FACULDADE DE MEDICINA
UNIVERSIDADE D
COIMBRA

INTEGRATED MASTER IN MEDICINE - FINAL PAPER

HELENA CRISTINA NUNES DA COSTA

Parkinson's disease: a link between systems.

NARRATIVE REVIEW ARTICLE

SCIENTIFIC AREA OF CELL BIOLOGY

Work done under the guidance of:

SANDRA MORAIS CARDOSO

ANA RAQUEL ESTEVES

MARCH/2021

Index

Abbreviations index

AS - Adaptive Immune System

ANS - Autonomic Nervous System

BMAA - neurotoxin β-Methylamino- l-alanine

CNS - Central Nervous System

DAMP - danger associated molecular pattern

ENS - Enteric Nervous System

ES - Endocrine System

BBB - Blood-Brain Barrier

GI - gastrointestinal

IEC - intestinal epithelial cells

IgA - immunoglobulin A

IGF-I - Insulin-Like Growth Factor-I

IL-10 - Interleukin-10

IS - Immune System

NMS - non-motor symptoms

NS - Nervous System

MAO B - monoamine oxidase B

MS - motor symptoms

PD - Parkinson's disease

RBD - REM sleep behavior disorder

RF - risk factors

ROS - reactive oxygen species

SCFAs - short chain fatty acids

TJ - tight junctions

TLR4 - Toll-Like 4 receptors

Abstract:

The way sporadic Parkinson's disease (PD) is perceived has undergone drastic changes in recent decades. Previously it was a reductive synonym for motor disorders associated with it. Today, with the appreciation of non-motor symptoms (NMS) and inherent pathological processes, there is a greater understanding of the involvement of other organ systems. For this reason, PD is increasingly seen as a multiorgan pathology that arises from the interaction of a susceptible individual with a challenging environment during the decline of the patient's *performance status* due to aging.

The identification of several risk factors (RF) and the hypothesis that PD has a gastrointestinal (GI) onset sheds light on the pathophysiological processes behind the pathology. The association of these processes between different systems puts emphasis on a systemic proinflammatory microenvironment in the Parkinson's patient. This pro-inflammatory state acts synergistically with *inflammaging*, contributing to PD development, predominantly in the elderly. This subclinical chronic inflammatory state contributes to the disruption of biological mechanical barriers, such as the intestinal barrier comprised by the GI epithelium and the endothelial cells of the Blood-Brain Barrier (BBB), which ultimately allows the progression and development of the disease.

Prodromal pathological processes' knowledge may, in the future, allow biomarkers' identification and establishment of PD risk scores for early diagnosis. This knowledge also paves the way for the discovery of new therapeutic targets.

Thus, the aim of this review is to present the Parkinson's patient in a complete perspective, highlighting the possible pathophysiological events that precede dopaminergic neuronal loss and the development of motor symptoms, as well as the communication and disruption of gut-brain barriers during the propagation of these events.

Keywords: Parkinson's disease, Gut Microbiome, Intestinal Barrier, α-Synuclein, Blood-Brain Barrier.

1. Introduction:

Parkinson's disease (PD) is currently described as a multi-organ neurodegenerative disease, since it concomitantly involves the Central Nervous System (CNS), Enteric Nervous System (ENS), Autonomic Nervous System (ANS), Adaptive Immune System (AS) and the Gastrointestinal (GI) tract [1–5]. Clinically it is characterized by the occurrence of both motor (MS) and non-motor (NMS) symptoms [6].

Bradykinesia associated with resting tremor and rigidity is still considered a cardinal manifestation of the disease, but it is insufficient for clinical diagnosis. The NMS have been gaining importance in the pathology's description. Thus, an established clinical diagnosis requires, besides the cardinal manifestations, the presence of at least two supporting criteria, in the absence of exclusion criteria or "*red flags*". The support criteria also include dopaminergic therapy's response, dyskinesia secondary to levodopa treatment and the presence of NMS, such as anosmia and cardiac sympathetic denervation [1,6].

NMS, such as hyposmia/anosmia, REM sleep behavior disorder (RBD), depression, anxiety and constipation, may precede dopaminergic neurons' loss and the consequent MS, by several years [6,7]. Although NMS are present during disease progression and are an important cause of loss of quality of life their presentation and timing vary. Sleep and ANS disturbances are present from early to later stages but dementia and psychosis are more likely to occur in later disease stages [5,7]. The presence of these manifestations is a reminder of the inherent pathophysiology of PD which extends beyond the *pars compacta's* dopaminergic neurons loss [1,6–9].

2. Before the race: Risk factors.

It is thought that there is a close relationship between genetic predisposition, a favorable environment, and patient's ageing in the onset of the pathology. Therefore, although hereditary forms of the disease have been described, most cases are sporadic with no known etiology. As a result, several possible risk factors have been suggested for this pathology [1,10–12].

2.1. Exposure to external agents and lifestyles

Rural living, farming and well-water consumption correlate with an increased risk of developing PD [13,14]. Although this epidemiological relationship is not completely understood, it is known that there is an increased risk of PD in individuals with occupational exposure to pesticides, as well as, a worse prognosis of the disease [13,15]. This risk is more clear with pesticides that affect the complex I of the mitochondrial respiratory chain and induce oxidative stress, such as Rotenone and Paraquat [1,13–18]. Although the heavy metal's neurotoxic action has been described in animal models, its exposure and PD' increased risk remains controversial [1,11]. Iron free form is capable of inducing oxidative stress and its accumulation in the *substantia nigra* in the brains of patients is also known [19,20]. However, and despite the hypothesis that alterations in iron metabolism may confer susceptibility to PD, the comparison of serum iron levels between Parkinson's patients and controls has contradictory results [1,19,19–22,22–26]. Similarly, the relationship of PD with polymorphisms in C282Y or H63D in the *HFE* gene, which encodes a protein responsible for regulating iron levels in liver cells and hepcidin production, also remains controversial. However, some data point to a protective effect of the HFE-C282Y polymorphism [25]. Low serum zinc levels have been also described as a risk factor for PD [24].

Exercise is a known protective factor for PD, probably due to the increased of serum urate. Higher serum urate levels have been shown to decrease the risk of PD and improve its prognosis in men. The same relationship remains controversial in females [11,16,26–29]. Urate is a product of purine metabolism, and it seems to have an antioxidant effect through Nrf2/ARE (antioxidant responsive element) pathway activation, protecting against the loss of dopaminergic neurons [16,26]. The Nrf2/ARE pathway has already been indicated as a possible therapeutic target in neurodegenerative pathology, since it is responsible for regulating, in a constitutional and induced manner, the expression of proteins essential to cellular protection against oxidative stress. This pathway is also capable of improving mitochondrial biogenesis [27,30]. Thus, physical exercise, and thereby raise of urate, has a possible dual beneficial effect on PD by preventing mitochondrial dysfunction and oxidative stress [11,16,26–28].

Coffee consumption is linked to a lower PD risk, this relationship is most evident in men [16,18,26,31]. The coffee´s protective effects are thought to be mostly due to caffeine. This hypothesis is strengthened by the fact that other sources of caffeine, such as teas

and soft drinks, demonstrate the same effect. Additionally decaffeinated coffee consumption has not been shown to reduce the risk of PD [26,32]. Among women, the decreased risk is controversial, probably because estrogens and caffeine are metabolized by the same enzyme, CYP1A2 [16,31,32].

Caffeine acts as a competitive adenosine receptors antagonist, having higher affinity for A_1R and $A_{2A}R$. Chronic A_1R blockade by caffeine is known to lead to upregulation of these receptors, resulting in pro-inflammatory cytokines decrease. $A_{2A}R$ inhibition has anti-inflammatory and anti-apoptotic effects. By this route, caffeine prevent adenosine-mediated neuroinflammatory processes by decreasing microglia reactivity, glutamate release and pro-inflammatory cytokines release [16,32–34].

