Alpha-Synuclein: Activation of Microglia and Iron Metabolism in Parkinson diseases

Chengkui Shi [1, a], Xinxing Du [2, b]

[1]Department of Physiology, Shandong Provincial Key Laboratory of Pathogenesis and Prevention of Neurological Disorders, Shandong Provincial Collaborative Innovation Center for Neurodegenerative Disorders and State Key Disciplines: Physiology, Medical College of Qingdao University, Qingdao 266071, China

[2]Five-year clinical medicine of Class 7, Grade 2014,Medical College of Qingdao University, Qingdao 266071, China

[a]shichengkui@163.com, [b]807102287@qq.com

Abstract

Microglial cells are central nervous system innate immune effector cells that respond to different stimuli, such as damage, neurodegeneration, stroke, and brain tumors. Some studies about Parkinson's disease (PD) specific markers Alpha-synuclein-dependent glial activation, particularly on mutant or aggregated forms of alpha-synuclein, have been shown to lead to more Strong activation of microglia and astrocytes, release of cytokines and oxidative stress. The substantia nigra compacta is associated with iron deposition in Parkinson's disease, mainly in neurons and microglia. At the same time studies have shown that abnormal aggregation of Alpha-synuclein is often associated with iron deposition, indicating that there is a link between iron and Alpha-synuclein aggregation. Alpha-synuclein is a high-iron reductase that reduces intracellular Fe3 + to Fe2 +. Therefore, alpha-synuclein may play a key role in microglia activation and iron deposition.

Keywords

Alpha-synuclein, Microglia, Iron metabolism.

1. Microglia Overview

1.1 Microglia origin and distribution

On the origin of microglia, there is still no conclusion. It is now generally accepted that microglia originate from mesodermal mononuclear cells, or from the neuroectoderm. Others believe that microglia are not true glial cells. Microglia are widely distributed in the central nervous system and retina. The distribution of microglial cells in the normal brain and resting state is not uniform, and the distribution in the substantia nigra is significantly higher than that in the midbrain and other regions of the brain, such as the hippocampus.

1.2 microglia function

In a healthy brain, the microglia are in a quiescent state, and they search for tissue damage and intruders. If tissue damage or infection, microglia will be transformed into activated and phagocytic state, mainly for the morphological changes, amplification, neurotrophic substances (such as: BDNF) or inflammatory factors (such as: tumor necrosis factor TNF- α) increased and oxidative stress strengthened (such as the generation of reactive oxygen species ROS) [1].

In vitro cell culture experiments, the release of proinflammatory cytokines indicates that Alpha-synuclein has a significant effect on the activation of microglia. In addition, overexpression of the Alpha-synuclein in the local area of the brain of the mouse may be caused by viral vectors or the tyrosine-hydroxylase promoter to cause early progressive microglia activation before the neuronal pathology takes place , T cell activation, substantia nigra and striatum pro-inflammatory factor

production. However, little is known about the time course and distribution of microglia activation and cytokine production [2]

1.3 dual role of microglia in Parkinson's disease

Parkinson's disease is a process of inflammatory changes. Many mechanisms of disease damage in the nervous system are characterized by the production of inflammation. Microglia activation, proliferation of central nervous system disease is an important manifestation of inflammatory response. Parkinson's patients, nigrostriatal microglia in a large number of activation, value, especially the most significant of the substantia nigra. Microglia activation, the value of the dopaminergic neuron degeneration before, but when the activation of microglia reached its peak, the neuronal damage has reached the highest value. Microglia activation after the release of a variety of proinflammatory cytokines and neurotrophic factors, causing a variety of inflammatory response and apoptosis. Therefore, in the nervous system, microglia have the dual of role damage and protection [3].

