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Molecular pathways in sporadic PD

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SUMMARY

Over the last decade, several autosomal dominant and recessive genes causative of Parkinson's disease (PD) have been identified. The functional studies on their protein products and the pathogenetic effect related to their mutations have greatly contributed to understand the many cellular pathways leading to neurodegeneration, that include oxidative stress damage, mitochondrial dysfunction, misfolded protein stress and impairment of cellular clearance systems, namely the ubiquitin-proteasome system (UPS) and the autophagy pathway.
Although mendelian genes are responsible only for a small subset of PD patients, it is expected that the same pathogenetic mechanisms could play a relevant role also in the more frequent sporadic PD, that is currently recognized as a multifactorial disorder. In this model, different genetic and environmental factors, either playing a protective or a susceptibility role, variably interact to reach a threshold of disease over which PD will become clinically manifest. As an example, mutations or multiplication of the alpha-synuclein gene cause autosomal dominant PD, while common genetic variants at the same locus have been consistently associated to the risk of developing PD by genome-wide association studies. These findings are opening novel interesting perspectives to identify critical molecular pathways leading to neurodegeneration.

1. Introduction

For a long time, Parkinson's disease has been largely considered a "non-genetic" condition. This concept has radically changed in the last decade, with the identification of several genes that, when mutated, cause monogenic PD with autosomal dominant or recessive inheritance. Genetic studies performed on very large cohorts of PD patients, as well as the availability of innovative, high throughput technologies to study the human genome, have clearly demonstrated that the genetic risk to develop PD represents a continuum from highly penetrant mutations at one end of the spectrum to low risk variants at the other end [1]. Autosomal recessive mutations in genes such as Parkin, PINK1 and DJ-1 are the only ones to present complete penetrance, meaning that the presence of homozygous or compound heterozygous mutations is sufficient *per se* to cause the parkinsonian phenotype. Even these fully penetrant, truly monogenic parkinsonisms can manifest in subjects with negative family history, and in fact mutations in recessive genes are found in up to 10% of sporadic patients with early onset PD [2]. Mutations in autosomal dominant genes, such as SNCA (encoding alpha-synuclein) and LRRK2, present markedly reduced penetrance, justifying the lack of family history in several mutated patients [1]. Besides these rare forms, in the majority of cases PD can be considered a multifactorial, complex disorder, in which several genetic and environmental factors concur to reach a threshold of disease. To date, genetic variants in several genes have been convincingly implicated in the etiology of sporadic PD. Among these, single heterozygous mutations in the glucocerebrosidase (GBA) gene play a key role, increasing the risk to develop the disease up to five times in carriers vs non carriers [3]. Other common variants in autosomal dominant genes (e.g. the SNCA REP1 promoter polymorphism, or the G2385R and S1647T variants in the LRRK2 gene) as well as heterozygous rare variants in autosomal recessive PD genes have also been associated to an increased risk to develop PD, although with low odds ratio values <2 [1,4–6]. The current technological revolution, with the advent of massive parallel sequencing techniques, is likely to reveal a far greater number of rare variants in known or novel genes, that could account for an additional proportion of the PD genetic risk in sporadic cases.

2. Molecular pathways in sporadic PD

The increasing number of genes and proteins linked to PD is unraveling a complex network of molecular pathways involved in its etiology, suggesting that common mechanisms underlie both familial and sporadic forms. Three main pathways have emerged that can trigger the neurodegenerative process: accumulation of misfolded and aggregated proteins, impairment of clearance systems such as the ubiquitin protein pathway (UPS) and the autophagy pathway, and mitochondrial dysfunction. All these

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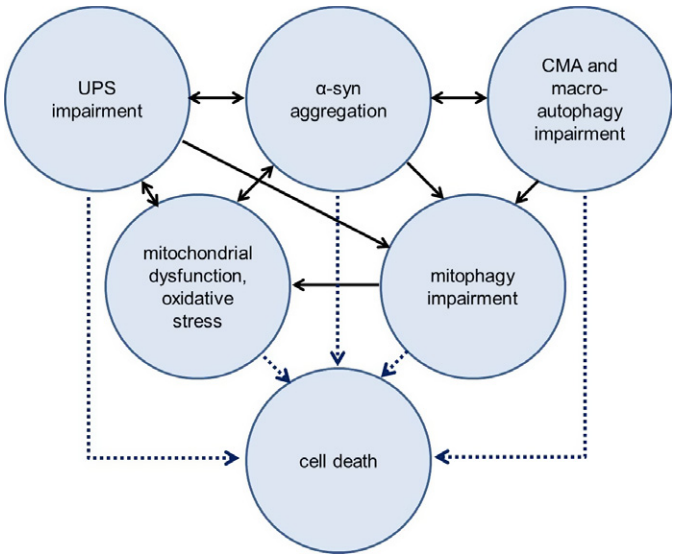