Smoking decreases the PD risk. The hypothesis of a mortality bias associated with smoking has been refuted. It is recognized that its protective effect is greater in men and among long-term smokers [16,18,35]. A PD premorbid personality was described, indeed PD patients that score high on neuroticism, have a greater risk aversion and lower sensation-seeking scores [18,36,37], therefore they are thought to be less prone to smoking. However, this behavior does not fully explain the protective effect found. Consequently, the mechanisms inherent in this relationship are still under scrutiny [11,13,16,18,35,38].

The protective effect of smoking is believed to be due to nicotine, since it inhibits ∝-synuclein fibrils formation and reduces the production of free radicals by monoamine oxidase B (MAO B). On the other hand, MAO B metabolizes dopamine, and it is involved in the production of inflammatory cytokines, as well as, apoptosis [11,13,16,18,35,38].

2.2. Patient Susceptibility.

Advanced age is the greatest known risk factor for PD. However, other individual intrinsic characteristics are potential risk factors. There is a higher incidence of PD in males. Being a male is a risk factor, as well as, a worse prognostic factor [7,16,26]. Male patients have a slightly increased mortality compared to female patients [39].

The presentation and age of disease onset also differ between genders. Women have a later MS onset and more benign progression [7]. Nevertheless, symptoms such as fatigue, depression, anxiety, constipation, pain, hypo/anosmia, hyperhidrosis, and

propensity for severe dysphagia are more common and severe in women. Nonetheless, the higher incidence of depression, anxiety and pain in women is not specific to PD. Women are also more likely to develop postural instability, motor complications and hallucinations due to iatrogenic symptomatic therapy [7,29].

Men have greater cognitive impairment, greater sexual dysfunction, severe REM sleep disturbances, association with impulse control disorder and severe sialorrhea [7,29].

Sex dissimilarities may depend on structural differences, such as a higher ratio of D1:D2 dopaminergic receptors in females in the dorsal and ventral *striatum*, but also on the contrasting hormonal background. [29] In an animal model of PD, sex differences are blurred after an oophorectomy. [40] Estradiol seems to have a neuroprotective effect by a pleiotropic action on the CNS. Its interaction with Insulin-Like Growth Factor-I (IGF-I) has been demonstrated [41], as well as, its ability to modulate cell death by inducing expression of Bcl-2 and Bcl-w (anti-apoptotic proteins) and decreasing Bad and Bim (apoptotic proteins). Estradiol is also thought to increase the synthesis, release and reuptake of dopamine, as well as, decrease the production of reactive oxygen species (ROS), improving mitochondrial function [42,43].

Having a family history of PD is also a known risk factor, although the monogenic form is a minority of diagnoses [18]. However, to describe PD as monogenic or sporadic is a simplification of the genetic contribution to the condition development. In reality there is a spectrum of genetic mutations with varying degrees of penetrance, which again emphasizes the interaction between individual susceptibility factors and the external environment [44,45]. At this spectrum we have high penetrance mutations responsible for PD familial forms and low penetrance mutations found in sporadic forms [45,46]. An example is that individuals with LRRK2 locus mutation, that did not develop PD, have higher serum urate values [29]. Note that, as previously discussed, the relationship of serum urate levels with PD is not equal in both genders, it is also known that the phenotypic expression of these mutations may be dependent on the sex of the patient [16,26,47].

DNA methylation levels may also influence a patient's vulnerability to pathology. In blood and saliva samples from patients, co-methylation modules in genes associated with mitochondrial protein coding or involved in fighting oxidative stress, such as *LARS2*, *MIR1977* and *DDAH2*, have been found to be associated with PD as well [48].

3. The starting point: The Gut of a Parkinson's patient.

The relationship between the gut and the human brain opened new opportunities to understand and perhaps explain neurological pathology, this is especially true in PD [49,50].

The human gut is the home of bacteria, viruses and fungi that together form the microbiome. In turn, the microbiome is essential for the development and maturation of the host Nervous System (NS), Immune System (IS) and Endocrine System (ES)[49]. It is now known that the microbiome composition is vulnerable to the complexity and interaction of the host with the external environment in a way that is not limited to diet or physical exercise [51]. Thus, the gut has a microbiome that is sensitive to neurotransmitters and therefore susceptible to information transmitted by the CNS. However, the microbiome is also able to influence the CNS by producing metabolites with tropism for the CNS, including neurotransmitters (serotonin, dopamine and GABA), neuromodulators (short chain fatty acids - SCFAs) or other substances such as histamine and tryptophan [52].

The association between PD and the gut arises from careful observation of the disease natural history. It has been hypothesized that the gut is the starting point of PD [2,28,52-54] since GI symptoms precede MS [7,55]. Also, the Lewy bodies presence in the gut precedes their appearance in the CNS [53,56], and the relationship of PD with intestinal inflammation is documented by the fact that patients with inflammatory bowel disease (IBD) have a higher risk of developing PD [53].

In 2007, Braak proposed the dual-hit Hypothesis which advocates the olfactory bulb and the gut as entry points for an unknown infectious agent capable of reaching the CNS and triggering PD [57]. Other PD pathophysiological hypotheses such as Johnson et al. and Cardoso & Empadinhas hypotheses emphasize gut dysbiosis as the starting point of at least some sporadic PD cases (Figure 1) [1,2,28,58].

Several cohort studies have been investigating the gut microbiome composition in Parkinson's patients (Table 1). As a result, although there are some common points in the different studies, there are also discordant points (Figure 2), and their significance is still uncertain, given the disparity in sample size, clinical presentation and stage of pathology progression of the selected patients [54–61]. Still, the changes found in the

microbiome of Parkinson's patients corroborate both hypotheses stating gut dysbiosis as a PD starting point [53,55,59,60].

