NEURAL REGENERATION RESEARCH
Volume 7, Issue 5, February 2012
www.nrronline.org

Cite this article as: Neural Regen Res. 2012;7(5):376-385.

Oxidative stress in neurodegenerative diseases☆

Xueping Chen[1], Chunyan Guo[2], Jiming Kong[1]

1*Department of Human Anatomy and Cell Science, University of Manitoba, Manitoba, Canada*
2*Department of Pharmacy, Hebei North University, Zhangjiakou 075000, Hebei Province, China*

Abstract

Reactive oxygen species are constantly produced in aerobic organisms as by-products of normal oxygen metabolism and include free radicals such as superoxide anion (O_2^-) and hydroxyl radical (OH^-), and non-radical hydrogen peroxide (H_2O_2). The mitochondrial respiratory chain and enzymatic reactions by various enzymes are endogenous sources of reactive oxygen species. Exogenous reactive oxygen species -inducing stressors include ionizing radiation, ultraviolet light, and divergent oxidizing chemicals. At low concentrations, reactive oxygen species serve as an important second messenger in cell signaling; however, at higher concentrations and long-term exposure, reactive oxygen species can damage cellular macromolecules such as DNA, proteins, and lipids, which leads to necrotic and apoptotic cell death. Oxidative stress is a condition of imbalance between reactive oxygen species formation and cellular antioxidant capacity due to enhanced ROS generation and/or dysfunction of the antioxidant system. Biochemical alterations in these macromolecular components can lead to various pathological conditions and human diseases, especially neurodegenerative diseases. Neurodegenerative diseases are morphologically featured by progressive cell loss in specific vulnerable neuronal cells, often associated with cytoskeletal protein aggregates forming inclusions in neurons and/or glial cells. Deposition of abnormal aggregated proteins and disruption of metal ions homeostasis are highly associated with oxidative stress. The main aim of this review is to present as much detailed information as possible that is available on various neurodegenerative disorders and their connection with oxidative stress. A variety of therapeutic strategies designed to address these pathological processes are also described. For the future therapeutic direction, one specific pathway that involves the transcription factor nuclear factor erythroid 2-related factor 2 is receiving considerable attention.
Key Words: oxidative stress; neurodegenerative diseases; reactive oxygen species; therapy; reviews

Xueping Chen☆, Doctor, Department of Human Anatomy and Cell Science, University of Manitoba, Manitoba, Canada

Xueping Chen and Chunyan Guo contributed equally to this work.

Corresponding author: Jiming Kong, Department of Human Anatomy and Cell Science, University of Manitoba, Manitoba, Canada
kongj@cc.umanitoba.ca

Received: 2011-09-24
Accepted: 2011-11-22
(N20111019001/H)

Chen XP, Guo CY, Kong JM. Oxidative stress in neurodegenerative diseases. Neural Regen Res. 2012;7(5):376-385.

www.crter.cn
www.nrronline.org

doi:10.3969/j.issn.1673-5374.2012.05.009

INTRODUCTION

Free radicals are molecules with at least one unpaired electron in the outermost shell; they are highly reactive due to the presence of unpaired electron. Any free radical involving oxygen can be referred to as a reactive oxygen species (ROS)[1]. Since ROS are common outcome of normal aerobic cellular metabolism, in-built antioxidant system of body plays its decisive role in prevention of any loss due to ROS overproduction. Oxidative stress arises as a result of an imbalance between the production of ROS and the biological system's ability to detoxify the reactive intermediates[2]. Oxidative stress has been implicated in the progression of Alzheimer's disease (AD), Parkinson's disease (PD) and other neurodegenerative diseases. Oxidative stress leading to free radical attack on neural cells contributes calamitous role to neurodegeneration. Toxicity of ROS contributes to protein misfolding, glia cell activation, mitochondrial dysfunction and subsequent cellular apoptosis[3]. However, the systems in place to cope with biochemistry of oxidative stress are complex, and this complexity provides a number of therapeutic targets. Recognition of upstream and downstream antioxidant therapy has been proved an effective tool in alteration of neuronal damage as well as novel metal-protein attenuating compound (MPAC). Furthermore, therapeutic approaches aiming at nuclear factor erythroid 2-related factor 2 (Nrf2) transcriptional pathway have shown promise in clinical studies. This review presents detailed information on oxidative stress and its connection with neurodegenerative diseases. The therapeutic strategies designed to address these diseases are also described.