1.3.1 microglia activation

There is some evidence that Alpha-synuclein is released by neuronal cells responding to neuronal activity, or released by Golgi independent endocytosis, and can directly activate microglia. Alpha-synuclein, once released, binds to microglia and leads to its activation. This process has been confirmed by the addition of exogenous Alpha-synuclein in murine primary culture, human microglia culture and mononuclear cell culture [2]. In a healthy brain, the microglia are in a quiescent state, and they search for tissue damage and intruders. If tissue damage or infection, microglia will be transformed into activated and phagocytic state, mainly for the morphological changes, amplification, neurotrophic substances (such as: BDNF) or inflammatory factors (such as: tumor necrosis factor TNF- α) and increased oxidative stress (such as the generation of reactive oxygen species ROS) [4]. Thus, in brain remodeling and maturation, microglia are thought to contribute to clearance of cells through programmed cell death. In the physiological state of the mature brain, the resting microglia have a branching morphological appearance, often with immune surveillance and host defense. However, microglia are particularly sensitive to their microenvironmental changes and are easily activated to respond to infection or injury. Activated microglial cells upregulate surface receptors, including major histocompatibility complexes and various complement receptors. They also undergo severe morphological changes from resting-branched cells into activated amoeboid microglia [5].

1.3.2 microglia damage

Study have reported that PD patients have significant iron deposition in the substantia nigra region of the brain, a large number of experiments have also been confirmed in PD cells and animal models of substantia nigra increased iron content, mainly deposited in neurons and microglia [6, 7,8,9]. Microglial cells are particularly sensitive to heavy metal damage, when the brain is damaged by heavy metals,it can be response to damage before neuronal degeneration, so microglia can be as a marker of heavy metal damage. Activation of microglia is through a variety of pathways in the pathogenesis of Parkinson's neurons. Oxidative stress is not negligible in the activation of microglia injury. Microglia is activated after the free radicals on the toxic effects of neurons. Reactive oxygen species include various oxygen radicals and non-radical oxygenates that are closely related to the behavior of various free radicals. Active oxygen production of cytotoxic effects known as oxidative stress, polyunsaturated fatty acids caused by lipid peroxidation can lead to macromolecular substances, especially DNA changes, can also lead to apoptosis and necrosis [10]. Excitotoxic effects of microglia also play a role in the injury mechanism. Activated microglia can be synthesized prostaglandin E2. Prostaglandin E2 is a mediator of the post-synaptic signaling cascade of the N-methyl-D-aspartate glutamate receptor. Prostaglandin E by inhibiting the reuptake of glutamate astrocytes, glutamatergic neurons to enhance transmission. Glutamate is an important excitatory neurotransmitter in the central nervous system. Pathological circumstances, the extracellular glutamate concentration increased, excessive stimulation of its receptor, the central nervous system has obvious excitotoxicity, causing dopaminergic neuronal death. In Parkinson's disease, microglia-

derived NGF can induce apoptosis by activating low-affinity nerve growth factor receptor p75 and activating transcriptional factor NF-kB to produce toxic effects on dopaminergic neurons. Neurotrophic factors play a role in neurological damage or protection by binding different types of receptors to different mechanisms of action. Tyrosine kinases and LNGFRp75 are expressed throughout the nervous system, the gene expression in the development and maturation of neuronal damage have changed, but the same kind of neurotrophic factor binding with different types of receptor affinity [11]. Microglia-derived neurotrophic factor can induce the development of retinal cell death by stimulating LNGFRp75, the retinal cell development of the tissue environment without nerve growth factor, cell death will be significantly reduced.