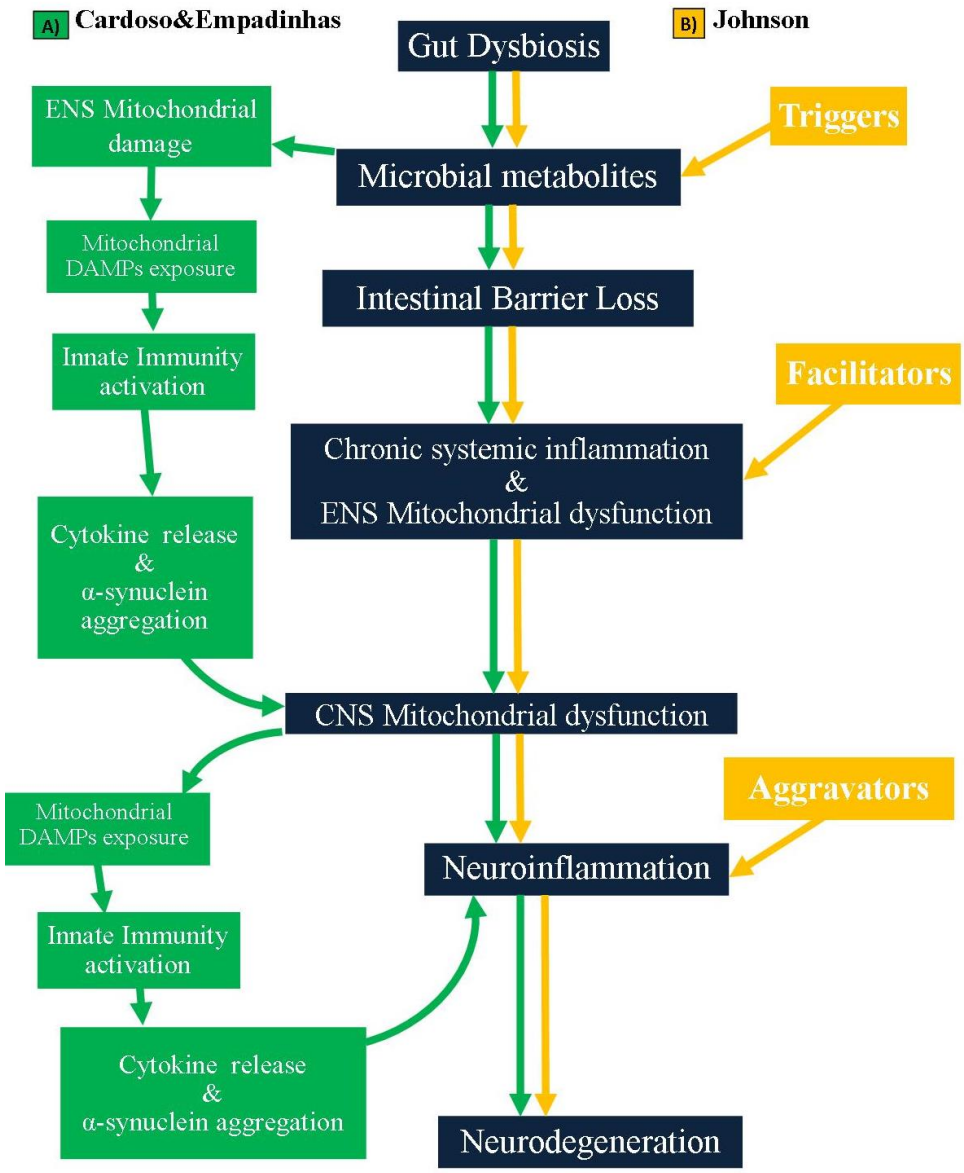

Figure 1- Schematic illustration of the Johnson et al. and Cardoso & Empadinhas hypothesis on gut dysbiosis as a starting point in PD. A) The Cardoso & Empadinhas hypothesis, proposes that the toxins produced by the microbiome, in the context of gut dysbiosis, leads to mitochondrial damage. Consequently, the release of mitochondrial DAMPs leads to neuronal sterile inflammation, via activation of Toll-like receptors (TLRs) and Nod-like receptors (NLRs). Ultimately, this inflammation leads to neurodegeneration of the ENS and CNS [28]; **B)** Johnson and colleagues describes 3 phases, which include: *triggers*, *facilitators* and *aggravators*. Triggers act transiently, e.g. trauma and exposure to toxins or pathogens, and although they are insufficient to generate the disease they allow it to develop in the presence of facilitators. In turn, facilitators, such as mitochondrial dysfunction or gene mutations associated with PD, are conditions that precede or are concomitant to triggers, allowing CNS involvement and lead to a state of chronic systemic inflammation. Finally, aggravators include changes in autophagy and neuroinflammation that are associated with the progression of the pathology [2].

It is still unclear what the etiology beyond PD gut dysbiosis might be. Since PD is so closely correlated with aging, one can point to its association with age-related dysbiosis. With aging there is an increase in proteobacteria and decrease in probiotics that translates into a decrease in SCFAs as a result of microbiome functional loss [52,62]. Aging is also associated with a chronic pro-inflammatory state (inflammaging) [63].

Exposure to antibiotics represents another possible cause of gut dysbiosis, but its relation to neurodegenerative diseases is ambiguous. These drugs lead to a decrease in the microbiome biodiversity and therefore lead to dysbiosis. However, some antibiotics have shown neuroprotective and anti-inflammatory effects [52]. In this sense, although some studies have found no association between antibiotic intake and the incidence of PD [64], others have associated the broad-spectrum penicillins consumption with a higher prevalence of PD [65], as well as a higher risk of PD associated with exposure to macrolites and lincosamides [66]. Rifampicin demonstrated, in a cell model, neuroprotective properties by reducing mitochondria-mediated oxidative stress, decreasing microglia activation, and suppressing expression of α-synuclein aggregates [67]. Tetracyclines, namely doxycycline and minocycline, are also thought to possess neuroprotective properties that may have utility in neurodegenerative diseases such as PD. Namely by their anti-inflammatory, anti-apoptotic and free radical scavenging actions that contribute to a decrease in mitochondrial dysfunction and a reduction in microglia activation [68,69]. Finally, it has been theorized that microbiome-derived toxins have a contribution to the development of neurodegenerative diseases, since endotoxins such as LPS can lead to mitochondrial dysfunction and neuroinflammation, both processes widely associated with PD [2,28,70,71]. An example is the neurotoxin β-Methylamino- l-alanine (BMAA), a non-proteinogenic amino acid produced by ubiquitous cyanobacteria [72–74], when in nitrogen deprivation [75]. BMAA was primarily associated with the Amyotrophic lateral sclerosis/parkinsonism-dementia complex of Guam [76]. Since then, BMAA has been hypothesized to contribute to the development of neurodegenerative diseases in susceptible individuals [75,77]. When present in high concentrations, this toxin was first shown to trigger excitotoxicity via NMDA receptors. However, given the suspicion that its contribution to the neurodegenerative process reflects a chronic exposure at low concentrations, other mechanisms of toxicity have been investigated [77,78]. Consequently, BMAA has been linked to mitochondrial dysfunction and fragmentation, as well as, to the production of inflammatory molecules by neurons, which in turn leads to the activation of microglia.

Table 1- comparison of eight cohort studies analyzing the microbiome of Parkinson's patients

Author (year)	Sample	sequencing	Increased in PD Family(genus)	Decreased in PD Family(genus)
Unger, et al (2016) [54]	Fecal samples (PD n=34; Controls n=34)	16S rRNA gene	• *Enterobacteriaceae*	• *Prevotellaceae*
Scheperjns, et al (2015) [55]	Fecal samples (PD n=72; Controls n=72)	V1-V3 regions 16S rRNA gene.	• *Enterobacteriaceae*	• *Prevotellaceae*
Qian, et al (2018)[56]	Fecal samples (PD n=45; Controls n=45)	V3-V4 region of 16S rRNA gene	• *Rikenellaceae (Alistipes),* • *Prevotellaceae (Paraprevotella)* • *Enterobacteriaceae (Klebsiella)* • *Sphingomonadaceae (Sphingomonas)* • *Moraxellaceae (Acinetobacter)* • *Comamonadaceae (Aquabacterium)* • *Desulfovibionaceae (Desulfovibrio)* • *Clostridiaceae (Clostridium)* • *Lachnospiraceae* • *Oscillospiraceae (Butyricicoccus)* • *Nitrososphaeraceae (Nitrososphaera)*	• *Lactobacillaceae (Lactobacillus)* • *Chitinophagaceae (Sediminibacterium)*
Petrov, et al. (2017)[57]	Fecal Samples (PD n=89; Controls n=66)	16S rRNA gene	• *Christensenellaceae (Christensenell)* • *Catabacteraceae (Catabacter)* • *Lactobacillaceae (Lactobacillu)* • *Oscillospiraceae (Oscillospira, Ruminococcus, Papillibacter)* • *Bifidobacteriaceae (Bifidobacterium)*	• *Lachnospiraceae (Dorea Blautia glucerasea, Coprococcus Eutactus)* • *Bacteroidaceae (Bacteroides)* • *Prevotellaceae (Prevotella)* • *Oscillospiraceae (Faecalibacterium, Ruminococcus callidus)*
Baldini, et al. (2020)[58]	Fecal samples (PD n=147; Controls n=162)	16S rRNA gene	• *Verrucomicrobiaceae (Akkermansia.)* • *Lactobacillaceae* • *(Lactobacillus)*	• *Turicibacteraceae (Turicibacter)* • *Prevotellaceae (Paraprevotella - female PD)*
Lin, et al. (2019)[59]	Fecal samples (PD n=80; Controls n=77)	V3-V4 region of the 16S rRNA gene	• *Deferribacteraceae (Mucispirillum),* • *Porphyromonadaceae (Porphyromonas)* • *Lactobacillaceae (Lactobacillus),* • *Tannerellaceae (Parabacteroides)*	• *Prevotellaceae (Prevotella)*
Li, et al (2017)[60]	Fecal samples (PD n=24; Controls n=14)	16S rRNA gene	• *Enterobacteriaceae (Escherichia, Shigella, Enterococcus)* • *Streptococcaceae (Streptococcus Proteus)*	• *Lachnospiraceae (Blautia)* • *Oscillospiraceae (Faecalibacterium, Ruminococcus)*
Hill-Burns E. et al (2017)[61]	Fecal samples (PD n=197; Controls n=130)	16S rRNA gene	• *Verrucomicrobiaceae (Akkermansia)* • *Lactobacillaceae (Lactobacillus)* • *Bifidobacteriaceae (Bifidobacterium)*	• *Lachnospiraceae*

.