ROS GENERATION

Free radicals, with at least one unpaired electron in the outermost shell, is highly reactive[4]. The most common reported cellular free radicals are hydroxyl (OH•), superoxide ($O_2^{\bullet -}$), nitric oxide (NO•), nitrogen dioxide (NO_2•), peroxyl (ROO•) and

lipid peroxyl (LOO•). Molecules such as hydrogen peroxide (H_2O_2), ozone (O_3), singlet oxygen ($1O_2$), hypochlorous acid (HOCl), nitrous acid (HNO_2), peroxynitrite ($ONOO^-$), dinitrogen trioxide (N_2O_3), lipid peroxide (LOOH), while not considered free radicals, can easily lead to free radical reactions in living organisms[5]. Cells exposed to environment fortified with oxygen continuously generate oxygen free radicals. ROS includes oxygen-related free radicals and reactive species[6], and they are produced as a result of aerobic metabolism. Formation of ROS can occur in two ways: enzymatic and non-enzymatic reactions. Enzymatic reactions generating free radicals include those involved in the mitochondrial respiratory chain, phagocytosis, prostaglandin synthesis and the cytochrome P450 system[7]. For example, the superoxide radical is generated *via* several cellular oxidase systems such as 5,10-methylenetetrahydrofolate reductase oxidase, xanthine oxidase, peroxidases. ROS can also be produced from non-enzymatic reactions of oxygen with organic compounds as well as those initiated by ionizing radiations. The non-enzymatic process can also occur during oxidative phosphorylation (*i.e.* aerobic respiration) in the mitochondria[8]. ROS is generated from either endogenous or exogenous sources. Endogenous free radicals are generated from immune cell activation, inflammation, mental stress, excessive exercise, ischemia, infection, cancer and aging. Exogenous ROS result from air and water pollution, cigarette smoke, alcohol, heavy or transition metals (Cd, Hg, Pb, Fe, As), certain drugs (cyclosporine, tacrolimus, gentamycin, bleomycin), industrial solvents, cooking (smoked meat, used oil, fat) and radiation[9-12]. After penetrated into the body by different routes, these exogenous compounds are decomposed or metabolized into free radicals. Generation of ROS can occur in various organelles, mitochondria is the main source of ROS production[13-14]. The generation of mitochondrial ROS is a consequence of oxidative phosphorylation, a process that occurs in the inner mitochondrial membrane and involves the oxidation of reduced form of nicotinamide-adenine dinucleotid to produce energy. Mitochondrial electron transport involves four-electron reduction of O_2 to H_2O. However, during mitochondrial electron transport, a one-electron reduction of O_2 results in superoxide ($O_2^{\bullet-}$). Superoxide anion is detoxified by the mitochondrial manganese superoxide dismutase to yield H_2O_2, and H_2O_2 in the presence of reduced transition metals can also be converted to hydroxyl radical (OH^-). For the ROS is not originated from mitochondria, peroxisomal β-oxidation of fatty acids was considered to be a second source of oxygen radicals. This reaction generates H_2O_2 as a by-product. Peroxisomes are organelles responsible for degrading fatty acids as well as other molecules[14]. Phagocytic cells are another important source of oxidants; these cells defend the central nervous system (CNS) against invading microorganisms and clear the debris from damaged cells by an oxidative burst of nitric oxide, H_2O_2, and O_2^-. Finally, cytochrome P450 enzymes in animals are one of the first defenses against natural toxic chemicals from plants. In addition, the generation of ROS in living systems is closely linked with the participation of redox-active metals such as iron and copper. As a general principle, the chemical origin of the majority of ROS is the direct interactions between redox-active metals and oxygen species *via* reactions such as the fenton and haber-weiss reaction. Free iron (Fe^{2+}) reacts through the Fenton reaction with H_2O_2, leading to the generation of very reactive and damaging hydroxyl radicals. Superoxide can also react with ferric iron in the Haber-Weiss reaction leading to the production of Fe^{2+}, which then again affects redox cycling[15]. Apart from direct ROS generation, indirect pathway involves calcium activation with metallo-enzymes such as phospholipases, nitric oxide synthase. Calcium stimulates the tricarboxylic acid cycle and enhances electron flow into the respiratory chain; it also stimulates the nitric oxide synthase and subsequently promotes nitric oxide generation, which would inhibit respiration at complex IV. These events would enhance ROS generation from Q cycle. Calcium is an important signaling molecule and it is required for many cellular responses and cell-cell communication. Thus, any disruption of calcium homeostasis may disrupt the cellular physiology[16].

