Why do dopaminergic neurons die in sporadic Parkinson's disease?

1 Introduction

Parkinson's disease (PD) is a late-life progressive neurodegenerative disorder characterised by the selective death of dopaminergic neurons in the substantia nigra pars compacta (SNpc) [1]. PD patients develop muscle rigidity, slowed movement, tremors at rest and difficulty in initiating and ending movement [2]. As of 2015, over six million people are affected by this disease [3], with approximately 90-95% of all cases being sporadic[4].

Dopaminergic neurons serve as modulators for the direct and indirect pathways of the basal ganglia. This compound system is known as the nigrostratal pathway, and its main function is the control of voluntary motor movement [5]. When dopaminergic neurons die, the lack of dopamine in the striatum prompts a decrease in the thalamic excitation of the motor cortex [6]. This selective neuronal death brings the principal motor-related symptoms of PD [5].

The exact causes of dopaminergic neuron death are not known to date. There is a general consensus of how the interplay of different hypotheses may lead to the death of dopaminergic neurons, and how these processes originate and lead to PD [1][7]. The four principal hypotheses underlying the pathogenesis of dopaminergic cell death are mitochondrial dysfunction, oxidative stress, neuroinflammation and the misfolding and aggregation of proteins [7].

The etiological factors behind these mechanisms are unknown as well, and current studies situate environmental toxic factors and genetic predisposition as the most likely causes [8]. This essay will study the work done on pathogenesis of dopaminergic neuronal death in order to better understand and assess the current limitations of its etiology. Additionally , different approaches to the study of neuronal death will be proposed.

2 Mitochondrial dysfunction, oxidative stress and neuroinflammation

Since PD mostly affects individuals over the age of 60, the vulnerability of dopaminergic neurons as a result of ageing is thought to be a risk factor [9]. The consequent deterioration of normal cell processes may make these neurons more susceptible to neuronal death. This aspect is of special importance, since many studies fail to take this aspect into consideration when it should be given more attention [10].

The increase of oxidized lipids in the SNpc and the decreased levels of glutathione (GSH) observed in PD patients [11][12] suggests that oxidative stress is an important factor in PD neuronal death. Oxidative stress originates when there is an imbalance between the production of reactive oxygen species and antioxidant activity from the cell.

Dopaminergic cells are specially prone to oxidative stress, since they have enzymes such as tyrosine hydroxylase which generate ROS. Dopamine(DA) is also a cause of oxidative stress, since excessive cytosolic DA can be oxidized enzymatically to produce DA quinone [4]. Due to DA metabolism transition metal ions in the cell also induce the conversion of hydrogen peroxide into reactive hydroxyl radicals, which contribute to oxidative stress [13].

However, this implies that other dopaminergic areas of the brain should theoretically be affected, whereas in PD, the death of these neurons is limited to the SNpc. The exact source and attributes of the ROS need to be determined, since they are not accounted for in DA metabolism and they would be able to explain the motive behind localised neuronal death.

DA quinones are able to covalently modify important cellular nucleophiles that are essential for basic cell survival, such as protein cysteinyl residues and GSH [4]. When mitochondrial GSH is consumed due to oxidative stress, the calcium homeostasis of the mitochondria will be disrupted [14]. DA quinone is also capable of modifying PD-related proteins such as α-synuclein, Parkin and DJ-1 [15]. It is also able to severally alter the respiratory chain of the mitochondria by altering the sub-units of Complex I and III of the respiratory chain[16]. This mitochondrial dysfunction produces more ROS in exchange, acting as a vicious cycle.

The last adverse effect of DA quinone is its indirect triggering of neuroinflammation. It is able to cyclize into aminochrome, whose redox-cycling generates superoxide which in turn polymerises to assemble neuromelanin. Neuromelanin may then cause neuroinflammation [17].

Neuroinflammation appears as a result of microglial reaction, which has been identified in the SNpc of sporadic PD patients [18]. This reaction could be explained as the response of oxidative-originated toxic stress. Since microglial overaction is susceptible to cause excessive noxious neuroinflammation [19], oxidative stress could be situated as the causal nexus. Further research is needed in this area.

The study of the first documented case of MPTP-induced parkinsonism [20] brought the hypothesis of mitochondrial dysfunction as the cause behind dopaminergic neuronal death. Users who injected drugs contaminated with MPTP into their systems developed parkinsonism, due to localised death of dopaminergic neurons in the SNpc. This brought the attention of several researchers, who later found out that monoamime oxidised MPTP into to MPP+, which is highly toxic [21].

MPP+ was discovered to be responsible for the death of dopaminergic neurons in the SNpc: the complex I respiratory pathway and the α-ketoglutarate dehodryganse complex (KGDHC) is inhibited by MPP+ [22]. The corresponding loss of effective electron transport and ATP synthesis from inhibited mitochondrial respiration leads the dopaminergic cells to their death.

After these discoveries, extensive analysis was done on PD patients and it was discovered that the same mitochondrial dysfunction was present. In complex I, the activity of nicotinamide adenine diriucleotide (NADH), a specific enzyme of complex I, was inhibited. Correspondingly, the activity of complex I was reduced as well [23]. KGDHC was also found in abnormally low proportions, which in turn aggravated mitochondrial dysfunction by inhibiting complex II and III [24].

The interplay between oxidative stress and mitochondrial dysfunction has been the object of many studies. Oxidative stress was defined as the cause of mitochondrial dysfunction[25], but modern research situates it as second to mitochondrial dysfunction [22]. The formation of superoxide anions by complex I and III inhibition [26] and the consequent electron leakage serve as compelling arguments to demonstrate Yoshikuni Mizuno's claims. Nevertheless, the results are not conclusive enough to fully

validate this reaction chain, and as such, information where the causality of either mechanism is stated as a fact should be approached critically.