1.4 microglia activation in three ways

A variety of signaling pathways lead to activation of microglia, however, in these diseases, what is the trigger of microglia activation still need to explore. First, there may be a factor that does not have direct side-effects on neuronal cells. Current research suggests that one of the best investigational drugs may be bacterial cell wall endotoxin LPS [5]. Lipopolysaccharide (LPS), which has been widely used in the activation of microglia, is an effective tool for the activation of microglia. Although there is no known evidence that LPS has toxic effects on neurons, it can indirectly release neurotoxin-induced neuronal death [12]. However, there are two different neurotoxic factors: MPTP and 6-OHDA, which can directly cause neuronal damage and activate microglia. In addition to the direct and indirect cytotoxic effects mentioned above, there is also an agent associated with a variety of neurodegenerative diseases that exhibit a mixed mode mechanism of neurotoxicity. These agents include beta-amyloid beta (Aβ), HIV-coated protein gp120, prion peptide, and rotenone [13,14]. Aβ enhances neurotoxicity by activating microglia and releasing superoxide radicals. Rotenone was initially thought to damage dopaminergic neurons by inhibiting the activity of mitochondrial complex I, whereas the presence of microglia enhances the neurotoxicity of rotenone while producing oxygen free radicals that enhance the cellular toxicity . Therefore, we believe that mixed mode neurotoxic effects include direct neurotoxic effects and indirect activation of microglia to produce toxicity (Figure 1) [15].

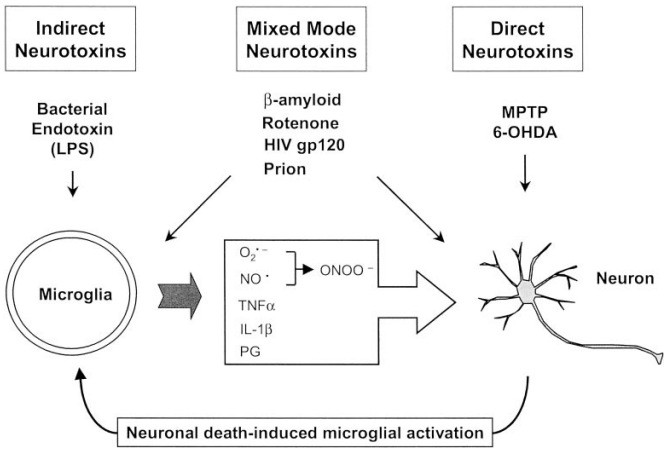

Figure1: Microglia activation in three ways

2. Overview of Alpha-synuclein

2.1 Alpha-synuclein discovery

The alpha-synuclein gene was the earliest discovery of Parkinson's disease-related genes [16]. In some patients with familial Parkinson's disease mutations in the gene. In both familial and sporadic Parkinson's disease, the main inclusion of the characteristic inclusions, Lewy bodies, are aggregated Alpha-synuclein. In 1988, a protein consisting of 143 amino acids was isolated from the cholinergic terminals of the electrical organ of torpedo [17] and named as synuclein. Thereafter, along with the subsequent discovery of the protein homologue, the initial synuclein was referred to as Alpha-synuclein. At the end of the 20th century, mutations in the Alpha-synuclein gene were found in some

patients with familial Parkinson's disease (PD), which led to a great interest in Alpha-synuclein. Alpha-synuclein is an essential component of PD-specific inclusion body-Lewy bodies, and is further evidence that alpha-synuclein is closely related to PD. Alpha-Synuclein has several structural states during the aggregation process: Monomers, intermediates, intermediate oligomers, alpha-synuclein fibrous aggregates, or amorphous aggregates [19,20]. Alpha-synuclein is normally in the form of a monomer, in a curl-free state, soluble in aqueous solution. Usually part of the intracellular Alpha-synuclein is present in the cytoplasm and a part is bound to the membrane. Alpha-synuclein agglutination was induced by rotenone in the cells. Alpha-synuclein oligomers were detected from the membrane components after 24h, but the cytoplasmic components could not be detected aggregates of synuclein after 48 h [21]. In addition, the Alpha-synuclein in the membrane fraction promotes the aggregation of the Alpha-synuclein in the cytoplasmic component. Therefore, Lee think that membrane-bound Alpha-synuclein, although only a fraction of total Alpha-synuclein in the brain, but may play an important role in the Alpha-synuclein aggregation. The main pathological features of Parkinson's disease is the progressive loss of dopaminergic neurons in the midbrain and the formation of Lewy bodies in the dopaminergic neurons, and why the death of DA neurons occurs. This has led the researchers to focus on the relationship between DA and Alpha-synuclein [22]. Conway found in the study, DA can inhibit Alpha-synuclein fiber formation, the mechanism is DA through oxidative connect Alpha-synuclein fibrils to form covalent compounds, stabilize the structure of fibrils, inhibition of fibrils to the fiber , DA-induced fibril accumulation is expected to aggravate cell damage, which may be one of the causes of DA neuron-specific death [23].

