The Neuromelanin Paradox and its Dual Role in Oxidative Stress and Neurodegeneration

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Abstract

Aging is associated with an increasing dysfunction of key brain homeostasis mechanisms and represents the main risk factor across most neurodegenerative disorders. However, the degree of dysregulation and the affectation of specific pathways set apart normal aging from neurodegenerative disorders. In particular, the neuronal metabolism of catecholaminergic neurotransmitters appears to be a specifically sensitive pathway that is affected in different neurodegenerations.

In humans, catecholaminergic neurons are characterized by an age-related accumulation of neuromelanin (NM), rendering the soma of the neurons black. This intracellular NM appears to serve as a very efficient quencher for toxic molecules. However, when a neuron degenerates, NM is released together with its load (many undegraded cellular components, transition metals, lipids, antibiotics) contributing to initiate and worsen an eventual immune response,

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exacerbating the oxidative stress, ultimately leading to the neurodegenerative process.

This review focuses on the analysis of the role of NM in normal aging and catecholaminergic metabolism due to its capability as a pro-oxidant and other harmful molecules, versus its involvement in oxidative stress and aberrant immune response, which it is highly dependent on NM saturation state and its extracellular release.

Keywords: reactive oxygen species (ROS), neuromelanin (NM), oxidative stress, neurodegeneration, immune response

1. Metals, oxidative stress, and neurodegeneration

Neurodegenerative diseases are characterized by progressive cognitive dysfunction and loss of neurons and synapsis. Neurodegeneration is a process extremely complex, which involves a multifactorial imbalance with the simultaneous interplay of numerous processes operating at different levels of functional organization. In this dynamic context, neurodegeneration may be considered a pathological brain-aging process, triggered by alterations on distinct molecular pathways, genetic predisposition, and environmental toxic exposure. Therefore, aging is by far the greatest risk factor for highly prevalent neurodegenerative disorders [1].

A key feature for most neurodegenerative disorders is the accumulation of misfolded protein aggregates, framing them within the classification concept of proteinopathies or “protein conformational disorders”. However, it is important to underscore that not all neurodegenerative diseases can be considered as protein conformational disorders [2].

Although the molecular underpinnings of neurodegeneration are still not completely understood, the increased oxidative stress associated with brain aging stands out as one of the common features across various neurodegenerative diseases[3–6]. Thus, aging is associated with increasing levels of pro-oxidant factors (reactive oxygen species, ROS) and the dysfunction

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of the antioxidant systems, leading to protein and cellular damage and ultimately to neurodegeneration.

The brain high vulnerability to oxidative stress appears to be mainly related to i) its high oxygen demand (~20% of the body total basal oxygen), ii) the high polyunsaturated fatty acids content of neuronal membranes, iii) the accumulation of transition metals, and iv) the absence of efficient antioxidant systems[7].

Transition metals, such as iron, copper, and zinc, are necessary for brain functioning, participating in essential biological reactions as cofactors of metalloproteins that intervene in myelination and neurotransmitters metabolism, among other roles[8–13]. However, iron and copper are very efficient catalysts of ROS production via the Fenton or Haber-Weiss reactions. Under physiological conditions, free radicals contribute to sustaining a normal cellular redox state; however, under certain conditions, ROS levels increase and in consequence, oxidative damage is triggered. For this reason, transition metal levels require a tight control that tunes their levels and location.

Metal dyshomeostasis yields high levels of ROS, leading to oxidative stress and mitochondrial dysfunction[14–17]. In turn, mitochondrial dysfunction triggers a higher ROS production, driving to a “vicious cycle” inducing cellular damage, by altering a wide variety of cellular components, resulting in DNA damage, protein oxidation, and peroxidation of polyunsaturated fatty acids in membrane lipids[12,18–20].

In neurodegenerative disorders, increases in transition metal levels have been reported, being iron the metal showing the most substantial change[21–23]. In Alzheimer´s disease (AD), the accumulation of metals is observed within β-amyloid senile plaques[24–27], and alterations in iron, copper, and zinc levels have been found in several regions of AD brain[28–31]. Similarly, disturbances in metal levels have been reported in Parkinson's disease (PD)[32–35]. The molecular mechanisms through which metal dysregulation triggers neurodegeneration are diverse, but they appear to be mediated mainly by the generation of free radicals and oxidative stress[4].

