, Dawson TM, Moore DJ, Emson PC. Localization of Parkinson's disease-associated LRRK2 in normal and pathological human brain. *Brain Res*. 2007; 1155:208–19. [PubMed: 17512502]
42. Melrose HL, Kent CB, Taylor JP, Dachsel JC, Hinkle KM, Lincoln SJ, Mok SS, Culvenor JG, Masters CL, Tyndall GM, Bass DI, Ahmed Z, Andorfer CA, Ross OA, Wszolek ZK, Delldonne A, Dickson DW, Farrer MJ. A comparative analysis of leucine-rich repeat kinase 2 (Lrrk2) expression in mouse brain and Lewy body disease. *Neuroscience*. 2007; 147:1047–58. [PubMed: 17611037]
43. Westerlund M, Belin AC, Anvret A, Bickford P, Olson L, Galter D. Developmental regulation of leucine-rich repeat kinase 1 and 2 expression in the brain and other rodent and human organs: Implications for Parkinson's disease. *Neuroscience*. 2008; 152:429–36. [PubMed: 18272292]
44. Lee H, Melrose HL, Yue M, Pare JF, Farrer MJ, Smith Y. Lrrk2 localization in the primate basal ganglia and thalamus: a light and electron microscopic analysis in monkeys. *Exp Neurol*. 2010; 224:438–47. [PubMed: 20483355]
45. Hermann A, Monoranu C, Schwarz J, Petri S, Storch A. Reduced LRRK2-positive neurons in the striatum of Parkinson's disease patients hypothesize a retrograde disease mechanism? *Basal Ganglia*. 2012; 2:67–72.
46. Altar CA, Cai N, Bliven T, Juhasz M, Conner JM, Acheson AL, Lindsay RM, Wiegand SJ. Anterograde transport of brain-derived neurotrophic factor and its role in the brain. *Nature*. 1997; 389:856–60. [PubMed: 9349818]
47. Altar CA, DiStefano PS. Neurotrophin trafficking by anterograde transport. *Trends Neurosci*. 1998; 21:433–7. [PubMed: 9786341]
48. Gold DA, Gent PM, Hamilton BA. ROR alpha in genetic control of cerebellum development: 50 staggering years. *Brain Res*. 2007; 1140:19–25. [PubMed: 16427031]
49. Goedert M, Spillantini MG, Del Tredici K, Braak H. 100 years of Lewy pathology. *Nat Rev Neurol*. 2013; 9:13–24. [PubMed: 23183883]
50. Kordower JH, Chu Y, Hauser RA, Freeman TB, Olanow CW. Lewy body-like pathology in long-term embryonic nigral transplants in Parkinson's disease. *Nat Med*. 2008; 14:504–6. [PubMed: 18391962]
51. 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 USA*. 2009; 106:13010–5. [PubMed: 19651612]
52. Kordower JH, Dodiya HB, Kordower AM, Terpstra B, Paumier K, Madhavan L, Sortwell C, Steece-Collier K, Collier TJ. Transfer of host-derived synuclein to grafted dopaminergic neurons in rat. *Neurobiol Dis*. 2011; 43:552–7. [PubMed: 21600984]

53. Luk KC, Kehm V, Carroll J, Zhang B, O'Brien P, Trojanowski JQ, Lee VM. Pathological α -synuclein transmission initiates Parkinson-like neurodegeneration in nontransgenic mice. *Science*. 2012; 338:949–53. [PubMed: 23161999]
54. Biskup S, Moore DJ, Celsi F, Higashi S, West AB, Andrabi SA, Kurkinen K, Yu SW, Savitt JM, Waldvogel HJ, Faull RL, Emson PC, Torp R, Ottersen OP, Dawson TM, Dawson VL. Localization of LRRK2 to membranous and vesicular structures in mammalian brain. *Ann Neurol*. 2006; 60:557–69. [PubMed: 17120249]
55. Sulzer D, Surmeier DJ. Neuronal vulnerability, pathogenesis, and Parkinson's disease. *Mov Disord*. 2013; 28:41–50. [PubMed: 22791686]
56. Rangasamy SB, Soderstrom K, Bakay RA, Kordower JH. Neurotrophic factor therapy for Parkinson's disease. *Prog Brain Res*. 2010; 184:237–64. [PubMed: 20887879]
57. Steiner H, Blum M, Kitai ST, Fedi P. Differential expression of ErbB3 and ErbB4 neuregulin receptors in dopamine neurons and forebrain areas of the adult rat. *Exp Neurol*. 1999; 159:494–503. [PubMed: 10506520]
58. Decressac M, Kadkhodaei B, Mattsson B, Laguna A, Perlmann T, Björklund A. α -Synuclein-induced down-regulation of Nurr1 disrupts GDNF signaling in nigral dopamine neurons. *Sci Transl Med*. 2012; 4:163ra56.
59. Liu J, Zhou Y, Wang C, Wang T, Zheng Z, Chan P. Brain-derived neurotrophic factor (BDNF) genetic polymorphism greatly increases risk of leucine-rich repeat kinase 2 (LRRK2) for Parkinson's disease. *Parkinsonism Relat Disord*. 2012; 18:140–3. [PubMed: 21924942]
60. Carta A. The role of microglia–lymphocyte interaction in PD neuropathology. *Basal Ganglia*. 2012; 2:123–30.
61. Van Waes V, Tseng KY, Steiner H. GPR88 - a putative signaling molecule predominantly expressed in the striatum: Cellular localization and developmental regulation. *Basal Ganglia*. 2011; 1:83–9. [PubMed: 21804954]
62. Oorschot DE. The percentage of interneurons in the dorsal striatum of the rat, cat, monkey and human: A critique of the evidence. *Basal Ganglia*. 2013; 3:19–24.

Fig. 1.

Illustration of LRRK2 mRNA expression in midbrain dopamine neurons vs. striatum of the rat, as determined by in situ hybridization histochemistry [61]. (A) Film autoradiograms depict LRRK2 (top) and tyrosine hydroxylase (TH, bottom) mRNA in the midbrain. As previously shown (e.g., [36,37,40]), the expression of LRRK2 in dopamine neurons of the substantia nigra (pars compacta, SNpc) and ventral tegmental area is very low or below the threshold of detection. The location of the dopamine neurons is shown in an adjacent section, by TH mRNA labeling (arrows). Note also the robust LRRK2 signal in cortex (Cx) and hippocampus (H). (B) Film autoradiograms show LRRK2 mRNA expression in the rostral (top) and caudal (bottom) striatum (S) and nucleus accumbens (NAC). In contrast to the midbrain, the striatum displays high levels of LRRK2 expression. Also, note the absence of a LRRK2 mRNA signal in the external globus pallidus (GPe) (see text). (C) LRRK2 expression varies between individual neurons of the striatum, but is present in projection neurons and most interneuron types [40,44]. Neurons were identified with NeuN immunoreactivity (top) [61], and LRRK2 mRNA was labeled with fluorescence in situ hybridization histochemistry (middle). Merged images (bottom) show double labeled cells (arrow heads). Based on their size and the fact that interneurons account for less than 3% of striatal neurons [62], most or all of the shown neurons are presumed projection neurons. Images courtesy of V. Van Waes and H. Steiner. Supported by USPHS grant DA011261.