BMAA is able to lead to NLRP3 inflammasome activation by an NF-kB dependent pathway, namely due to cardiolipin exposure that functions as a mitochondrial danger associated molecular pattern (DAMP), and increases the TLR receptors expression on the neurons [71]. Thus, it is hypothesized that gut chronic exposure to BMAA may lead to a pro-inflammatory intestinal state and to enteric mitochondrial dysfunction that in a susceptible individual, will lead to ENS and then to CNS neurodegeneration [77]. This is not only in line with the Cardoso & Empadinhas hypothesis [28] previously discussed, but is also supported by BMAA identification in water reserves [72–74]. Additionally, BMAA can progress between neurons and seems to have tropism for motor neurons or NADPH- diaphorase positive neurons, in which its transport is carried out retrogradely [78].

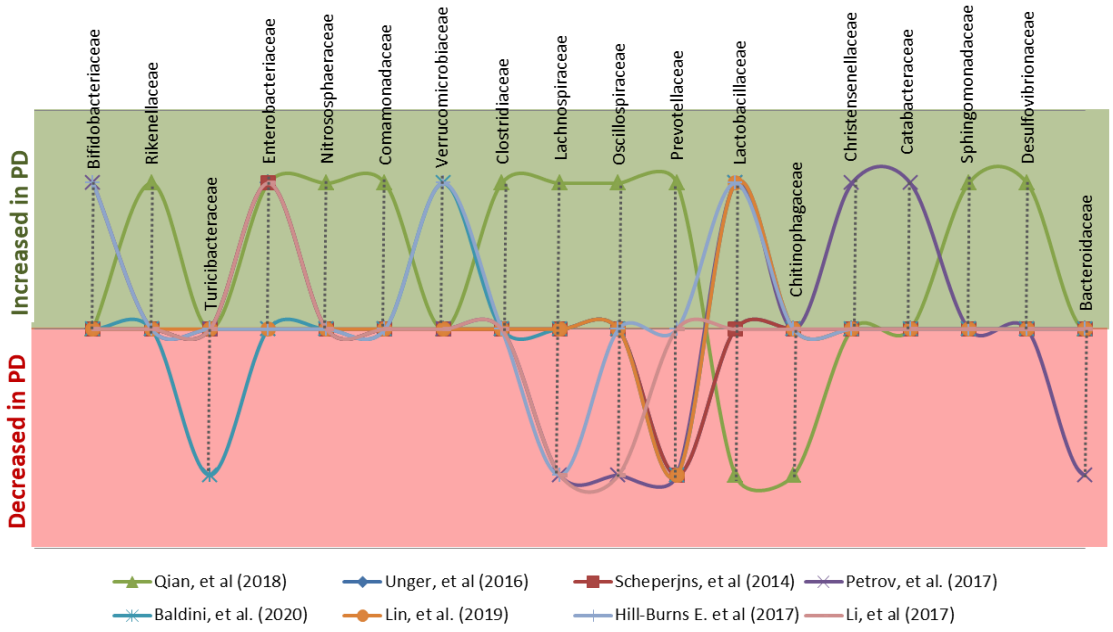

Figure 2- Illustrative summary of the results presented in the eight cohort studies consulted. The increase of Enterobacteriaceae in four studies is noteworthy. A decrease in Prevotellaceae in Parkinson's disease patients was also reported in five of the studies, although one study reported an increase. Four of the studies showed an increase in the Lactobacillaceae family, but one study found its decrease in PD. Expression of the Lachnospiraceae family is decreased in Parkinson's patients in three studies, with one study coming to opposite conclusions.

4. The first obstacle: from the gut to the rest of the body.

The GI boundary of the "*self*" is conferred by a single layer of intestinal epithelial cells (IEC) interconnected by tight junctions (TJ) and covered by mucus produced by goblet cells, into which paneth cells secrete immunoglobulin A (IgA) and antimicrobial peptides. By itself, the intestinal barrier provides mechanical protection and selective permeability to water and ions [79,80].

It is up to the IS to discern which stimuli pose as a survival threat of the "*self*", without triggering unnecessary sterile colitis, in the face of the gut microbial population, as well as, the constant food passageway [79,81]. The shaping of this immune response remains to be understood in a way, but communication between IECs and resident macrophages is thought to be a highlight to this mystery. IECs stimulation by the presence of the microbiome, via Toll-Like 4 receptors (TLR4), induces Interleukin-10 (IL-10) expression. In sequence, IL-10 is known to have anti-inflammatory properties and is essential for maintaining the integrity of the epithelium. On the other hand, IL-10 expression in IEC is enhanced by resident macrophages signaling [82]. In turn, the gut inflammatory status itself is responsible for shaping the phenotype acquired by resident macrophages [83].

CX3CR1+ resident macrophages are a heterogeneous group with high adaptive capacity, which concomitantly express IL-10 and TNF-α, while maintaining an alert resting state [79]. Through the transepithelial extensions, these macrophages are sensitive to the intraluminal environment and respond phenotypically accordingly. In case of threat they acquire a proinflammatory phenotype, communicate with dendritic cells, so that they recruit the action of adaptive immunity, or switch themselves to a phenotype similar to that of a dendritic cell, assuming similar roles [83].

The intestinal mucosa permeability degree depends on different stimuli and components. The microbiome is able to shape this barrier, increasing its permeability by inducing miR-21-5p expression in epithelial cells [84]. This is a curious finding given that inhibition of miR-21-5p in an animal model of Ulcerative Colitis, was able to decrease inflammation, as well as, cell apoptosis through the IL-6/STAT3 pathway, demonstrating to be a possible therapeutic target against intestinal inflammation [85]. On the other hand, excessive proteolysis results in increased permeability in the intestinal epithelium [86]. Finally, when the integrity of TJs is compromised, an increase in intestinal permeability occurs inducing local and systemic inflammation [87].

Intestinal epithelial barrier disruption and increased permeability have been demonstrated in several pathologies, notably in the context of hyperglycemia, but also in PD [88,89]. Upon administration of *Proteus mirabilis* and purified LPS from *P. mirabilis,* to a PD animal model, it was shown dopaminergic neuronal loss in the substantia *nigra*, neuroinflammation, and aggregation of α-synuclein in the colon and the brain [90]. These findings are endorsed by an increase in calprotectin, α1-antitrypsin and zonulin (the first one a marker of intestinal inflammation, and the latter two are markers of increased intestinal permeability) in the Parkinson's patients fecal material. [91]. Thus, an increase in intestinal permeability in PD may be associated with a systemic pro-inflammatory state, which ultimately will translate into CNS changes compatible with the motor disturbances documented in PD [28,92,93]. The serological increase of pro-inflammatory cytokines such as NT-proCNP and TNF-α in Parkinson's patients supports this hypothesis [94]. A serological increase in anti-inflammatory cytokines has also been demonstrated, which is thought to be due to compensatory mechanisms [94,95]. In the plasma of patients, an increase in IL-1β and α-synuclein that correlates positively with the motor condition severity, has also been found [96]. This findings highlights the PD systemic involvement and, may one day, be used as disease biomarker [2,96–98].

The topography of the α-synuclein aggregates propagation is highly suggestive of the pathway taken by pathological processes [99]. Increased permeability of the intestinal mucosa is known to correlate positively with the α-synuclein aggregates localization [100]. And, although elevated α-synuclein levels in the mucosa muscularis or submucosa is not a PD-specific outcome, this is considered a quite suggestive PD histological finding, being Lewy pathology detected more often in colon biopsies than in rectal biopsies [101].