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Among these noxious mechanisms, lipid peroxidation enhanced by iron appears to be a relevant triggering factor for neurodegeneration since the brain is highly enriched in polyunsaturated fatty acids and iron excess promotes ROS production. Lipid peroxidation exerts its deleterious effects through two general mechanisms: i) Loss of the integrity of cellular membranes (it is important to note that this process affects not only the cell membrane, but also the mitochondria, endoplasmic reticulum, and nuclear membranes), and ii) Generation of intermediate- and end-products, such as lipid hydroperoxides, malondialdehyde, 4-hydroxynonenal and acrolein that leads to genotoxicity, cytotoxicity and ultimately, cellular death[36–40]. This type of regulated non-apoptotic peroxidation-driven and iron-dependent cell death mechanism is called “ferroptosis”[41]. Ferroptosis has been established in neurodegenerative diseases[42–45], being considered as an alternative cell death mechanism in the absence of downstream indicators of apoptotic death[46].

Additionally, iron, copper, and zinc cations can be coordinated to Aβ peptides and α-synuclein, promoting their aggregation and leading to the formation of the histopathological hallmarks of AD and PD, respectively[47–54]. The resulting protein-metal complexes aggregates are directly involved in ROS production, thus exacerbating the oxidative damage[49,55–58].

2. Local origin of neurodegenerative diseases: function and involvement of neuromelanin in disease

In general, neurodegenerative disorders appear to have a focal origin from which the pathology spreads. AD pathology expands from the cholinergic nuclei in the basal forebrain and the noradrenergic nuclei in the brainstem, most importantly the *Locus coeruleus* (LC). PD brain pathology appears to initiate in the brainstem or limbic system with the greatest affectation of the *Substantia nigra* (SN). Interestingly, degeneration of cholinergic nucleus basalis is characteristic of AD, but also occurs in PD; while, neuronal loss in dopaminergic SN is the pathological hallmark of PD, but also occurs to a variable degree in AD[59,60]. Depletion of neurons in the noradrenergic LC is also recognized in both disorders, and for both AD and PD the greatest neuronal loss is found within the LC[61].

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Despite that aging-related oxidative stress is a global process that affects all neurons, certain neuronal populations are selectively vulnerable in different neurodegenerative diseases, suggesting that additional factors play a role in the pathophysiological mechanisms of neurodegeneration of specific neurons and brain regions[62,63].

Dopaminergic and noradrenergic neurons in SN and the LC, respectively, are exposed to additional oxidative stress due to their inherent metabolism of catecholamines, which generates significant amounts of ROS[64]. These neuronal populations are characterized by an age-dependent accumulation of neuromelanin (NM)[65]. It has been suggested that NM plays a protective role in these neurons by preventing the accumulation of catechol derivatives generated from dopamine or noradrenaline auto-oxidation, thus avoiding their toxic effect[66].

NM is a dark neuronal pigment, therefore it can be mainly found in brain regions enriched in dopaminergic or noradrenergic neurons, such as SN or LC, but also to a lesser extent in other regions of the central nervous system (CNS) and peripheral nervous system (PNS) [63][67]. NM belongs to the family of melanins, a big group of biopolymers ubiquitous in nature[68]. Although there is no consensus about its structure, it appears to be formed by a heme-like structure in a π-conjugated matrix. Due to this conformation, it has been attributed with many functionalities from which stands out its ability to trap free radicals, toxins, and metals due to its oligomeric structure of benzothiazine rings containing conjugated double bonds.

Since the presence of NM in catecholaminergic neurons is associated with the catecholamine metabolism itself, the accumulation of NM in the soma of these neurons begins as early as the first decade of human life. NM levels gradually increases with age throughout life, until we reach the age of 60, approximately[69,70]. Afterwards, the total amount of NM appears to stabilize due to the balance between the number of neurodegenerating melanized neurons, and the ever-increasing NM levels in the surviving neurons, as

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confirmed by early histological studies in higher animals [70,71]. Thus, NM appears to play a role both in normal aging and neurodegeneration.