Fig. 1. In sporadic PD, the neurodegenerative process is elicited at the molecular level by a complex intersection of distinct pathways, that are variably compromised due to aging, individual genetic background and environmental exposure. The ongoing dysfunction of the neuronal pool of mitochondria, the increased levels of oxidative stress and of protein aggregates, along with the malfunctioning of protective pathways such as the UPS, chaperon-mediate autophagy (CMA), macroautophagy and mitophagy, concur to impair neuronal homeostasis beyond its ability to recovery. This long-term process triggers the apoptotic cell death of selected neuronal populations and progressively leads to the development of clinically evident PD.

models are supported by functional studies on the proteins encoded by PD-related genes, but at the same time have found confirmation by pathological and biochemical studies performed in patients with sporadic PD and no apparent genetic cause [5,7,8]. Overall, autosomal dominant PD genes encode for proteins that are toxic when mutated or overexpressed; conversely, proteins encoded by autosomal recessive genes are actively involved in neuroprotective pathways aimed at counteract neuronal damage, and their haploinsufficiency (as it happens in carriers of heterozygous mutations) may significantly impair their function and make the neurons more vulnerable to aging and stress exposure. In particular, aging is a crucial trigger of the neurodegenerative process, since most cellular protective pathways such as the UPS, autophagy and regulation of mitochondria dynamics, are known to lose efficiency with time, and there is progressive accumulation of somatic mutations especially in the mitochondrial DNA during life [9] (Fig. 1). Moreover, recent studies have implicated a role for chronic neuroinflammation and microglia activation in PD pathogenesis, suggesting that different molecular or cellular events (such as haploinsufficiency in neuroprotective genes, protein aggregation or oxidative stress) may contribute to neurodegeneration by activating resident microglial populations in selected brain areas, with potential detrimental effects on vulnerable neuronal populations [10].

3. Pathological protein aggregation

The pathological accumulation of misfolded or aggregated proteins within neuronal cells has long been recognized a central pathogenetic mechanism shared by most neurodegenerative disorders. In PD, striking evidence came from the finding that Lewy bodies, the intracytoplasmic inclusions found in dopaminergic neurons that are pathognomonic for PD, are mainly constituted by aggregated alpha-synuclein, the first protein to be found mutated in rare cases of dominant PD. Remarkably, aggregated alpha-synuclein has been found as the major component of Lewy bodies also in the brains of patients with sporadic PD, suggesting a key role for this protein in the pathogenesis of the common, non-mendelian form of the disease. Not only pathogenic mutations, but also overexpression of alpha-synuclein increases its propensity to aggregate, and in fact multiplications of the SNCA gene are also causative of autosomal dominant PD, with a consistent correlation between the total number of gene copies and the severity of the disease [11]. In sporadic cases, a specific polymorphic variant in the promoter region of the SNCA gene (REP1) is known to increase the levels of gene expression and alpha-synuclein production, and carriers of this variant bear a significantly higher risk to develop PD compared to non-carriers. Additional variants at the 3′ untranslated region of the SNCA gene have also been significantly associated with a higher risk to develop PD, and correlate with the levels of alpha-synuclein both in peripheral blood and brain tissue [12]. Several post-translational modifications of alpha-synuclein, including nitration and hyperphosphorylation at Ser129, are known to increase its tendency to aggregate and misfold, as well as the presence of dopamine adducts, which could partly explain the selective susceptibility to dopaminergic neurons to the neurodegenerative process [13]. Interestingly, it has recently been demonstrated that alpha-synuclein may propagate between neuronal cells following a prion-like mechanism, following the observation of alpha-synuclein-containing Lewy bodies in embryonic dopamine cells transplanted in PD patients. This intriguing phenomenon has been advocated to explain the spreading of PD pathology over time, but the underlying molecular pathways still need to be fully understood [14].