The multiorgan dysfunction in PD becomes even more evident if we focus on changes in the blood's elements, particularly among leukocytes. Platelets structural alterations, resulting in increased platelet activation and hypercoagulation, have been found in Parkinson's patients [102]. Deficits in the energy function of blood cells have also been found, with blood cells showing increased vulnerability to oxidative stress, increased glycolysis, mitochondrial dysfunction, and lower glutathione peroxidase activity in early stages of the disease [103,104]. This demonstrates that the mitochondrial dysfunction described in PD is not limited to neurons [105]. Parkinson's patients blood cells express α-synuclein, and it is possible to differentiate between controls and patients base on that [98]. NLRP3 inflammasome activation has also been described in mononuclear blood cells of patients [96].

Finally, several studies have demonstrated lymphoproliferative response alterations in Parkinson's patients at very early disease stages. Thus, a decrease in T cells CD4+ and a Th1 bias have been described. Notice that Th1 is considered a pro-inflammatory phenotype [3,4,106]. Dysfunction in T_{reg} cells, which contributes to a pro-inflammatory microenvironment, may also be attributed to the disease [106]. α-Synuclein specific T cells have also been identified at early stages of the disease, which may demonstrate an autoimmune component associated with PD [95].

5. The final hurdle: BBB and Parkinson's disease.

It is believed that pathological processes, initiated in the gut and possibly triggering PD, may reach the brain by different routes, either through the vagus nerve or through the circulatory system [2]. This perspective is supported by the α-synuclein identification in the vagus nerve dorsal motor nucleus in early stages [92] and by the protective effect that full truncal vagotomy confers to PD [107].

The CNS is immunologically privileged due to the existence of the BBB. A disruption of this barrier translates into a greater vulnerability of the CNS to the external environment [108]. It is known that there is a positive correlation between the appearance of white matter lesions and systemic inflammation in PD [106,109]. On the other hand, advanced age, in itself, is already a factor of increased vulnerability of the BBB. With aging there is an increase in this barrier permeability [110]. The elderly have a decrease number of endothelial cells available and a decrease in capillary density. Also, it has been demonstrated in elderly brains an IgG, IgA and IgM leakage. In addition the involvement of the BBB in PD has been likewise demonstrated [111]. Dysfunction of TJs and transporters such as P-glycoprotein has been shown to be associated with aging and neurodegenerative diseases [111]. LPS administration, in an MPTP model of PD, led to the expression of A1 astrocytes, which are considered neurotoxic, death of *substantia nigra* dopaminergic neurons , increased permeability of the BBB and galectin-3 microglia expression [108]. This microglia galectin-3 positive phenotype is a surveillance state previously identified in the context of cerebral ischemia [112].

Microglia performs a dual behavior concerning peripheral inflammation. During the acute peripheral inflammation phase microglia contribute to BBB integrity by migrating towards to the vasculature and producing claudin-5, which helps maintain TJs between

endothelial cells. But if the inflammation is sustained and becomes a chronic systemic inflammation, microglia adopt a phagocytic phenotype that contributes to the increase permeability of this barrier [113].

Following LPS administration, high α-synuclein expression lead to an increase in BBB permeability [114]. Therefore intestinal pathological processes translocation to the CNS is intrinsically linked to the α-synuclein intercellular and interregional migration capacity, which is consequently believed to have a prion-like behavior in PD [115]. Its relationship with the BBB is also quite peculiar, since its passage occurs bidirectionally and it is mediated by the LRP-1 receptor. This shows that α-synuclein, formed in the gut in response to an initial local inflammation, is able to reach the CNS by routes other than the vagus nerve [116,117].

The BBB seems to be especially vulnerable in the *striatum*. It has been shown in *postmortem* samples a significant increase in BBB permeability at the posterior commissural *putamen* in the *striatum* of Parkinson's patients [118].

Although BBB disruption possibly plays a major role in the progression of PD, there are laboratories that see this vulnerability as an opportunity to optimize the currently available therapies efficacy [119].

6. Conclusion

Currently, the Parkinson's patient is understood as a genetically susceptible individual, inserted at a pathology favorable environment, subjected to various triggers, facilitators, and aggravators of the disease [2]. As a result there is a wider involvement of the organism than previously speculated. This perspective brings new challenges, but also new opportunities for management and treatment of the disease in the future [106]. In view of the absence of a disease-modifying therapy and an ineffective therapy for symptomatic non-motor manifestations, the full contemplation of the Parkinson's disease and the Parkinson's patient may indicate new paths to follow [6,119].

This review concludes that there is strong evidence of systemic involvement in PD and a predominant role of the disruption of biological mechanical barriers, such as the intestinal barrier and the BBB, in the progression and development of the pathology [94,104,108,114].

7. Bibliography

1. Nandipati S, Litvan I. Environmental Exposures and Parkinson's Disease. Int J Env Res Public Health. 2016;19.

2. Johnson ME, Stecher B, Labrie V, Brundin L, Brundin P. Triggers, Facilitators, and Aggravators: Redefining Parkinson's Disease Pathogenesis. Trends Neurosci. 2019 Jan;42(1):4–13.

3. Kustrimovic N, Comi C, Magistrelli L, Rasini E, Legnaro M, Bombelli R, et al. Parkinson's disease patients have a complex phenotypic and functional Th1 bias: cross-sectional studies of CD4+ Th1/Th2/T17 and Treg in drug-naïve and drug-treated patients. J Neuroinflammation. 2018 Dec;15(1):205.

4. Chen Z, Chen S, Liu J. The role of T cells in the pathogenesis of Parkinson's disease. Prog Neurobiol. 2018 Oct;169:1–23.

5. Postuma RB, Berg D, Stern M, Poewe W, Olanow CW, Oertel W, et al. MDS clinical diagnostic criteria for Parkinson's disease: MDS-PD Clinical Diagnostic Criteria. Mov Disord. 2015 Oct;30(12):1591–601.

6. Schapira AHV, Chaudhuri KR, Jenner P. Non-motor features of Parkinson disease. Nat Rev Neurosci. 2017 Jul;18(7):435–50.

7. Marinus J, Zhu K, Marras C, Aarsland D, van Hilten JJ. Risk factors for non-motor symptoms in Parkinson's disease. Lancet Neurol. 2018 Jun;17(6):559–68.

8. Lees AJ, Hardy J, Revesz T. Parkinson's disease. 2009;373:12.

9. Cersosimo MG, Benarroch EE. Pathological correlates of gastrointestinal dysfunction in Parkinson's disease. Neurobiol Dis. 2012 Jun;46(3):559–64.

10. Campdelacreu J. Enfermedad de Parkinson y enfermedad de Alzheimer: factores de riesgo ambientales. 2014:9.

11. Bellou V, Belbasis L, Tzoulaki I, Evangelou E, Ioannidis JPA. Environmental risk factors and Parkinson's disease: An umbrella review of meta-analyses. Parkinsonism Relat Disord. Fevereiro de 2016;23:1–9.

12. Delamarre A, Meissner WG. Epidemiology, environmental risk factors and genetics of Parkinson's disease. Presse Médicale. 2017 Mar;46(2):175–81.

13. Breckenridge CB, Berry C, Chang ET, Sielken RL, Mandel JS. Association between Parkinson's Disease and Cigarette Smoking, Rural Living, Well-Water Consumption, Farming and Pesticide Use: Systematic Review and Meta-Analysis. Guedes RNC, editor. PLOS ONE. 2016 Apr 7;11(4):e0151841.

14. Pouchieu C, Piel C, Carles C, Gruber A, Helmer C, Tual S, et al. Pesticide use in agriculture and Parkinson's disease in the AGRICAN cohort study. Int J Epidemiol. 2018 Feb 1;47(1):299–310.

15. Schneider Medeiros M, P. Reddy S, P. Socal M, Schumacher-Schuh AF, Mello Rieder CR. Occupational pesticide exposure and the risk of death in patients with Parkinson's disease: an observational study in southern Brazil. Environ Health. 2020 Dec;19(1):68.

16. Ascherio A, Schwarzschild MA. The epidemiology of Parkinson's disease: risk factors and prevention. Lancet Neurol. 2016 Nov;15(12):1257–72.

17. Palin O, Herd C, Morrison KE, Jagielski AC, Wheatley K, Thomas GN, et al. Systematic review and meta-analysis of hydrocarbon exposure and the risk of Parkinson's disease. Parkinsonism Relat Disord. 2015 Mar;21(3):243–8.