2.2 Alpha-synuclein distribution and biological activity

Alpha-synuclein is a soluble protein expressed in the presynaptic and pericentral regions of the central nervous system. It is closely related to the pathogenesis of Parkinson's disease and related dysfunction. It is the main component of Lewy bodies. Its structure depends largely on its intracellular environment, and will show different structures such as monomers, oligomers, fibrils and fibers, etc., pathological state of synuclein easily aggregated to form insoluble Of the fibrin precipitation, leading to nerve cell death [24]. Studies of human genetics have shown that the Alpha-synuclein gene mutations are the major pathogenic states in familial Parkinson's disease, and that the aggregation of Alpha-synuclein has similar prion-like characteristics of intercellular transmission . A-synuclein is functionally diverse and may be involved in many aspects of synaptic structural maintenance, neural plasticity, learning, memory, development, cell adhesion, phosphorylation, cell differentiation, and uptake of dopamine. Alpha-synuclein is mainly expressed in neuronal remodeling sites and is involved in the remodeling of synapses. Osterova [26] found that Alpha-synuclein can regulate nerve transmission to affect neural plasticity. Steidl [27] found that transgenic mouse models (overexpressing human Alpha-synuclein, including A30P Parkinson's disease mutant) to study the synaptic plasticity of the hippocampus and found that when the neurotransmitter utilization is limited, The mutant accumulation of alpha-synuclein attenuates synaptic concentrations. Alpha-synuclein is released mainly from neurons into the extracellular space, both as part of normal extracellular processing, and can cause oxidative and digestive stress and neurodegeneration [28]. A large number of Alpha-synuclein released by denatured neurons produce cytotoxicity through different mechanisms. Previously study demonstrated that NMDA receptor-mediated NO synthesis can be activated by exposure to Alpha-synuclein in the form of extracellular monomers and oligomers for a short time [29].

2.3 Alphay-synuclein metal binding characteristics

Metals often play a role as cofactors for proteins. They are necessary either for structural stability, such as the zinc cofactor of superoxide dismutase, or for a role in catalytic activities of enzymes.The reason for the investigation into metal binding to Alpha-synuclein is the possible insight that it provides for its function. As metal are often cofactors associated with catalytic activity, this could provide a key piece of evidence linking the protein to a biochemical function.

2.3.1 Alpha-synuclein binding copper

It has previously been suggested that Alpha-synuclein is a copper binding protein[30].Initial results showed Alpha-synuclein to bind 5–10 molecules of copper [31] while a later study showed Alpha-synuclein to bind copper at two high affinity sites with more copper able to bind at other lower affinity sites.The co-ordination of copper has been investigated and two potential sites of metal interaction have been defined. In particular, a site at the N-terminus has been shown to be the high-affinity site [32]. Cu(II) mainly interacts with residues located in the N-terminal region of Alpha-synuclein[33]. Copper is normally a cofactor associated with electron transfer in oxidation and reduction reactions. Alpha-synuclein with copper bound showed a strong ability to cycle between oxidized and reduced forms. This implies that, in terms of copper binding to the protein, it could play a role in catalytic processes of electron donation. We propose that the concentrations of Cu(II) in neurons and glia are sufficient to contribute to potential abnormal interaction with proteins such as Alpha-synuclein under certain adverse circumstances. Although more studies are needed to explore other biological aspects of Cu(II)–Alpha-synuclein interactions, the structural features emerging from this work indicate that perturbations in copper metabolism may constitute a more widespread element in neurodegenerative disorders than has been recognized previously.