Opposite to other melanins (eumelanin and pheomelanin) that are localized in skin melanocytes and their synthesis depends on the enzyme tyrosinase that carries out the initial oxidation of the melanin precursor L-tyrosine, the synthesis of NM in the neurons is not enzymatic regulated, getting accumulated in organelles of autophagic nature[67,72,73].

NM seems to be composed of a mixture of cysteinyl derivatives of oxidized dopamine or noradrenalin produced during the neurotransmitter synthesis[74,75]. Regarding its properties and functionality, opposite to other melanins, whose function is to yield protection against UV, historically brain NM has been considered a waste product of catecholaminergic neurotransmitter synthesis. However, recent evidence suggests new roles for NM ranging from neuroprotection to neurodegeneration[76]. It has been suggested that the vulnerability of dopaminergic and noradrenergic neurons in SN and the LC, respectively, is related to their pigment content[77].

In normal conditions, NM production appears to play a protective role within the cell by preventing the accumulation of toxic catechol derivatives produced via the catecholamine auto-oxidation pathway[66,78], preventing ROS generation. NM presents also a chemoprotective role by interacting with a variety of potentially damaging molecules such as pesticides and neuroleptics[79]; as well as, potentially toxic cations such as iron, zinc, copper, manganese, chromium, cobalt, mercury, lead, and cadmium[75,80], acting like a ‘black hole’ capable of chelating redox-active metals, especially iron[66,81,82]. Hence, NM is the main iron storage molecule in dopaminergic neurons in the SN[83,84]. Interestingly, NM possesses two types of iron-binding sites (high and low-affinity sites); and under physiological conditions, iron binds preferentially to NM by the high-affinity site, avoiding its participation in ROS production and protecting cells from oxidative stress. However, this protective function appears to be lost at high iron concentrations, where the high binding site is saturated and the remaining iron binds to the NM by the low-affinity site. Thus, NM becomes into pro-oxidative

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burden, where iron could easily be released to participate in oxidative processes[85]. Therefore, the NM anti-oxidant or pro-oxidant role will greatly depend on the result of the regulation of iron/NM ratio[3].

3. Crosstalk of neuromelanin with lipofuscin in lipid peroxidation in catecholaminergic neurons

Besides the accumulation of NM, another striking morphological change in neurons throughout the brain during normal aging is related to the accumulation of lipofuscin (LF) aggregates, which is traditionally considered as an “aging pigment” [86,87]. LF, as a undigested bioproduct of central processes of cellular detoxification of autophagy, is associated with both aging and neurodegeneration, reflecting the impairment of the exocytosis and lysosomal secretion systems [88]. Interestingly, the main component of LF comes from highly reactive lipid derivatives such as 4-hydroxy-2-nonenal [89] that form adducts by reacting with histidine, lysine, and cysteine residues, which also block proteasomal activity.

Particularly, in catecholaminergic neurons, there is a simultaneous buildup of LF and NM in cytoplasmic vacuoles. As both NM and LF accumulate throughout life and are poorly degraded in catecholaminergic neurons, the colocalization of both types of polymers may produce a redox crosstalk between NM and LF, where the highly pro-oxidant effect of transition metals such as iron maybe quenched by NM or amplified by a lipid peroxidation cascade initiated by LF lipids.

Besides, since the formation of NM is a non-enzymatically controlled auto-oxidation process, the presence of metals and LF may produce and increased oxidative state that drives NM formation [90–92]. According to this crosstalk, NM and LF in the SN present a certain level of inter-convertibility, revealing that under certain conditions NM can arise from melanized LF [93]. Reciprocally, Barden et al. (11) demonstrated that in presence of ferrous sulfide, LF could be transformed into NM after a pseudoperoxidation process, becoming melanized in the presence of DOPA or its precursors showing properties similar to NM. Thus, LF could be considered as a product of an early stage in the maturation of the

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lipopigmentary metabolism, being the melanic pigments the result from a pseudoperoxidation of LF or its precursors, catalyzed by metals [94].

The same process was observed in the peroxidation of spheroids, where oxidation products of myelin metabolism occurred in the presence of iron [95]. Thus, oxygen peroxide is replaced by lipid peroxide originated in the oxidation of myelin fatty acids, considered in the process as a precursor of LF. The fact that NM could come, at least in part, from the conversion of LF through a process of peroxidation suggests that NM is an end product of the neurodegenerative metabolism of lipids. The potential interconversion between NM and LF mediated by a dynamic phenomenon pseudoperoxidation of lipids supports the colocalization of both age-related pigments observed in different brain areas [96].