BENEFICIAL AND DELETERIOUS ACTIVITIES OF ROS

At low or moderate concentrations, ROS are necessary for the maturation process of cellular structures and can act as weapons for the host defense system, supporting cell proliferation and survival pathways. Indeed, phagocytes (neutrophils, macrophages, monocytes) release free radicals to destroy invading pathogenic microbes as part of the body's defense mechanism against disease[7]. Other beneficial effects of ROS involve their physiological roles in the function of a number of cellular signaling systems. ROS signaling can affect cellular energetics by acutely regulating adenosine-triphosphate production *via* activation of uncoupling proteins[17]. Moreover, ROS are required for transduction growth signals *via* certain receptor tyrosine kinases[18]. Specific example includes nitric oxide, which is an intercellular messenger for modulating blood flow, thrombosis, and neural activity[19]. Nitric oxide is also important for nonspecific host defense, and for killing intracellular pathogens and tumors. Another beneficial activity of free radicals is the induction of a mitogenic response[19]. In brief, ROS at low or moderate levels are vital to human health. When produced in excess, these highly reactive radicals can start a pathological chain reaction, like dominoes[20], damaging all components of the cells, and leading to a progressive decline in physiological function[21]. This will generate a phenomenon called oxidative stress. Oxidative stress is

a deleterious process that can seriously alter the cell membranes and other structures such as proteins, lipids, lipoproteins, and deoxyribonucleic acid (DNA)[3, 6]. Oxidative stress can arise when cells cannot adequately destroy the excess of free radicals formed. In other words, oxidative stress is caused by an imbalance between the production of reactive oxygen and a biological system's ability to detoxify the reactive intermediates. Furthermore, in the CNS, ROS production has been linked to another key feature of neurodegenerative diseases, such as accumulation of protein aggregates, increase in intracellular free Ca^{2+}, release of excitatory amino acids, autophagy and apoptosis, and all mechanisms play a critical role in the pathogenesis of many neurological disorders, such as PD, AD and amyotrophic lateral sclerosis[22].

For example, ROS overproduction within mitochondria can lead to oxidative damage to mitochondrial proteins, membranes, and mitochondrial DNA, finally resulting in mitochondrial injury[23]. ROS affect the heme-containing cytochrome c oxidase I molecule of complex IV of the respiratory chain, as well as induce additional damage to complex I, II, and III components[24]. Mitochondrial oxidative damage leads to the release of cytochrome c into the cytosol resulting in apoptosis. Increased permeability makes the inner membrane permeable to small molecules. Despite a large amount of scientific evidence supporting oxidative stress as a pathogenic factor in these diseases, human also experience with antioxidant neuroprotectants[2]. The body counteracts oxidative stress by producing antioxidants, either naturally generated *in situ* (endogenous antioxidants), or externally supplied through foods (exogenous antioxidants). The role of antioxidants is to neutralize excess of free radicals, protecting the cells against their toxic effects, and to contribute to disease prevention. However, overproduction of ROS which could not be fully neutralized can cause oxidative damage to biomolecules, and eventually leading to many chronic diseases, such as atherosclerosis, cancer, diabetics, rheumatoid arthritis and degenerative diseases.

OXIDATIVE STRESS IN NEURODEGENERATIVE DISEASES

Neurodegenerative diseases are clinically characterized by their insidious onset and chronic progression, and are pathologically characterized by progressive dysfunction and death of cells that frequently affect specific neural system. Morphologically, neuronal loss is associated with gliosis and frequently, with misfolding and aggregation of proteins leading to the relentless accumulation of abnormal extracellular and intracellular filamentous deposit in specific cell types, representing the core features/hallmarks of many neurodegenerative disorders. While many brain neurons can cope with a rise in oxidative stress, there are selected populations of neurons that are vulnerable to increase oxidative stress, this phenomenon in neurodegenerative conditions is called selective neuronal vulnerability[25]. Selective neuronal vulnerability refers to the differential sensitivity of neuronal populations in the CNS to stresses that cause cell injury or death and lead to neurodegeneration. For example, neurons in the entorhinal cortex, hippocampal CA1 region, frontal cortex, and amygdala are the populations of neurons most sensitive to the neurodegeneration associated with AD. In PD, dopaminergic neurons of the substantia nigra are the primary neurons undergoing cell death[26]. Amyotrophic lateral sclerosis is characterized by the degeneration of, primarily, spinal motor neurons, but also cortical and brain stem neurons[27]. The fact that specific brain regions exhibit differential vulnerabilities to oxidative stress in various neurodegenerative diseases is a reflection of the specificity in the etiology of each disease, and it is possible that the selected cells involved in the pathology of neurodegenerative diseases may share a common increased vulnerability to the detrimental effects of oxidative stress. Neuronal cells are highly sensitive to oxidative stress, because (1) their large dependence on oxidative phosphorylation for energy as compared with other cells; (2) they are exposed to high oxygen concentration, utilizing about 20% of respired oxygen, even though the brain represents only 5% of the body weight. Under physiological condition, 1–2% of consumed O_2 is converted to ROS, leading to oxidative stress, and this percentage goes up dramatically in aged brain[28]; (3) they are enriched in metal ions, which accumulated in the brain as a function of age and can be a potent catalyst for oxidative species formation; (4) they are rich in polyunsaturated fatty acids that are prone to oxidation; (5) they contain relatively poor concentrations of antioxidants and related enzymes. The brain is lower in antioxidant activity in comparison with other tissues, for example, about 10% of the liver. Under normal conditions, cells are capable of counteracting the oxidant insults by regulating their homeostatic balance. However, during the progression of age-related neurodegenerative conditions, the capacity of cells to maintain the redox balance decreases, leading to the accumulation of free radicals, mitochondrial dysfunction, and neuronal injury. It is widely accepted that oxidative stress increases during aging[29], and it can be considered as an important age-dependent factor making the neuronal systems more susceptible to several neurodegenerative diseases such as AD and PD.