2.3.2 Alpha-synuclein binding iron

While analysis of copper binding to Alpha-synuclein has been relatively extensive, there has been little consideration of iron in this regard,other study have shown that Alpha-synuclein can bind two different metals simultaneously. Cu(II) saturate Alpha-synuclein was still able to bind Fe(III) with little change in affinity. This clearly indicates that the two metals occupy different sites on the protein.The majority of data suggest that copper binds to the N-terminus of Alpha-synuclein while iron binds to the C-terminus.Both copper and iron can bind to the protein under normal physiological conditions and allow it catalyse the reduction of Fe(III) to Fe(II).

3. Iron metabolism

3.1 The body of iron absorption

Iron in heme iron and inorganic iron in two different forms exist in food. The iron in the food is absorbed from the duodenum to compensate for daily iron loss. DcytB, or duodenal cytochrome B, is a trivalent iron reductase on the cell membrane of the small intestine. It is expressed in descending order on the apical membrane of the small intestine. Iron in food (mainly ferric iron) through the apical membrane of small intestinal cells of duodenal cytochrome b reduced to ferrous iron, which can be divalent metal ion transporter 1 transport and then absorbed by the body [29] . The iron-depleted iron-transport protein on the basal surface of duodenal villi epithelial cells, which is not used or stored by intestinal cells, is transported into the bloodstream and bound to the major iron transferrin transferrin. Iron transport by the membrane iron transporter is dependent on ferric and ferrous iron ions that are activated by multi-copper oxidase (plasma ceruloplasmin present in the blood circulation and membrane iron transporting auxiliary protein in the basolateral membrane of the intestinal cell) [34]. Although a variety of transporters have been demonstrated [35], the mechanism of heme absorption and release in the intestine remains unclear, since the role of these transporters is not yet fully understood clear.

3.2 intracellular iron metabolism

As with systemic iron homeostasis, the maintenance of intracellular unstable iron iron levels is achieved by coordinating the expression of transporters involved in iron uptake, iron transfer, iron storage, and iron utilization. This process is regulated by multiple steps, but the post-transcriptional regulation mediated by IRE and IRPs is the most fundamental and important. Because intracellular iron metabolism is IRP1 and IRP2 effect with a variety of different mRNAs in the cis-acting element[36].

3.3 Alpha-synuclein is involved in the iron metabolism of microglia

Recent studies have confirmed that high expression of Alpha-synuclein does induce iron accumulation in neurons. In the small intestine, Fe^{3+} is first reduced to Fe^{2+} by the duodenal cytochrome b (Dcytb) (iron reductase) on the luminal membrane of small intestinal cells and then transferred to the cells via DMT1. SDR2 (stromal cell-derived receptor 2) is a homologue of Dcytb in the choroid plexus and the ependymal cells of the fourth ventricle. The expression of SDR2 in substantia nigra neurons suggests that SDR2 may act as an iron reductase aided DMT1 need for further studies to confirm. Recent studies have shown that Alpha-synuclein is an iron-reductase that can metabolize Fe^{3+} in the cell. Studies have shown that all three mutant forms of the Alpha-synuclein have iron reductase activity [37] and are not different from wild-type. Excessive amounts of Fe^{2+} are produced when too much Alpha-synuclein is expressed, leading to excessive iron transfer to DMT1-mediated cells, whereas excessive Fe^{2+} generates large amounts of hydroxyl radicals through the Fenton reaction resulting in DA neuronal damage. The deposition of iron further aggregates the Alpha-synuclein, leading to a vicious cycle. Alpha-synuclein has three aggregated forms, three forms have different biological functions. The monomer interacts with a variety of phospholipids, which disrupt negatively charged vesicles, whereas polymers can act on negatively charged vesicles and cell membranes [38]. Studies have shown that oligomeric and multimeric forms of Alpha-synuclein on cytotoxicity through the interaction with the cell membrane and destroy membrane integrity, but the current study is still not to reach a consensus on the three forms of Alpha-synuclein protein toxicity (Figure 2) [39].