18. Noyce AJ, Bestwick JP, Silveira-Moriyama L, Hawkes CH, Giovannoni G, Lees AJ, et al. Meta-analysis of early nonmotor features and risk factors for Parkinson disease. Ann Neurol. 2012 Dec;72(6):893–901.

19. Bergsland N, Zivadinov R, Schweser F, Hagemeier J, Lichter D, Guttuso T. Ventral posterior substantia nigra iron increases over 3 years in Parkinson's disease. Mov Disord. 2019 Jul;34(7):1006–13.

20. An H, Zeng X, Niu T, Li G, Yang J, Zheng L, et al. Quantifying iron deposition within the substantia nigra of Parkinson's disease by quantitative susceptibility mapping. J Neurol Sci. 2018 Mar;386:46–52.

21. Green AJ, Planchart A. THE NEURODEVELOPMENTAL TOXICITY OF HEAVY METALS: A FISH PERSPECTIVE. 2019;17.

22. Mostile G, Cicero CE, Giuliano L, Zappia M, Nicoletti A. Iron and Parkinson's disease: A systematic review and meta-analysis. Mol Med Rep. 2017 May;15(5):3383–9.

23. Mariani S, Ventriglia M, Simonelli I, Donno S, Bucossi S, Vernieri F, et al. Fe and Cu do not differ in Parkinson's disease: A replication study plus meta-analysis. 2013:2.

24. Sun H, Liu X, Ge H, Wang T, Wang Y, Li W. Association Between Serum Zinc Levels and the Risk of Parkinson's Disease: a Meta-Analysis. Biol Trace Elem Res. 2017 Sep;179(1):45–51.

25. Xia J, Xu H, Jiang H, Xie J. The association between the C282Y and H63D polymorphisms of HFE gene and the risk of Parkinson's disease: A meta-analysis. Neurosci Lett. 2015 May;595:99–103.

26. Boulos C, Yaghi N, Hayeck RE, Heraoui GN, Fakhoury-Sayegh N. Nutritional Risk Factors, Microbiota and Parkinson's Disease: What Is the Current Evidence? 2019;24.

27. Nguyen T, Nioi P, Pickett CB. The Nrf2-Antioxidant Response Element Signaling Pathway and Its Activation by Oxidative Stress. J Biol Chem. 2009 May 15;284(20):13291–5.

28. Cardoso SM, Empadinhas N. The Microbiome-Mitochondria Dance in Prodromal Parkinson's Disease. Front Physiol. 2018 May 9;9:471.

29. Cerri S, Mus L, Blandini F. Parkinson's Disease in Women and Men: What's the Difference? 2019;15.

30. Vomhof-DeKrey EE, Picklo MJ. The Nrf2-antioxidant response element pathway: a target for regulating energy metabolism. J Nutr Biochem. 2012 Oct;23(10):1201–6.

31. Kim IY, O'Reilly ÉJ, Hughes KC, Gao X, Schwarzschild MA, Ascherio A. Differences in Parkinson's Disease Risk with Caffeine Intake and Postmenopausal Hormone Use. J Park Dis. 2017 Nov 1;7(4):677–84.

32. Kolahdouzan M, Hamadeh MJ. The neuroprotective effects of caffeine in neurodegenerative diseases. CNS Neurosci Ther. 2017 Apr;23(4):272–90.

33. Lopes JP, Pliássova A, Cunha RA. The physiological effects of caffeine on synaptic transmission and plasticity in the mouse hippocampus selectively

depend on adenosine A1 and A2A receptors. Biochem Pharmacol. 2019 Aug;166:313–21.

34. Sebastião AM, Ribeiro JA. Neuromodulation and metamodulation by adenosine: Impact and subtleties upon synaptic plasticity regulation. Brain Res. 2015 Sep;1621:102–13.

35. Nag N, A. Jelinek G. A Narrative Review of Lifestyle Factors Associated with Parkinson's Disease Risk and Progression. Neurodegener Dis. 2019 Sep;(19):9.

36. Poletti M, Bonuccelli U. Personality traits in patients with Parkinson's disease: assessment and clinical implications. J Neurol. 2012 Jun;259(6):1029–38.

37. Santangelo G, Piscopo F, Barone P, Vitale C. Personality in Parkinson's disease: Clinical, behavioural and cognitive correlates. J Neurol Sci. 2017 Mar;374:17–25.

38. Li X. Association between cigarette smoking and Parkinson's disease: A meta-analysis. Arch Gerontol Geriatr. 2015;7.

39. Hirsch L, Jette N, Frolkis A, Steeves T, Pringsheim T. The Incidence of Parkinson's Disease: A Systematic Review and Meta-Analysis. Neuroepidemiology. 2016;46(4):292–300.

40. Chen Y-H, Wang V, Huang EY-K, Chou Y-C, Kuo T-T, Olson L, et al. Delayed Dopamine Dysfunction and Motor Deficits in Female Parkinson Model Mice. Int J Mol Sci. 2019 Dec 11;20(24):6251.

41. Garcia-Segura LM, Sanz A, Mendez P. Cross-Talk between IGF-I and Estradiol in the Brain: Focus on Neuroprotection. 2006:5.

42. Ishihara Y, Sakurai H, Oguro A, Tsuji M, Vogel CFA, Yamazaki T. Retinoid X receptor-mediated neuroprotection via CYP19 upregulation and subsequent increases in estradiol synthesis. J Steroid Biochem Mol Biol. 2019 Oct;193:105421.

43. Guo J, Duckles SP, Weiss JH, Li X, Krause DN. 17β-Estradiol prevents cell death and mitochondrial dysfunction by an estrogen receptor-dependent mechanism in astrocytes after oxygen–glucose deprivation/reperfusion. Free Radic Biol Med. 2012 Jun;52(11–12):2151–60.

44. Billingsley KJ, Bandres-Ciga S, Saez-Atienzar S, Singleton AB. Genetic risk factors in Parkinson's disease. Cell Tissue Res. 2018 Jul;373(1):9–20.

45. Blauwendraat C, Nalls MA, Singleton AB. The genetic architecture of Parkinson's disease. Lancet Neurol. 2020 Feb;19(2):170–8.

46. Kim C, Alcalay R. Genetic Forms of Parkinson's Disease. Semin Neurol. 2017 May 16;37(02):135–46.

47. San Luciano M, Wang C, Ortega RA, Giladi N, Marder K, Bressman S, et al. Sex differences in LRRK2 G2019S and idiopathic Parkinson's Disease. Ann Clin Transl Neurol. 2017 Nov;4(11):801–10.

48. Chuang Y-H, Paul KC, Bronstein JM, Bordelon Y, Horvath S, Ritz B. Parkinson's disease is associated with DNA methylation levels in human blood and saliva. Genome Med. 2017 Dec;9(1):76.

49. Fung TC, Olson CA, Hsiao EY. Interactions between the microbiota, immune and nervous systems in health and disease. Nat Neurosci. 2017 Feb;20(2):145–55.

50. Rinninella E, Raoul P, Cintoni M, Franceschi F, Miggiano G, Gasbarrini A, et al. What is the Healthy Gut Microbiota Composition? A Changing Ecosystem across Age, Environment, Diet, and Diseases. Microorganisms. 2019 Jan 10;7(1):14.

51. Singh Y, El-Hadidi M, Admard J, Wassouf Z, Schulze-Hentrich JM, Kohlhofer U, et al. Enriched Environmental Conditions Modify the Gut Microbiome Composition and Fecal Markers of Inflammation in Parkinson's Disease. Front Neurosci. 2019 Oct 15;13:1032.

52. Angelucci F, Cechova K, Amlerova J, Hort J. Antibiotics, gut microbiota, and Alzheimer's disease. J Neuroinflammation. 2019 Dec;16(1):108.

53. Brudek T. Inflammatory Bowel Diseases and Parkinson's Disease. van Laar T, editor. J Park Dis. 2019 Oct 30;9(s2):S331–44.