4. Mitochondrial damage

Overexpression of alpha-synuclein has been shown to induce mitochondrial depolarization and release of cytochrome c in association with cell death, suggesting an effect on the mitochondrial permeability. Also, alpha-synuclein can affect components of the electron transport chain and result in the generation of excessive reactive oxygen species in dopaminergic neurons. The oxidative stress that is known to occur in dopaminergic neurons due to dopamine metabolism and mitochondrial dysfunction may further precipitate protein misfolding and aggregation, perpetuating a deleterious vicious circle [13]. Finally, alpha-synuclein is also known to imbalance mitochondrial dynamics by inhibiting fusion and promoting fragmentation [15]. Both parkin and PINK1 proteins are also critically involved in regulation of mitochondrial dynamics and in selective removal of damaged mitochondria through mitophagy (see below), and their malfunction leads to impaired mitochondrial morphology and integrity [16]. Moreover, parkin is directly implicated in the control of mitochondrial biogenesis through regulation of PARIS [17]. Several studies have consistently shown that PINK1 is an antiapoptotic protein implicated in the maintenance of mitochondrial membrane potential, in regulation of intramitochondrial calcium levels and in the activation of anti-apoptotic chaperon proteins. Similarly, DJ-1 is also essential to counteract oxidative stress acting as a mitochondrial chaperone and oxidative stress sensor, that palys a fundamental role in preserving the integrity and function of the mitochondrial pool [18]. The impairment of mitochondrial function in sporadic PD is also supported by evidences that exposure to environmental toxins known to damage mitochondrial function (such as complex I blockers) represents a significant risk factor for PD. Moreover somatic mitochondrial DNA deletions were found to accumulate in the brain of aging individuals as well as PD patients, leading to respiratory chain deficiency [9].

5. Impairment of clearance systems

Two pathways have been found to degrade misfolded and aggregated proteins, the ubiquitin-proteasome system (UPS) and

the autophagy pathway. It has recently been shown that over-expression of mutant or dopamine-modified alpha-synuclein is able to impair both clearing mechanisms, leading in turn to a further accumulation of toxic aggregated species as well as of other damaging components of the cell, such as dysfunctional mitochondria [19].

The parkin protein is a ubiquitin E3-ligase that ubiquitinates protein targets for UPS-mediated degradation, thus contributing to the removal of misfolded and damaging proteins such as PARIS [17]. Parkin-mediated ubiquitination is also required to promote the degradation of several proteins of the outer mitochondrial membrane (including mitofusin and VDAC1), a step which precedes the removal of dysfunctional mitochondria through mitophagy [20]. In the mitophagy pathway, Parkin is specifically recruited by PINK1, that selectively accumulates on the surface of depolarized mitochondria and targets them to autophagy-based degradation [21]. PINK1 itself is known to directly activate autophagy and to interplay with autophagic proteins such as Beclin1 [22], and a reduced clearance of mitochondria was also demonstrated in cells lacking DJ-1 [23].

Other proteins encoded by recessive genes are also implicated in the lysosome-autophagy pathway. Mutant LRRK2, that represents a relevant genetic cause of sporadic PD, has been suggested to promote alpha-synuclein accumulation through impairment of the autophagy pathway [7]. Autosomal recessive mutations in ATP13A2, a lysosome ATPase, are responsible for Kufor-Rakeb syndrome, a complex disorder characterized by early onset parkinsonism associated with other clinical features. Single heterozygous mutations in this gene have been occasionally detected in patients with early onset PD, suggesting that the haploinsufficiency of this enzyme could contribute to PD pathogenesis by impairing lysosomal function [1,24]. Finally, the GBA gene encodes for glucocerebrosidase, another lysosomal enzyme. It has been postulated that heterozygous mutations in GBA could lead to lysosomal dysfunction or possibly interfere with binding of alpha-synuclein to its specific receptor at the lysosome membrane. A recent study has shown that the accumulation of the GBA substrate glucosylceramide is also able to stabilize alpha-synuclein soluble oligomers and, in turn, alpha-synuclein can inhibit normal GBA lysosomal activity, creating a "positive feedback loop" that could directly contribute to the neurodegenerative process [25].

6. Conclusions

Growing evidence from genetic and functional studies is demonstrating a convergence of shared pathogenetic pathways in familiar and sporadic PD, such as mitochondrial dysfunction, oxidative stress damage, toxic protein aggregation and derangement of cellular clearing systems. The incomplete penetrance associated with genetic variations in most PD-related genes clearly speaks for a multifactorial basis that underlies the vast majority of PD cases. In these patients, several genetic and environmental factors, as well as aging, concur to the progressive neuronal damage that eventually results in clinically manifest PD. The understanding of these complex, convergent pathways, along with the identification of effective preclinical biomarkers, will pave the way for innovative therapeutic approaches, aimed not only at alleviating the symptoms of disease but also at preventing its manifestation through concerted neuroprotective strategies.

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Conflict of interests

The authors have no conflicts of interest to declare.