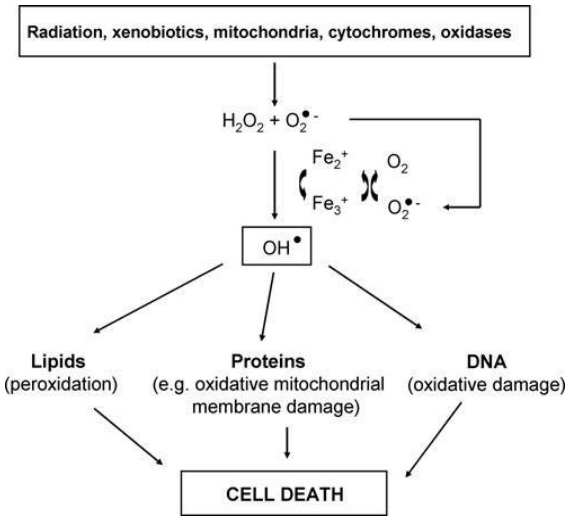

Figure 2 Effect of reactive oxygen species leading to cell damage and death

A number of different stimuli can result in the production of moderately active hydrogen peroxide and superoxide (O_2^- and H_2O_2), which react with iron and catalyze the reactive toxic hydroxyl (O_2^- and H_2O_2) free radical formation, this hydroxyl radicals can damage lipids, proteins and nucleic acids important cell macromolecules and eventually lead to cell death.

4. Conclusion

Microglial cells are central nervous system innate immune effector cells that respond to different stimuli, such as damage, neurodegeneration, stroke, and brain tumors. In a healthy brain, the microglia are in a quiescent state, and they search for tissue damage and intruders. If tissue damage or infection, microglia will be transformed into activated and phagocytic state, mainly for the morphological changes, amplification, neurotrophic substances (such as: BDNF) or inflammatory factors (such as: tumor necrosis factor TNF- α) and increased oxidative stress (eg, production of reactive oxygen species ROS). A number of studies exploring Alpha-synuclein -dependent glial activation, particularly in mutant or aggregated forms of Alpha-synuclein, have been shown to lead to stronger activation of microglia and astrocytes, release of cytokines and oxidative stress. In addition, recent evidence suggests that activation of microglia and astrocytes may lead to the degradation of

dopaminergic neurons, regulating the development of neurodegenerative diseases such as PD. Our lab study in recent years have been proved real high iron can lead to dopaminergic neuron death in substantia nigra par compacta. Neuropathology study have found Alpha-synuclein in abnormal aggregation often accompany with iron deposits,it is prominent indicate that Alpha-synuclein aggregation is related with iron deposits [13] and Alpha-synuclein maybe involved in the iron metabolism of microglia.Therefore, it is necessary to determine the Alpha-synuclein toxicity. Understanding the effect of Alpha-synuclein on microglia iron transport is the main goal and significance of future research.

References

[1] Fellner L, Irschick R, Schanda K, et al. Toll‐like receptor 4 is required for α‐synuclein dependent activation of microglia and astroglia[J]. Glia, 2013, 61(3):349-360.

[2] Watson M B, Richter F, Lee S K, et al. Regionally-specific microglial activation in young mice over-expressing human wildtype alpha-synuclein.[J]. Experimental Neurology, 2012, 237(2): 318–334.

[3] Pei Zhengbin, Peng Guoguang. Dual role of microglia in Parkinson's disease [J]. Chinese Journal of Rehabilitation, 2004 (1): 130-132.

[4]Alvarez-Erviti L, Couch Y, Richardson J, Cooper JM, Wood MJ. Alpha-synuclein release by neurons ativates the inflammatory response in a microglial cell line. J Neurosci Res. 2011; 69:337–342.

[5]Weijiang W, Junfei S, Hua L, et al. Guard of Delinquency? A Role of Microglia in Inflammatory Neurodegenerative Diseases of the CNS.[J]. Cell Biochemistry & Biophysics, 2014, 70(1):1-8.