The lipid component and the presence of metal ions seems to be a common pattern to NM and LF that will determine their antioxidant or pro-oxidant role. It is widely accepted that LF originates from lipid and lipoprotein peroxidation during normal cell-aging; however, the interconvertibility between NM and LF and the crosstalk between both molecules points out to an important role in neurodegeneration [97].

4. Neuromelanin and Immune response

With aging, there is a global low-level increase in the brain inflammatory response. However, in neurodegenerative diseases, there is also a strong increase in the inflammatory response in specific and restricted areas of the brain, where extracellular cell-released NM could be playing a relevant role in triggering and sustaining the inflammation process[98]. Regardless of the cell death mechanism, once a catecholaminergic neuron dies, the NM, loaded with all the substances that were bound while in the cell, is released to the brain parenchyma. Each component bound to the biopolymer might be released or not; but depending on their nature, they may contribute to initiate or exacerbate an immunological response (Figure 1).

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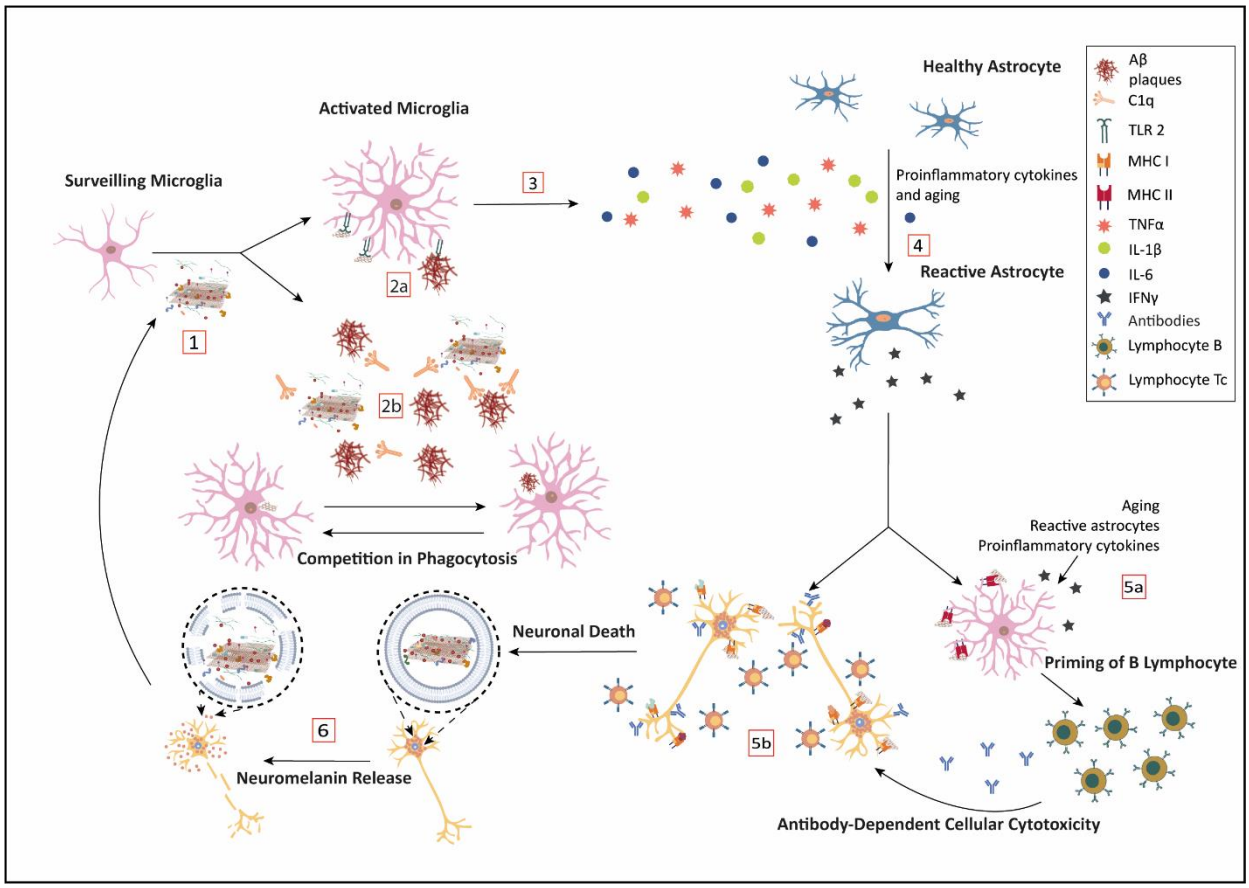