EVIDENCE OF OXIDATIVE STRESS IN NEURODEGENERATIVE DISEASES

Oxidative overload in the neuronal microenvironment causes oxidation of lipids[30], proteins[31] and DNA[32] and generates many by-products such as peroxides, alcohols, aldehydes, ketones and cholesterol oxide[33]. (1) Lipid oxidation. Unsaturated lipids are particularly susceptible to oxidative modification and lipid peroxidation is a

sensitive marker of oxidative stress. Lipid peroxidation is the result of attack by radicals on the double bond of unsaturated fatty acids to generate highly reactive lipid peroxy radicals that initiate a chain reaction of further attacks on other unsaturated fatty acids. The chain reaction leads to the formation of breakdown products including 4-hydroxy-2, 3-nonenal (HNE) and F_2-isoprostanes[34-37]. HNE is able to modify proteins, resulting in a multitude of effects, including inhibition of neuronal glucose and glutamate transporters, inhibition of Na^+-K^+-ATPases, activation of kinases and dysregulation of intracellular calcium signaling, that ultimately induce an apoptotic cascade mechanism[38-40]. These findings, together with the recent demonstration that HNE is cytotoxic to neurons and that it impairs the function of membrane proteins including the neuronal glucose transporter 3, indicate that HNE is a characteristic marker and a toxin leading to neurodegeneration[41]. (2) Protein oxidation. ROS mediated oxidation of protein side-chains and resulted in the introduction of hydroxyl groups or in the generation of protein based carbonyls[42]. Carbonyl groups are introduced in proteins by oxidizing amino acid residue side-chain hydroxyls into ketone or aldehyde derivatives[43]. Carbonyl groups can also be introduced in proteins by direct oxidation of lysine, arginine, proline and threonine residues, or from the cleavage of peptide bonds by the α-amidation pathway or by the oxidation of glutamyl residues[44]. Measurement of protein carbonylation is thought to be a good estimate for the extent of oxidative damage of proteins associated with various conditions of oxidative stress[45-47]. (3) DNA oxidation. DNA bases are vulnerable to oxidative stress damage involving hydroxylation, carbonylation and nitration[48-50]. DNA and RNA oxidation is marked by increased levels of 8-hydroxy-2-deoxyguanosine and 8-hydroxyguanosine[51-53]. It is now widely accepted that oxidative damage is responsible for DNA strand breaks and this is consistent with the increased free carbonyls in the nuclei of neuronal cells in neurodegenerative diseases. (4) Glycoxidation. Advanced glycation end products (AGEs), which are formed by a non-enzymatic reaction of sugars with long lived protein deposits, are also potent neurotoxins and proinflammatory molecules. A cascade of reactions results thereafter in the formation of AGEs, which are composed of irreversibly cross-linked heterogeneous protein aggregates[54]. Accumulation of extracellular AGEs in neurodegenerative diseases is caused by an accelerated oxidation of glycated proteins (“glycoxidation”)[55].