54. Unger MM. Short chain fatty acids and gut microbiota differ between patients with Parkinson's disease and age-matched controls. 2016;7.

55. Scheperjans F, Aho V, Pereira PAB, Koskinen K, Paulin L, Pekkonen E, et al. Gut microbiota are related to Parkinson's disease and clinical phenotype. Mov Disord. 2015 Mar;30(3):350–8.

56. Qian Y, Yang X, Xu S, Wu C, Song Y, Qin N, et al. Alteration of the fecal microbiota in Chinese patients with Parkinson's disease. Brain Behav Immun. 2018 May;70:194–202.

57. Petrov VA, Saltykova IV, Zhukova IA, Alifirova VM, Zhukova NG, Dorofeeva YuB, et al. Analysis of Gut Microbiota in Patients with Parkinson's Disease. Bull Exp Biol Med. 2017 Apr;162(6):734–7.

58. on behalf of the NCER-PD Consortium, Baldini F, Hertel J, Sandt E, Thinnes CC, Neuberger-Castillo L, et al. Parkinson's disease-associated alterations of the gut microbiome predict disease-relevant changes in metabolic functions. BMC Biol. 2020 Dec;18(1):62.

59. Chiang H-L, Lin C-H. Altered Gut Microbiome and Intestinal Pathology in Parkinson's Disease. J Mov Disord. 2019 May 31;12(2):67–83.

60. Li W, Wu X, Hu X, Wang T, Liang S, Duan Y, et al. Structural changes of gut microbiota in Parkinson's disease and its correlation with clinical features. Sci China Life Sci. 2017 Nov;60(11):1223–33.

61. Hill-Burns EM, Debelius JW, Morton JT, Wissemann WT, Lewis MR, Wallen ZD, et al. Parkinson's disease and Parkinson's disease medications have distinct signatures of the gut microbiome: PD, Medications, and Gut Microbiome. Mov Disord. 2017 May;32(5):739–49.

62. Mangiola F, Nicoletti A, Gasbarrini A, Ponziani FR. Gut microbiota and aging. European Review for Medical and Pharmacological Sciences. 2018;22:7404–13.

63. Fulop T, Larbi A, Dupuis G, Le Page A, Frost EH, Cohen AA, et al. Immunosenescence and Inflamm-Aging As Two Sides of the Same Coin: Friends or Foes? Front Immunol. 2018 Jan 10;8:1960.

64. Palacios N, O'Reilly ÉJ, Schwarzschild MA, Ascherio A. Long-Term Use of Antibiotics and Risk of Parkinson's Disease in the Nurses' Health Study. Park Dis. 2020 Jan 20;2020:1–5.

65. Ternák G, Kuti D, Kovács KJ. Dysbiosis in Parkinson's disease might be triggered by certain antibiotics. Med Hypotheses. 2020 Apr;137:109564.

66. Mertsalmi TH, Pekkonen E, Scheperjans F. Antibiotic Exposure and Risk of Parkinson's Disease in Finland: A Nationwide Case-Control Study. Mov Disord. 2020 Mar;35(3):431–42.

67. Bi W, Zhu L, Jing X, Liang Y, Tao E. Rifampicin and Parkinson's disease. Neurol Sci. 2013 Feb;34(2):137–41.

68. Bortolanza M, Nascimento GC, Socias SB, Ploper D, Chehín RN, Raisman-Vozari R, et al. Tetracycline repurposing in neurodegeneration: focus on Parkinson's disease. J Neural Transm. 2018 Oct;125(10):1403–15.

69. Garrido-Mesa N, Zarzuelo A, Gálvez J. Minocycline: far beyond an antibiotic: Minocycline: far beyond an antibiotic. Br J Pharmacol. 2013 May;169(2):337–52.

70. Brown GC. The endotoxin hypothesis of neurodegeneration. J Neuroinflammation. 2019 Dec;16(1):180.

71. Silva DF, Candeias E, Esteves AR, Magalhães JD, Ferreira IL, Nunes-Costa D, et al. Microbial BMAA elicits mitochondrial dysfunction, innate immunity activation, and Alzheimer's disease features in cortical neurons. J Neuroinflammation. 2020 Dec;17(1):332.

72. Li A, Tian Z, Li J, Yu R, Banack SA, Wang Z. Detection of the neurotoxin BMAA within cyanobacteria isolated from freshwater in China. Toxicon. 2010 May;55(5):947–53.

73. Banack SA, Johnson HE, Cheng R, Cox PA. Production of the Neurotoxin BMAA by a Marine Cyanobacterium. Mar Drugs. 2007;17.

74. Faassen E. Presence of the Neurotoxin BMAA in Aquatic Ecosystems: What Do We Really Know? Toxins. 2014 Mar 21;6(3):1109–38.

75. Cox PA, Kostrzewa RM, Guillemin GJ. BMAA and Neurodegenerative Illness. Neurotox Res. 2018 Jan;33(1):178–83.

76. Murch SJ, Cox PA, Banack SA. A mechanism for slow release of biomagnified cyanobacterial neurotoxins and neurodegenerative disease in Guam. Proc Natl Acad Sci. 2004 Aug 17;101(33):12228–31.

77. Nunes-Costa D, Magalhães JD, G-Fernandes M, Cardoso SM, Empadinhas N. Microbial BMAA and the Pathway for Parkinson's Disease Neurodegeneration. Front Aging Neurosci. 2020 Feb 7;12:26.

78. Tan VX, Lassus B, Lim CK, Tixador P, Courte J, Bessede A, et al. Neurotoxicity of the Cyanotoxin BMAA Through Axonal Degeneration and Intercellular Spreading. Neurotox Res. 2018 Jan;33(1):62–75.

79. Marelli G, Belgiovine C, Mantovani A, Erreni M, Allavena P. Non-redundant role of the chemokine receptor CX3CR1 in the anti-inflammatory function of gut macrophages. Immunobiology. 2017 Feb;222(2):463–72.

80. Böttner M, Zorenkov D, Hellwig I, Barrenschee M, Harde J, Fricke T, et al. Expression pattern and localization of alpha-synuclein in the human enteric nervous system. Neurobiol Dis. 2012 Dec;48(3):474–80.

81. De Schepper S, Verheijden S, Aguilera-Lizarraga J, Viola MF, Boesmans W, Stakenborg N, et al. Self-Maintaining Gut Macrophages Are Essential for Intestinal Homeostasis. Cell. 2018 Oct;175(2):400-415.e13.

82. Hyun J, Romero L, Riveron R, Flores C, Kanagavelu S, Chung KD, et al. Human Intestinal Epithelial Cells Express Interleukin-10 through Toll-Like Receptor 4-Mediated Epithelial-Macrophage Crosstalk. J Innate Immun. 2015;7(1):87–101.

83. Regoli M, Bertelli E, Gulisano M, Nicoletti C. The Multifaceted Personality of Intestinal CX 3 CR1 + Macrophages. Trends Immunol. 2017 Dec;38(12):879–87.

84. Nakata K, Sugi Y, Narabayashi H, Kobayakawa T, Nakanishi Y, Tsuda M, et al. Commensal microbiota-induced microRNA modulates intestinal epithelial permeability through the small GTPase ARF4. J Biol Chem. 2017 Sep;292(37):15426–33.

85. Lu X, Yu Y, Tan S. The role of the miR-21-5p-mediated inflammatory pathway in ulcerative colitis. Exp Ther Med [Internet]. 2019 Dec 4 [cited 2021 Feb 18]; Available from: http://www.spandidos-publications.com/10.3892/etm.2019.8277

86. Van Spaendonk H, Ceuleers H, Witters L, Patteet E, Joossens J, Augustyns K, et al. Regulation of intestinal permeability: The role of proteases. World J Gastroenterol. 2017;23(12):2106.

87. Suzuki T. Regulation of intestinal epithelial permeability by tight junctions. Cell Mol Life Sci. 2013 Feb;70(4):631–59.