Apoptosis was also thought to be a cause of nigral death. Studies later found apoptosis as the cause of dopaminergic neuronal death in PD patients [27]. Even though oxidative stress and ATP deficiency may trigger apoptosis [28], apoptosis may hold no relation with these mechanisms and be a part of a yet undiscovered process. Therefore, the factors behind cell apoptosis need to be fully understood before any claim can be made, since for all we know, apoptosis may be acting as a natural process and not as a PD-related trigger.

3 α-synuclein and Lewy Bodies

Lewy bodies(LB) are a pathological characteristic of sporadic and familial PD. They serve as a means of identifying cell suffering and are not limited to either PD or dopaminergic neurons in the SNpc [29]. Their composition is unknown, but studies have proved that they contain neurofilament antigens [30] and α-synuclein [31], the latter being the most relevant to us.

The functions of LB are still unidentified as a whole. The only lead available that connects them with PD is its composition of α-synuclein. Unfortunately, most research in PD does not focus on LB and instead targets α-synuclein, which may seem like a possible mistake, since we are interested in how the presence of LB may impact neuronal death.

On the other hand, the protein α-synuclein seems to play a crucial role in neuronal death situated in the SNpc. Its cellular functions are unknown, but extensive research has demonstrated that α-synuclein misfolds and creates aggregates which are able to spread trans-synaptically between neurons [32]. The aggregation of α-synuclein is the only lead available to discern the origin of LB in sporadic PD. Interistingly enough, a mutation in α-synuclein of familial PD elicits the same effects (discussed later).

α-synuclein seems to be an important factor behind cell death in sporadic PD. Numerous studies have found accumulation of α-synuclein aggregates to be the culprit of protein alteration [33], neuroinflammation [4], mitochondrial dysfunction [34][35] and the leakage of dopamine via vesicle membrane permeabilization [36] which exerts oxidative stress.

However, this does not imply that α-synuclein is the origin of these mechanisms, but it may accelerate them. In the same fashion in which Mitochondrial dysfunction, oxidative stress and neuroinflammation all interplay, α-synuclein seems to take an important part in this scheme by boosting their harmful effects. The possibility of α-synuclein taking advantage of enfeebled and susceptible dopaminergic neurons by bringing their already weakened and balance to a critical point is also another possibility.

4 Etiology

The origin of neuronal death is unknown, but extensive research has led to two main hypotheses: the environmental toxin hypothesis and the genetic predisposition hypothesis.

The environmental toxin hypothesis postulates that dopaminergic toxins in the environment are responsible for all of the damage listed in the previous sections. Toxins are tested and compared with the effects of MPP+ to determine their degree of toxicity. However, individual reports on different neurotoxins and environmental influences have generated conflicting results. One example is how some studies claim elevated risk from herbicide use [37] , but others do not [38].

Other approaches use animal models by studying the reaction of these animals to different potential neurotoxins and analysing the results, in an attempt to detect PD symptoms and MPTP-like effects. Two distinguishable components are tetrahydroisoquinolines (TIQ) and Rotenone. Both are natural components found in the environment, although the latter is used for insecticide and fish poison production. Results here are conflicting as well but the main issue lies with the animal model itself. Animal models may not be the best approach, since they lack the ability to imitate factors such as ageing and do not possess sufficient level of detail. As such, findings obtained by these means may be easily misinterpreted and need to be analysed carefully.

On the other hand, genetic predisposition research seems more promising and has yielded more conclusive results. The initiative of this hypothesis originates in the near identical similarity found between familial and sporadic PD, which implies that the pathogenic mechanisms remain identical as well [39]. This gives researchers the opportunity to study familial PD genetic mutations by identifying affected proteins and then seeking similar processes in sporadic PD.

This approach led to the discovery of a genetic basis in sporadic PD. One particular mutation in this basis may explain the origin of α-synuclein misfolding, which is still unknown. This mutation, situated in α-synuclein seems to create α-SynL, a α-synuclein transcript isoform with longer 3'UTR [40]. The presence of α-SynL aggravates risk factors ,such as ageing and oxidative stress, in dopaminergic neurons. Amongst other familial genes such as PARK2 and PARK6, LRRK2 was found to contribute to sporadic PD as well.

After studying both etiological hypotheses it would seem that the genetic predisposition hypothesis has brought better insights. Nevertheless, even if there are conflicting results in environmental toxin research, in all likelihood they play an important part in neuronal death and their study helps us understand neuronal death better. An ideal approach would be to study the interplay of both hypotheses, instead of focusing on each one separately with minor emphasis on the other. The increment of twin studies in this area would also prove useful.

5 Discussion and conclusion

As it can be surmised, dopaminergic neural death cannot be attributed to a single factor; it is an interplay between many different factors of various origins. The discoveries of MPTP induced parkinsonism and genetic factors of PD laid the foundation to most research in the field. Still, further research is needed and new approaches might need to be developed, since even after two centuries of research, not much is known. Perhaps our issues in the past lied in our failure of seeing PD as a

multi-causal disease.

Due to this multi-causal nature, there may be overlooked research in other areas which could add to our understanding of PD. Data Mining approaches could be used in public repositories to find links between information that researchers could not, due to the vast quantity of information available. Natural Language Processing techniques are specially useful and they are used nowadays for similar purposes.

Future lines of research could tackle current limitations, such as our current toxin-based animal models. Seeing that our models are insufficiently complex, *in silico* models could fill in these gaps, since we could simulate PD infected brains with ease. Projects such as the HBP could ultimately serve this purpose.

Lastly, different types of studies could be proposed, such as the possible causes for which men suffer PD more than women. Work on this topic could ultimately help us discover natural defence mechanisms present in women.

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