Protein aggregation and oxidative stress in neurodegenerative diseases

Abnormal interactions between proteins that result in aberrant intracellular and extracellular deposition of self aggregating misfolded proteins with formation of high-ordered insoluble fibrils are common pathological hallmarks of multiple neurodegenerative disorders. Although the pathogenicity of protein aggregates remains uncertain[56], a causative link between the formation of protein aggregates and neurodegeneration has been established, which may occur as a result of the toxic action of substances produced during early phases, and soluble oligomers and protofibrillar derivatives of misfolded proteins may play a pathogenic role[57-58]. The exact mechanisms of abnormal folding are not fully understood; however, speculations lead to the presumption that genetic and environmental factors (especially oxidative stress) are involved[59]. Aberrant proteins, the result of inherited or acquired amino acid substitution or damage, especially oxidative modification, cannot fold correctly and will be trapped in misfolded conformations. Growing evidence supports the hypothesis that oxidative stress, combined with protein aggregation, triggers a cascade of events leading to cell death in multiple neurodegenerative diseases. Because proteins modified by oxidative reactive species tend to form aggregates, and highly oxidized and cross-linked proteins may act as endogenous inhibitors of proteasomal activity. Since the proteasome represents the major proteolytic machinery for the removal of oxidized and misfolded protein[60], inhibition of the proteasome or decreasing proteasomal activity will result in an accumulation of abnormal proteins[61]. Therefore, timely removal of oxidatively damaged proteins is of critical importance to maintain normal cellular homeostasis and viability. If homeostasis is not restored, cells ultimately undergo apoptotic or necrotic cell death. To prevent cytotoxicity induced by oxidized proteins, normal proteasome-dependent degradation is essential for cells to cope with oxidative stress[62]. In a vicious cycle, proteasomal dysfunction can lead to decreased degradation of misfolded proteins, resulting in accumulation of oxidized proteins and subsequent protein aggregation. Protein aggregates can then feedback to further inhibit proteasome activities, stimulate reactive species formation, and lead to cytotoxicity and human pathologies. Such phenomena have been implicated in many oxidative stress-associated disorders, including neurodegenerative diseases[63-64].

Metal ions and oxidative stress in neurodegenerative diseases

Metals are known to play a fundamental role in numerous essential metabolic processes of living systems. Homeostasis of metal ion is maintained through tightly regulated mechanism of uptake, storage, and secretion[65]. In the brain, the movement of metals across the blood brain barrier is highly regulated, and there is no passive flux of metals from the circulation to the brain[66]. Since the generation of free radicals is closely linked with the participation of redox-active metals, the disruption of metal homeostasis may lead uncontrolled metal-mediated formation of deleterious free radicals participating in pathogenesis of neurodegenerative disease. While iron, copper, and zinc are being increasingly implicated in interaction with

major protein components of neurodegenerative diseases, this is not merely due to increased exposure to metals but rather because a breakdown in the homeostasis mechanisms that compartmentalize and regulate metals[67]. The ability of metal ions to accept and donate electrons can lead to the formation of ROS, which may trigger the oxidative stress, therefore contributing to disease and perhaps aging itself[68-69]. Increasing age is a dominant risk factor associated with the neurodegenerative diseases. Several studies in mice have shown that one of the consequences of normal aging is a rise in the levels of copper and iron in brain tissue[70-71]. The brain is an organ that concentrates metal ions and recent evidence suggests that a breakdown in metal homeostasis is a key factor in a variety of age-related neurodegenerative diseases. Impaired iron metabolism is a hallmark in several neurodegenerative diseases such as PD and AD, multiple sclerosis, amyotrophic lateral sclerosis, and neuroferritinopathies. In the case of PD and AD, iron has been shown to play a key role in neuronal fate: depending on the extent and intensity of the oxidative stress caused by the increase in the labile iron pool, it affects transcriptional activity and signaling cascades that could participate in neuronal survival or death[72]. Furthermore, in several age-dependent neurodegenerative disorders, the proteins might abnormally present Cu^{2+} or Fe^{3+} ligands for inappropriate reaction with O_2, examples include β-amyloid in AD, α-synuclein in PD, Cu-Zn superoxide dismutase in amyotrophic lateral sclerosis. These proteins might have some aspect of their function subserved by these metal ions, which normally occupy higher-affinity, embedded, redox-shielded binding sites. As metal concentration rises in the brain with age, the probability increases that a redox-competent, low-affinity metal-binding site will recruit a metal ion from the normally redox-silent cellular pool. In this manner, proteins such as Aβ can harness endogenous biometals to foster the release of inappropriate redox activity and ROS generation.