[6]Cairo G, Bernuzzi F, and Recalcati S. A precious metal: Iron, an essential nutrient for all cells. Genes Nutr 2006,1: 25–39.

[7]Zhang AS and Enns CA. Iron homeostasis: Recently identified proteins provide insight into novel control mechanisms. J Biol Chem 2009,284: 711–715.

[8]Church C, Lee S, Bagg EA, McTaggart JS, Deacon R, Gerken T, Lee A, Moir L, Mecinovic J, Quwailid MM, Schofield CJ, Ashcroft FM, and Cox RD. A mouse model for the metabolic effects of the human fat mass and obesity associated FTO gene. PLoS Genet2009, 5: e1000599.

[9]Iyer LM, Tahiliani M, Rao A, and Aravind L. Prediction of novel families of enzymes involved in oxidative and other complex modifications of bases in nucleic acids. Cell Cycle 2009, 8: 1698–1710.

[10]Love S. Oxidative Stress in Brain Ischemia[J]. Brain Pathology, 1999, 9(1):119–131.

[11]FradeJM ,Barde YA .micriglia-derived nerve growth factor caused cell death in thedeveloping retina.Neuron 1998;20(1):35-41

[12]Weijiang W, Junfei S, Hua L, et al. Guard of Delinquency? A Role of Microglia in Inflammatory Neurodegenerative Diseases of the CNS.[J]. Cell Biochemistry & Biophysics, 2014, 70(1):1-8.

[13]Castano A, Herrera AJ, Cano J, and Machado A (2002) The degenerative effect of a single intranigral injection of LPS on the dopaminergic system is prevented by dexamethasone and not mimicked by rh-TNF- α , IL-1 β and IFN- r . J Neurochem81:150–157.

[14]Chang RC, Rota C, Glover RE, Mason RP, and Hong JS (2000b) A novel effect of anopioid receptor antagonist, naloxone, on the production of reactive oxygen species by microglia: a study by electron paramagnetic resonance spectroscopy. Brain Res854:224–229.

[15]Chang RC, Chen W, Hudson P, Wilson B, Han DS, and Hong JS (2001) Neurons reduce glial responses to lipopolysaccharide (LPS) and prevent injury of microglial cells from over- activation by LPS. J Neurochem 76:1042–1049.

[16]Maroteaux L;Campanelli J T;Scheller R H Synuclein:a neuron-specific protein localized to thenucleus and presynaptic nerve terminal 1988(08)

[17] Conway K A, Lee S J, Rochet J C, et al. Acceleration of oligomerization, not fibrillization, is a shared property of both alpha-synuclein mutations linked to early-onset Parkinson's disease:

implications for pathogenesis and therapy.[J]. Proceedings of the National Academy of Sciences, 2000, 97(2):571-6.
[18] Kessler J C, Jeanchristophe Rochet A, Lansbury P T. The N-Terminal Repeat Domain of α-Synuclein Inhibits β-Sheet and Amyloid Fibril Formation†[J]. Biochemistry, 2003, 42(3): 672-8.
[19]Agata Adamczyk*, Anna Kaz ́mierczak, Grzegorz Arkadiusz Czapski, Joanna Benigna Strosznajder(2010) a -Synuclein induced cell death in mouse hippocampal (HT22) cells is mediated by nitric oxide-dependent activation of caspase-3.FEBS Letters 584 (2010) 3504–3508.
[20] Fortin, D.L., Nemani, V.M., Nakamura, K. and Edwards, R.H. (2010) The behavior of a -synuclein in neurons. Mov. Disord. 25, S21–S26.
[21]Norris, E.H., Giasson, B.I. and Lee, V.M. (2004) Alpha-synuclein: normal function and role in neurodegenerative diseases. Curr. Top. Dev. Biol. 60, 17–54.
[22]Sung J Y, Kim J, Paik S R, et al. Induction of Neuronal Cell Death by Rab5A-dependent Endocytosis of α-Synuclein[J]. Journal of Biological Chemistry, 2001, 276(29):27441-27448.
[23]Kaufman SK, Diamond MI. Prion-like propagation of protein aggregation and related therapeutic strategies. Neurotherapeutics. 2013; 10:371–382.
[24] Luk KC, et al. Exogenous alpha-synuclein fibrils seed the formation of Lewy body-like intracellular inclusions in cultured cells. Proc Natl Acad Sci U S A. 2009; 106:20051–20056.
[25]Agata, Adamczyk, Anna Kaz, mierczak, Grzegorz Arkadiusz Czapski, Joanna Benigna Strosznajder a -Synuclein induced cell death in mouse hippocampal (HT22) cells is mediated by nitric oxide-dependent activation of caspase-3.FEBS Letters 584 (2010) 3504–3508.
[26] Watanabe, Y., Kato, H. and Araki, T. (2008) Protective action of neuronal nitricoxide synthase inhibitor in the MPTP mouse model of Parkinson‘s disease.Metab. Brain Dis. 23, 51–69.
[27]Aquilano, K., Baldelli, S., Rotilio, G. and Ciriolo, M.R. (2008) Role of nitric oxide synthases in Parkinson‘s disease: a review on the antioxidant and antiinflammatory activity of polyphenols. Neurochem. Res. 33, 2416–2426.
[28]Reyes J F, Rey N L, Luc B, et al. Alpha-synuclein transfers from neurons to oligodendrocytes[J]. Glia, 2014, 62(3):387–398.
[29]Ahn KJ, Paik SR, Chung KC, Kim J. 2006. Amino acid sequence motifs and mechanistic features of the membrane translocation of alpha-synuclein. J Neurochem 97:265–279.
[30] Reeves R M. Structural characterization of copper(II) binding to alpha-synuclein: Insights into the bioinorganic chemistry of Parkinson's disease.[J]. Proceedings of the National Academy of Sciences, 2005, 102(12):4294-9.
[31] Lee EN, Lee SY, Lee D, Kim J, Paik SR (2003) Lipid interaction of alpha-synuclein during the metal-catalyzed oxidation in the presence of Cu2+ and H2O2. Journal of Neurochemistry 84: 1128–1142.
[32] Drew S C, Leong S L, Pham C L, et al. Cu2+ binding modes of recombinant alpha- synuclein--insights from EPR spectroscopy.[J]. Journal of the American Chemical Society, 2008, 130(24): 7766-73.
[33] Uversky VN, Li J, Fink AL. Metal-triggered structural transformations, aggregation, and fibrillation of human alpha-synuclein. A possible molecular NK between Parkinson's disease and heavy metal exposure.[J]. Journal of Biological Chemistry, 2001, 276(47):44284-44296.
[34]Wessling–Resnick M. Iron imports. III. Transfer of iron from the mucosa into circulation. Am J Physiol Gastrointest Liver Physiol 2006,290: G1–6.
[35]Latunde–Dada GO, Takeuchi K, Simpson RJ, and McKie AT. Haem carrier protein 1 (HCP1): Expression and functional studies in cultured cells. FEBS Lett 2006,580: 6865–6870.
[36]Breuer W, Shvartsman M, and Cabantchik ZI. Intracellular labile iron. Int J Biochem Cell Biol 2008, 40: 350–354.
[37]Brown D R. α-Synuclein as a ferrireductase.[J]. Biochemical Society Transactions, 2013, 41(6): 1513-1517.

[38]Clayton, D.F. and George, J.M. (1998) The synucleins: a family of proteins involved in synaptic function, plasticity, neurodegeneration and disease.Trends Neurosci. 21, 249–254.
[39]Dettmer, U., Newman, A.J., Luth, E.S., Bartels, T. and Selkoe, D. (2013) In vivo crosslinking reveals principally oligomeric forms of alpha-synuclein and beta-synuclein in neurons and non-neural cells. J.Biol. Chem. 288, 6371–6385.