88. Camilleri M. Leaky gut: mechanisms, measurement and clinical implications in humans. Gut. 2019 Aug;68(8):1516–26.

89. Lubomski M, Tan AH, Lim S-Y, Holmes AJ, Davis RL, Sue CM. Parkinson's disease and the gastrointestinal microbiome. J Neurol. 2020 Sep;267(9):2507–23.

90. Choi JG, Kim N, Ju IG, Eo H, Lim S-M, Jang S-E, et al. Oral administration of Proteus mirabilis damages dopaminergic neurons and motor functions in mice. Sci Rep. 2018 Dec;8(1):1275.

91. Schwiertz A, Spiegel J, Dillmann U, Grundmann D, Bürmann J, Faßbender K, et al. Fecal markers of intestinal inflammation and intestinal permeability are elevated in Parkinson's disease. Parkinsonism Relat Disord. 2018 May;50:104–7.

92. Sharma S, Awasthi A, Singh S. Altered gut microbiota and intestinal permeability in Parkinson's disease: Pathological highlight to management. Neurosci Lett. 2019 Nov;712:134516.

93. Chen H-L, Yamada K, Sakai K, Lu C-H, Chen M-H, Lin W-C. Alteration of brain temperature and systemic inflammation in Parkinson's disease. Neurol Sci. 2020 May;41(5):1267–76.

94. Koziorowski D, Tomasiuk R, Szlufik S, Friedman A. Inflammatory cytokines and NT-proCNP in Parkinson's disease patients. Cytokine. 2012 Dec;60(3):762–6.

95. Lindestam Arlehamn CS, Dhanwani R, Pham J, Kuan R, Frazier A, Rezende Dutra J, et al. α-Synuclein-specific T cell reactivity is associated with preclinical and early Parkinson's disease. Nat Commun. 2020 Dec;11(1):1875.

96. Fan Z, Pan Y-T, Zhang Z-Y, Yang H, Yu S-Y, Zheng Y, et al. Systemic activation of NLRP3 inflammasome and plasma α-synuclein levels are correlated with motor severity and progression in Parkinson's disease. J Neuroinflammation. 2020 Dec;17(1):11.

97. Ugalde-Muñiz P, Fetter-Pruneda I, Navarro L, García E, Chavarría A. Chronic Systemic Inflammation Exacerbates Neurotoxicity in a Parkinson's Disease Model. Oxid Med Cell Longev. 2020 Jan 13;2020:1–19.

98. Abd Elhadi S, Grigoletto J, Poli M, Arosio P, Arkadir D, Sharon R. α-Synuclein in blood cells differentiates Parkinson's disease from healthy controls. Ann Clin Transl Neurol. 2019 Dec;6(12):2426–36.

99. Braak H, Tredici KD, Rüb U, de Vos RAI, Jansen Steur ENH, Braak E. Staging of brain pathology related to sporadic Parkinson's disease. Neurobiol Aging. 2003 Mar;24(2):197–211.

100. Forsyth CB, Shannon KM, Kordower JH, Voigt RM, Shaikh M, Jaglin JA, et al. Increased Intestinal Permeability Correlates with Sigmoid Mucosa alpha-Synuclein Staining and Endotoxin Exposure Markers in Early Parkinson's Disease. Oreja-Guevara C, editor. PLoS ONE. 2011 Dec 1;6(12):e28032.

101. Pouclet H, Lebouvier T, Coron E, Bruley des Varannes S, Rouaud T, Roy M, et al. A comparison between rectal and colonic biopsies to detect Lewy pathology in Parkinson's disease. Neurobiol Dis. 2012 Jan;45(1):305–9.

102. Adams B, Nunes JM, Page MJ, Roberts T, Carr J, Nell TA, et al. Parkinson's Disease: A Systemic Inflammatory Disease Accompanied by Bacterial Inflammagens. Front Aging Neurosci. 2019 Aug 27;11:210.

103. Smith AM, Depp C, Ryan BJ, Johnston GI, Alegre-Abarrategui J, Evetts S, et al. Mitochondrial dysfunction and increased glycolysis in prodromal and early Parkinson's blood cells: Mitochondria and Glycolysis In PD Blood Cells. Mov Disord. 2018 Oct;33(10):1580–90.

104. Vida C, Kobayashi H, Garrido A, Martínez de Toda I, Carro E, Molina J, et al. Lymphoproliferation Impairment and Oxidative Stress in Blood Cells from Early Parkinson's Disease Patients. Int J Mol Sci. 2019 Feb 12;20(3):771.

105. Guerra F, Girolimetti G, Beli R, Mitruccio M, Pacelli C, Ferretta A, et al. Synergistic Effect of Mitochondrial and Lysosomal Dysfunction in Parkinson's Disease. Cells. 2019 May 14;8(5):452.

106. Garretti F, Agalliu D, Lindestam Arlehamn CS, Sette A, Sulzer D. Autoimmunity in Parkinson's Disease: The Role of α-Synuclein-Specific T Cells. Front Immunol. 2019 Feb 25;10:303.

107. Svensson E, Horváth-Puhó E, Thomsen RW, Djurhuus JC, Pedersen L, Borghammer P, et al. Vagotomy and subsequent risk of Parkinson's disease: Vagotomy and Risk of PD. Ann Neurol. 2015 Oct;78(4):522–9.

108. García-Domínguez I, Veselá K, García-Revilla J, Carrillo-Jiménez A, Roca-Ceballos MA, Santiago M, et al. Peripheral Inflammation Enhances Microglia Response and Nigral Dopaminergic Cell Death in an in vivo MPTP Model of Parkinson's Disease. Front Cell Neurosci. 2018 Nov 6;12:398.

109. Chiang P-L, Chen H-L, Lu C-H, Chen P-C, Chen M-H, Yang I-H, et al. White matter damage and systemic inflammation in Parkinson's disease. BMC Neurosci. 2017 Dec;18(1):48.

110. Popescu BO, Toescu EC, Popescu LM, Bajenaru O, Muresanu DF, Schultzberg M, et al. Blood-brain barrier alterations in ageing and dementia. J Neurol Sci. 2009 Aug;283(1–2):99–106.

111. Pan Y, Nicolazzo JA. Impact of aging, Alzheimer's disease and Parkinson's disease on the blood-brain barrier transport of therapeutics. Adv Drug Deliv Rev. 2018 Oct;135:62–74.

112. Satoh K, Niwa M, Goda W, Binh NH, Nakashima M, Takamatsu M, et al. Galectin-3 expression in delayed neuronal death of hippocampal CA1 following transient forebrain ischemia, and its inhibition by hypothermia. Brain Res. 2011 Mar;1382:266–74.

113. Haruwaka K, Ikegami A, Tachibana Y, Ohno N, Konishi H, Hashimoto A, et al. Dual microglia effects on blood brain barrier permeability induced by systemic inflammation. Nat Commun. 2019 Dec;10(1):5816.

114. Jangula A, Murphy EJ. Lipopolysaccharide-induced blood brain barrier permeability is enhanced by alpha-synuclein expression. Neurosci Lett. 2013 Sep;551:23–7.

115. Kujawska M, Jodynis-Liebert J. What Is the Evidence that Parkinson's Disease Is a Prion Disorder, Which Originates in the Gut? Int J Mol Sci. 2018 Nov 12;19(11):3573.

116. Sui Y-T, Bullock KM, Erickson MA, Zhang J, Banks WA. Alpha synuclein is transported into and out of the brain by the blood–brain barrier. Peptides. 2014 Dec;62:197–202.

117. Kim S, Kwon S-H, Kam T-I, Panicker N, Karuppagounder SS, Lee S, et al. Transneuronal Propagation of Pathologic α-Synuclein from the Gut to the Brain Models Parkinson's Disease. Neuron. 2019 Aug;103(4):627-641.e7.

118. Gray MT, Woulfe JM. Striatal Blood–Brain Barrier Permeability in Parkinson'S Disease. J Cereb Blood Flow Metab. 2015 May;35(5):747–50.

119. Karakatsani ME, Blesa J, Konofagou EE. Blood–brain barrier opening with focused ultrasound in experimental models of Parkinson's disease. Mov Disord. 2019 Sep;34(9):1252–61.