Figure 1. Immune response to the NM release to brain parenchyma. Upon neuronal cell death, surveilling microglia engulf cellular debris. The NM released to the brain parenchyma is less stable than the intracellular vesicle-contained NM, and therefore may distribute its toxic load throughout the tissue, triggering microglial activation (1). At the same time, NM may interact with the protein C1q which facilitates its phagocytosis, hindering in turn the clearance of amyloid deposits by competition (2b). Simultaneously, NM structure could interact with microglial TLR 2 (2a) promoting the secretion of pro-inflammatory interleukins (3). These secreted interleukins will amplify neuroinflammation, impairing other neurological structures and transforming astroglial cells into reactive astrocytes that produce INFγ (4). Activated microglia by INFγ stimulus will express MHC I, displaying portions of NM and its load that in the deep cervical lymph nodes could prime B cells, potentially inducing an autoimmune response (5a). Parallelly, INFγ also induce the expression of MHC I in healthy neurons and similar to MHC II in microglia, they could present modified antigens by its binding to NM and trigger Tc lymphocytes attack (5b). Finally, an increased cell death is associated with higher release of NM, stablishing a feed-back loop that exacerbates a pathological immune response (6).

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4.1. Neuromelanin and the Innate Immunity

Microglia, as brain-specific resident macrophages, can engulf the released NM to clear it (see Figure 1, steps 1 and 2). In this line, Zhang and colleagues showed that human NM can be phagocytized by primary enriched microglia Fisher 334 rat cell culture[99]. Its degradation seems to be mediated by ROS production by the phagocyte oxidase (PHOX) enzyme, which is responsible for a respiratory burst. Also, Depboylu *et al.*[100] demonstrated through immunohistochemistry and *in situ* hybridization techniques that in the SN and other brain regions from PD patients, there is an increased expression of C1q in microglia cells surrounding extracellular coming from dead NM-containing neurons, thus facilitating opsonization and phagocytosis by microglia. Interestingly, the same analogous process, mediated by C1q, appears to be relevant for the clearance of Aβ peptide plaques by the microglia (see Figure 1, step 2)[101,100]. Recently, an important role is being attributed to C1q in neurodegeneration[102]. Here, we propose that the competition between the clearance of NM and amyloid plaques by C1q opsonization and microglia phagocytosis may result in the blockage of the physiological system of clearance, leading to the accumulation of deleterious components and the progression of the neurodegenerative process.

Another relevant pathway for NM to activate the innate immunity mechanisms of microglia is through Pattern Recognition Receptors (PRR). Since NM has a repetitive structure, it is a good candidate to interfering with this pathway. Extracellular NM can activate *in vitro* microglia towards the M1 proinflammatory profile by induction of pattern-recognition receptors such as toll-like receptor 2 (TLR2) and nucleotide-binding oligomerization domain 2 (NOD2)(see Figure 1, step 2a)[103]. Extracellular NM fragments bind to TLR, also expressed in astroglial cells, to induce the NF-κB expression by the MyD88 transduction signaling, which is a known pathway to start inflammation by releasing proinflammatory cytokines, such as IL-1β, IL-6, and TNFα (see Figure 1, step 4) [103]. By the same pathway, Aβ brain deposits seem to be cleared by TLR2, lessening cognitive decline in AD and aging[104]. Once again, these results point to a competition for the resources and mechanisms capable of removing amyloid plaques and extracellular NM deposits.