Oxidative stress in AD

AD is the most common neurodegenerative disease in elderly people. It is characterized by progressive memory deficits, cognitive impairment and personality changes. Pathological features in AD are loss of neurons and synapses in the neocortex, hippocampus and other sub-cortical regions of the brain[73]. The main histological features are extracellular protein deposits called senile Aβ-amyloid plaques and intraneuronal neurofibrillary tangles[73]. Oxidative stress plays a major role in AD, believed to be stronger than in other neurodegenerative diseases[74]. In addition, an accumulation of misfolded protein in the aging brain results in oxidative and inflammatory damage, which in turn leads to energy failure and synaptic dysfunction[75]. Oxidative damage in AD exhibited through increased levels of DNA oxidation products like 8-hydroxydeoxyguanosine in mitochondria and nucleus[76]. Protein carbonyls and 4-HNE are also found to be increased in brain tissues[77]. Elevated levels of oxidized, nitrated and glycated proteins are found in plaques, helical filaments and cerebrospinal cord fluid from AD patients[78-80]. AGEs are found to accumulate in Aβ and neurofibrillary tangles and it could be shown that AGEs induce the release of various potentially neurotoxic inflammatory mediators such as nitric oxide, Il-1 and tumor necrosis factor-a[81]. The activity of the proteasome is also impaired, as hyperphosphorylated tau that is heavily ubiquitinated forms cross-linked aggregates and inhibits the proteasome[82-83]. Transition metals are abnormally distributed in AD as well, studies revealed a marked association between redox-active iron and both Aβ-rich senile plaques and neurofibrillary tangles[84]. It is well known that AD is characterized by Aβ accumulation in senile plaques, and Aβ deposition has also been demonstrated to participate in a positive feedback loop, where oxidative stress leads to increased Aβ generation, and, conversely, the mechanism of Aβ polymerization generates oxidative stress which in turn enhances Aβ production[85]. Additionally, Aβ has been characterized as a metalloprotein, which is able to bind transition metals (*e.g.*, zinc, iron, copper) *via* three histidine (positions 6, 13, and 14) and one tyrosine (position 10) residues located in the hydrophilic N-terminal part of the peptide[86-87]. Binding of Cu^{2+} and Fe^{3+} produce toxic chemical reaction, alter oxidation state both the metals, produce H_2O_2 catalytically in presence of transition metals. The H_2O_2 can initiate a number of different events, including Fenton reactions to form toxic hydroxyl radicals and calcium dysregulation. As calcium is pivotal in signal transduction, it can induce further production of ROS and elicit an excitotoxicity response. In health, soluble Aβ is not present in the cortical synapse. In AD, soluble oxidized Aβ accumulates within the synapse, at which the high Zn^{2+} concentrations precipitate the copper/iron-metallated Aβ, creating a reservoir of potentially toxic Aβ. Augmented metal ions concentrations and oxidative stress have been found to correlate with changes in the concentration of both soluable and deposited Aβ[88]. When Aβ interacts with these metals, the peptide aggregates, forming toxic oligomers and ultimately amyloid plaques. The toxicity of oligomers is elicited through interactions with the glutamatergic receptors such as the N-methyl-D-aspartate receptor. Interestingly, the metal-dependent generation of ROS by Aβ may be a good target for therapeutics[89]. MPACs such as clioquinol (CQ, 5-chloro-7-iodo-8- hydroxyquinoline) and a copper/zinc ionophore (PBT2) seek to inhibit metal interactions with Aβ and prevent the subsequent formation of ROS and facilitate neuroprotective signaling.

Oxidative stress in PD

PD is characterized by the loss of dopaminergic neurons of the substantia nigra, and the deposition of intracellular inclusion bodies[90-91]. It is the most common movement disorder in elderly people and the second most common

neurodegenerative disease. PD is associated with the appearance of round, intracytoplasmic proteinaceous inclusions lewy dodies. Several cellular components have been found in lewy bodies, including synphilin-1, α-synuclein and others[92]. A characteristic feature of the neurons within the substantia nigra is the age-dependent accumulation of neuromelanin, and these neuromelanin-containing cells are most likely to be lost in PD. Neuromelanin is a dark brown pigment that accumulates metal ions, particularly iron. Significant evidence shows enhanced oxidative stress in PD, because markers of oxidative damage to biological structures, such as lipid, protein, and DNA oxidation are found to be increased in PD. Proteasomal function is affected in the substantia nigra in patients with sporadic PD[93], suggesting that the ubiquitin-proteasome pathway as well as the lysosomal enzyme is defective in PD[94]. Furthermore, proteins in Lewy bodies are generally oxidized, nitrated and contain products from lipid peroxidation that all promote aggregation[95]. Especially selective tyrosine nitration of α-synuclein may play a role in fibril formation of unfolded native synuclein and decrease the rate of degradation by the proteasome. Defective mitochondrial function like an impaired complex I activity is also revealed in Parkinson's brain tissue, and inhibitors of mitochondrial complex-1 lead to aggregation of α-synuclein *in vitro* as well as in animal models[96-97]. PD is typically associated with an increased iron content of the substantia nigra. Recent studies demonstrated raised iron levels in individual dopaminergic neurons of the substantia nigra. Moreover, accumulation of iron into mitochondria might lead to oxidative stress damaging iron-sulphur cluster-containing proteins[98]. Iron-mediated cellular destruction is mediated primarily *via* reactive oxygen or/and nitrogen species induced oxidative stress. Furthermore, these pathogenic mechanisms appear to be closely interlinked to the cascade of events leading to cellular death[99]. Dopamine coordinates metals such as Cu^{2+} and Fe^{3+}, reduces the oxidation state of the metals, and subsequently engenders production of H_2O_2, setting up conditions for Fenton chemistry[100]. Furthermore, studies have showed synthetic melanins were produced by incubating dopamine with Cu^{2+} and Fe^{3+}[101]. At low iron concentrations, melanins are known to have antioxidant properties, but at higher metal loads melanins are pro-oxidant. The oxidative stress associated with PD could be the result of a breakdown in the regulation of dopamine (neuromelanin)/iron biochemistry. Studies have showed that α-synuclein has a role in modulating the activity of dopamine. The mutation of α-synuclein associated with familial PD impairs vesicular storage of dopamine in the cytoplasm and subsequent generation of ROS through its interaction with iron[102]. In the presence of iron and under conditions of oxidative stress α-synuclein will aggregate and form deposits. In addition to the regulation of dopamine by α-synuclein, studies have shown a direct interaction of α-synuclein with metal ions, leading to protein aggregation.