References

1. Lesage S, Brice A. Parkinson's disease: from monogenic forms to genetic susceptibility factors. Hum Mol Genet 2009;18:R48–59.
2. Lucking CB, Durr A, Bonifati V, Vaughan J, De Michele G, Gasser T, et al. Association between early-onset Parkinson's disease and mutations in the Parkin gene. N Engl J Med 2000;342:1560–7.
3. Sidransky E, Nalls MA, Aaslu JO, Aharon-Peretz J, Annesi G, Barbosa ER, et al. Multicenter analysis of glucocerebrosidase mutations in Parkinson's disease. N Engl J Med 2009;361:1651–61.
4. Farrer MJ, Stone JT, Lin CH, Dachsel JC, Hulihan MM, Haugarvoll K, et al. Lrrk2 G2385R is an ancestral risk factor for Parkinson's disease in Asia. Parkinsonism Relat Disord 2007;13:89–92.
5. Burbulla LF, Kruger R. Converging environmental and genetic pathways in the pathogenesis of Parkinson's disease. J Neurol Sci 2011;306:1–8.
6. Lin CH, Wu RM, Tai CH, Chen ML, Hu FC. Lrrk2 S1647T and BDNF V66M interact with environmental factors to increase risk of Parkinson's disease. Parkinsonism Relat Disord 2011;17:84–8.
7. Martin I, Dawson VL, Dawson TM. Recent advances in the genetics of Parkinson's disease. Annu Rev Genom Human Genet 2011;12:301–25.
8. Cookson MR, Bandmann O. Parkinson's disease: insights from pathways. Hum Mol Genet 2010;19:R1–27.
9. Bender A, Krishnan KJ, Morris CM, Taylor GA, Reeve AK, Perry RH, et al. High levels of mitochondrial DNA deletions in substantia nigra neurons in aging and Parkinson disease. Nat Genet 2006;38:515–7.
10. Tansey MG, Goldberg MS. Neuroinflammation in Parkinson's disease: its role in neuronal death and implications for therapeutic intervention. Neurobiol Dis 2010;37:510–8.
11. Singleton AB, Farrer M, Johnson J, Singleton A, Hague S, Kachergus J, et al. Alpha-Synuclein locus triplication causes Parkinson's disease. Science 2003;302:841.
12. Pihlstrøm L, Toft M. Genetic variability in SNCA and Parkinson's disease. Neurogenetics 2011 Jul 29 [Epub ahead of print]. doi: 10.1007/s10048-011-0292-7.
13. Venda LL, Cragg SJ, Buchman VL, Wade-Martins R. alpha-Synuclein and dopamine at the crossroads of Parkinson's disease. Trends Neurosci 2010;33:559–68.
14. Angot E, Steiner JA, Hansen C, Li JY, Brundin P. Are synucleinopathies prion-like disorders? Lancet Neurol 2010;9:1128–38.
15. Schapira AH. Mitochondria in the aetiology and pathogenesis of Parkinson's disease. Lancet Neurol 2008;7:97–109.
16. Bueler H. Mitochondrial dynamics, cell death and the pathogenesis of Parkinson's disease. Apoptosis 2010;15:1336–53.
17. Shin JH, Ko HS, Kang H, Lee Y, Lee YI, Pletinkova O, et al. PARIS (ZNF746) repression of PGC-1alpha contributes to neurodegeneration in Parkinson's disease. Cell 2011;144:689–702.
18. Cookson MR. DJ-1, PINK1, and their effects on mitochondrial pathways. Mov Disord 2010;25(Suppl 1):S44–8.
19. Matsuda N, Tanaka K. Does impairment of the ubiquitin-proteasome system or the autophagy-lysosome pathway predispose individuals to neurodegenerative disorders such as Parkinson's disease? J Alzheim Dis 2010;19:1–9.
20. Karbowski M, Youle RJ. Regulating mitochondrial outer membrane proteins by ubiquitination and proteasomal degradation. Curr Op Cell Biol 2011;23:476–82.
21. Vives-Bauza C, Przedborski S. Mitophagy: the latest problem for Parkinson's disease. Trends Mol Med 2011;17:158–65.
22. Michiorri S, Gelmetti V, Giarda E, Lombardi F, Romano F, Marongiu R, et al. The Parkinson-associated protein PINK1 interacts with Beclin1 and promotes autophagy. Cell Death Differ 2010;17:962–74.
23. McCoy MK, Cookson MR. DJ-1 regulation of mitochondrial function and autophagy through oxidative stress. Autophagy 2011;7:531–2.
24. Di Fonzo A, Chien HF, Socal M, Giraudo S, Tassorelli C, Iliceto G, et al. ATP13A2 missense mutations in juvenile parkinsonism and young onset Parkinson disease. Neurology 2007;68:1557–62.
25. Mazzulli JR, Xu YH, Sun Y, Knight AL, McLean PJ, Caldwell GA, et al. Gaucher disease glucocerebrosidase and alpha-synuclein form a bidirectional pathogenic loop in synucleinopathies. Cell 2011;146:37–52.