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Microglia over-activated by extracellular NM secretes large quantities of proinflammatory cytokines (Figure 1, step 3); which in turn, induce the expression of NO synthase (iNOS) and the production of NO, further increasing free ROS. NO is associated with neurodegeneration by different mechanisms: (i) iron dyshomeostasis, (iron imbalance is involved in both AD and PD)[105]; (ii) lipoperoxidation in association with transition metals[106], and (iii) antigenic modifications due to ROS derivates of NO[107,108]. Together with the intrinsic effect of ROS production, these NO-related mechanisms represent a strong inflammatory stimulus that can mount a sterile immune response involving other cells as astroglia and T lymphocytes and the production of other proinflammatory cytokines (Figure 1, step 5).

4.2. Neuromelanin and Acquired Immunity

In this context, two relevant findings point to the activation of microglia by NM as a potential triggering factor of the autoimmune response involved in PD. Orr and colleagues studied a humoral mechanism capable of causing microglial-meditated injury in tissue from idiopathic and genetic PD compared to controls[109]. They detected IgG immunoglobulins recognizing dopaminergic neuronal structures that correlated with pigmented neurons. Moreover, they found MHC class II immunopositive microglia expressing the high-affinity IgG receptor FcγRI, which is consistent with a potential phagocytic attack on the IgG-immunopositive pigmented neurons (Figure 1, step 5). Besides, the targeting of pigmented neurons by antibodies may also trigger antibody-mediated cytotoxicity. In line with this, Double *et al.* found that antibodies against melanin-skeleton were increased in patients with a clinical diagnosis of PD[110]. Moreover, the concentration of antibodies detected correlated negatively with the disease duration, while no correlation was found with the severity of the disease.

Thus, it seems that there is a first innate response, which induces a beneficial inflammation to help clearing the melanin remnants, evolves into a more complex and difficult-to-manage pernicious immune response mediated by cellular immunity.

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Microglia, as a macrophage-related cell, expresses MHC class II and it is the main cell population able to present antigens in the CNS[111]. Nevertheless, under a pro-inflammatory environment, several cells inside the CNS can act as Antigen-Presenting Cells (APC), such as astrocytes, border-associated macrophages (BAM, which reside in the choroid plexus, the pia, and the dura maters), and dendritic cells (DC) in the blood-brain barrier[112]. In this line, Oberländer *et al* used human NM to stimulate murine DC, and observed how the DC can capture the NM and be activated as APC expressing MHC II and CD86 in their cell surface[113]. Thus, they propose microglia as the initial APC that triggers NM-driven autoimmune-based pathogenesis of PD.

The phagocytosis of NM by APC cells opens a new potential avenue with immunological consequences[114]. Extracellular NM and their derived components would be presented by MHC class II, leading to two different pathways: i) activating B cells to produce autoantibodies, and ii) cytotoxic response priming naïve T cells, which under a pro-inflammatory environment mediated by cytokines that could infiltrate from subarachnoid vessels (see Figure 1, step 5a)[115].

Despite the immune privilege of the CNS, the priming process is possible because the CSF containing immune cells and brain antigens is transported by brain lymphatic vessels to the deep cervical lymph nodes (dCLN), where the APC can present all those components attached to NM as brain antigens to naïve T cells[100]. Upon their activation in the context of neuroinflammation, they can be chemoattracted and mobilized towards the CNS[116]. At the same time, APC would prime B cells to turn into plasmatic cells to secrete IgGs, thus explaining the presence of IgG against NM in sera from PD patients[110].

As NM and their components are presented by MHC II and activate an immune response, it is also plausible that in the proper environment (*e.g.* presence of IFNγ), they may be presented by MHC I (Figure 1, step 5b). TLR, proinflammatory cytokines such as TNFα, or even aging[116] can induce astrocytes to become reactive astrocytes, leading to the secretion of IFNγ, which promotes MHC I expression in catecholaminergic neurons. Then,

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catecholaminergic cells may present neuromelanin portions together with protein oligomers or toxic components. Previously, these proteins or peptides may have experienced post-translational changes enhancing its antigenicity, and thus triggering an autoimmune response that targets both degenerating and healthy dopaminergic neurons for cytotoxic T lymphocytes (Tc)[117]. The infiltration of activated Tc cells, exacerbation of the neuroinflammation, and loss of neurons may contribute to the progression of pathology in different neurodegenerative diseases.