Therapeutic options targeting oxidative stress in neurodegenerative diseases

Antioxidants

Antioxidants are exogenous or endogenous molecules which act against any forms of oxidative stress and its associated ill effects on the cellular system. They could neutralize ROS and other kinds of free radicals to inhibit oxidative stress. Many foods we consume contain a variety of antioxidant supplements, including flavonoids and phenolic compounds, lipoic acid (thioctic acid), ubiquinone and idebenone, β-carotene and vitamin C[103]. These natural antioxidants prevent oxidation of proteins, lipid peroxidations and prevent generation of ROS, thus act as an upstream therapeutic barrier to oxidative stress. One consequence of ROS generation is the initiation of excitotoxicity, which is modulated through the over-activation of glutamate receptors. Drugs that target these receptors are efficient upstream approaches in treating neurodegenerative diseases. For example, Memantine slows the development of AD and is of modest benefit to patients in the moderately severe to severe range of the disease by targets the N-methyl-D-aspartate receptor[104]. One of important futuristic upstream therapeutic aspect that can regulate oxidative stress to protect neuronal cells from death is vaccination against potential toxic protein formed in different types of neuronal disorders. A promising example is β-amyloid vaccination in AD that prevents plaque formation and subsequent neuron inflammation[105]. Downstream antioxidant activity functions as coverage for post oxidative stress events. For example, ginkgo biloba extracts (EGb 761), have been found to possess excellent antioxidant properties that restrict β-amyloid toxicity after plaque formation[106].

MPACs

MPACs are molecules that compete with the target protein for the metal ions. Since the breakdown in metal homeostasis in neuodegerative diseases leads to tissue saturation with metal, the intention of the MPAC is to disrupt an abnormal metal-protein interaction, to achieve a subtle repartitioning of metals and a subsequent normalization of metal distribution[107]. The prototypic MPAC is clioquinol. CQ is able to bind a range of metal ions with moderate affinity, that is, in the nM range. CQ was then given orally in a blinded study to Tg2576 transgenic mice, the results showed a 49% decrease in brain Aβ burden compared with non-treated controls after 9 weeks of treatment[108]. Moreover, the effect of oral CQ treatment in a randomized, double-blind, placebo-controlled pilot Phase II clinical trial[109] of moderately severe AD patients was evaluated. The results showed a statistically significant prevention of cognitive deterioration during a 36-week period in the more severely affected patients. There was also a significant decline in plasma $A\beta_{42}$ in the CQ group

compared with an increase in the placebo group. A novel second generation MPAC PBT2 has been synthesized that has higher solubility and increased blood-brain barrier permeability as compared with CQ. When tested in the APP/PS1 transgenic mouse model of AD[110], PBT2 decreased soluble interstitial Aβ within hours; this was accompanied by improved cognitive performance. In addition, there were significant decreases in insoluble Aβ load and tau phosphorylation. The randomized, double-blind, placebo-controlled Phase II clinical trial demonstrated reduced cerebrospinal fluid levels of $A\beta_{42}$ and improved cognitive performance in patients taking PBT2[111].