In this situation, the CXCL10 pathway is another checkpoint where NM could interfere. CXCL10 is a chemokine strongly secreted in response to interferon, as well as by the expression of NF-κB induced by TNFα. CXCL10 appears to function as a chemo-attractive for activated T cells, monocytes/macrophages, and microglia, promoting antimicrobial activity and inducing astrocyte proliferation. Although its role is not well understood, depending on the context may be neuroprotective or neurotoxic. CXCL10 levels have correlate with both β-amyloid[118] plaques and mini-mental state examination (MMSE) [119], but it is still not clear whether it is increased in the CSF of AD patients *versus* controls [120,121]. In the PD murine model induced by 1-methyl 4-phenyl 1,2,3.6-tetrahydropiridine (MPTP), CXCL10 mRNA expression was up-regulated in the striatum and the ventral midbrain[122], and it also seems to be related to HIV-1 dementia and Tick-borne encephalitis neuropathology[121]. Tousi and colleagues[123] showed *in vitro* how NM in the presence of TNFα inhibits CXCL10 expression by inhibition of NF-κB activation in the human A172 astroglial cell line. Nevertheless, no effect was detected when NM was present alone without a proinflammatory background. In the context of CXCL10 neurotoxicity, NM appears to be neuroprotective. Moreover, the fact that NM is able of inhibiting the expression of CXCL10 in an NF-κB-depending manner may have wider implications, since the same pathway regulates many pro-inflammatory cytokines at the same time.

5. Conclusions

As previously discussed, the accumulation of NM is a process that is not enzymatically regulated. With aging, the cell appears to be unable to balance

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adequately between the production of NM that helps to cope with increasing oxidative stress, toxicants binding on one side, and the production of catecholamines on the other side. Thus, the aging cell cannot keep increasing NM production at the expense of a reduced production of neurotransmitters that would lead to a neurodegeneration associated to loss of function of the cell.

In normal physiological states, intracellular NM plays a protective role for the neuron as a metal chelator, preventing ROS production and therefore oxidative damage. However, when the concentration of active metals and other toxicants surpasses a certain threshold, the antioxidant potential of NM turn into a pro-oxidant role, in a feedback loop, producing an increased cellular oxidative stress.

Oxidative damage includes, among others, lipid peroxidation that compromises the cellular integrity and promote the production of noxious products. Both processes lead to cellular death and the consequent release of NM from catecholaminergic neurons into the brain parenchyma. Thus, the release of NM, loaded with all the substances that were bound while in the cell, may contribute to initiate or exacerbate the strong brain inflammatory status observed in neurodegenerative processes, mediated by both an autoimmune and cytotoxic response that result is cell death and further NM release, closing a vicious circle that leads to the progression of neurodegeneration. Thus, the paradox of NM is that its greatest virtue is its worst defect. NM can play both a cytoprotective or neurotoxic role depending on its cellular and extracellular context and the load that carries (Table 1).

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Table 1. The properties of neuromelanin.

NEUROPROTECTOR	NEUROTOXIC
Antioxidant activity: trapping free radicals, toxicants, and metals	Neuroinflammation trigger: Release trapped toxic components (*i.e.*: metals, drugs, amyloids, lipids…)
Potential dopamine reservoir[76]	Its clearance compete with other harmful molecules[100,101]
Immunomodulator (NK-κB inhibition)[123]	Potentially autoimmune response inducer[113]

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Abbreviations

AD	Alzheimer´s disease
APC	Antigen presenting cell
BAM	Border associated macrophage
CNS	Central nervous system
DC	Dentritic cell
dCLN	Deep cervical lymph node
FcγRI	High-affinity IgG receptor
IFNγ	Interferon γ
IgG	Immunoglobulin G
iNOS	Inducible nitric oxygen synthase

LC	*Locus coeruleous*
LF	Lipofuscin
MHC	Major histocompatibility complex
MPTP	1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine
NFκB	Nuclear factor κ B
NM	Neuromelanin
NOD2	nucleotide-binding oligomerization domain containing protein 2
PD	Parkinson´s disease
PHOX	phagocyte oxidase
PNS	Peripheral nervous system
PRR	Pattern recognition receptors
ROS	Reactive oxygen species
SN	*Substantia nigra*
TLR2	Toll-like receptor 2
TNFα	Tumor necrosis factor

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