Concluding remarks and future strategies-targeting Nrf2 pathway

Accumulating data suggests that oxidative stress is involved in the pathogenesis of neurodegenerative diseases, and that antioxidant administration may be useful in the prevention and treatment of neurodegenerative diseases. To obtain efficacy in delaying diseases progression, the candidate antioxidant must be given as early as possible, before irreversible neuronal loss. It also should be tailored to the precise oxidative stress physiology, *e.g*. the type of ROS involved, the place of generation, and the severity of the damage. The chosen antioxidant should also be able to penetrate the blood-brain barrier after systemic administration in order to attain a critical therapeutic level within the CNS. Cellular protection against oxidative stress-induced toxicities is provided by two types of antioxidants: (1) direct antioxidants, which are redox active, short-lived, are sacrificed in the process of their antioxidant actions and need to be replenished or regenerated, and may evoke pro-oxidant effects; and (2) indirect antioxidants, that may or may not be redox active. Indirect antioxidants activate the Kelch like-ECH-associated protein 1 (Keap1)/Nrf2/antioxidant response element pathway resulting in transcriptional induction of a battery of cytoprotective proteins (also known as phase 2 enzymes) that act catalytically, are not consumed, have long half-lives, and are unlikely to evoke pro-oxidant effects[112]. Indirect antioxidants act through the augmentation of cellular antioxidant capacity by enhancing gene expression[113]. Nrf2, an important stress-responsive transcription factor of the “cap-and-collar” β-leucine zipper family, is widely activated in response to stimuli, such as oxidative and reactive species. The activation of Nrf2 leads to upregulation of an entire array of genes that impart protection, as a result, this pathway has been identified as a promising therapeutic target for neurodegenerative diseases where oxidative stress and neuro-inflammation occur[114]. Eventually, this effect influences the physiological, biochemical, and/or cellular processes that inactivate free radicals or that prevent free radical-initiated chemical reactions[115]. The role of Nrf2 is now considered instrumental to several neurodegenerative disorders[116]. Under normal or unstressed conditions, Nrf2 is tethered in the cytoplasm by another protein called Keap1. Keap1 acts as a substrate adaptor protein for Cullin 3-based ubiquitination, which results in degradation of Nrf2 and, under normal conditions; Nrf2 has a hal flife of only 20 minutes. Oxidative stress disrupts critical cysteine residues in Keap1, resulting in a disruption of the Keap1-Cullin 3 ubiquitination system and a build-up of Nrf2 in the cytoplasm. Unbound Nrf2 is then able to translocate into the nucleus, where it heterodimerizes with a small Maf protein and binds to the antioxidant response element in the upstream promoter region of many anti-oxidative genes, where it initiates their transcription. Activation of Nrf2 results in the induction of many cytoprotective proteins. These include heme oxygenase-1, nicotinamide adenine dinucleotide phosphate hydratenucleotide NAD(P)H quinone oxidoreductase 1, glutathione S-transferase and glutamatecysteine ligase[117-118]. In response to oxidative stress, Nrf2 normally translocates from the cytoplasm into the nucleus and transactivates expression of genes with antioxidant activity. Despite this cellular mechanism, severe oxidative damage is not uncommon in AD and PD. Intense mechanistic investigations in this arena have revealed that Nrf2 expression is abundant in both the nucleus and the cytoplasm of neurons in normal hippocampus with predominant expression in the nucleus. However, in AD, Nrf2 was predominantly cytoplasmic rather than nuclear in hippocampal neurons and was not a major component of beta amyloid plaques or neurofibrillary tangles. In contrast, the magnitude of expression of nuclear Nrf2 was much stronger in PD nigral neurons, but it was cytoplasm centric in substantia nigra from normal Alzheimer’s patients. Such observations suggest that Nrf2-mediated transcription is not robust in neurons in AD despite the presence of oxidative stress. But in PD, despite a stronger nuclear localization of Nrf2, the impact of Nrf2 may be inadequate to protect neurodegeneration. Because of this differential Nrf2 expression, it can be considered as a potential therapeutic target for conditions that are sensitive to free radical damage[89]. Pre-clinical and clinical studies of the therapeutic potential of phytochemicals that activate the Nrf2/antioxidant response element pathway in several different neurodegenerative disorders are in progress[119].

Author contributions: Xueping Chen is responsible for designing and writing the manuscript. Chunyan Guo helps to revise the manuscript. Jiming Kong is responsible for manuscript oversight and instruction.

Conflicts of interest: None declared.

Funding: This work was supported by the Muscular Dystrophy Association (MDA) USA; the National Natural Science Foundation of China, No. U0632007.

Acknowledgments: We gratefully acknowledge Dr. Huifang Shang, from Department of Neurology, West China Hospital, China, for providing the constructive suggestions for this review.

REFERENCES

[1] Aiken CT, Kaake RM, Wang X, et al. Oxidative stress-mediated regulation of proteasome complexes. Mol Cell Proteomics. in press.
[2] Ienco EC, LoGerfo A, Carlesi C, et al. Oxidative stress treatment for clinical trials in neurodegenerative diseases. J Alzheimers Dis. 2011;24 Suppl 2:111-126.
[3] Fulda S, Gorman AM, Hori O, et al. Cellular stress responses: cell survival and cell death. Int J Cell Biol. 2010;2010:214074.
[4] Martin LJ. Mitochondrial and cell death mechanisms in neurodegenerative diseases. Pharmaceuticals (Basel). 2010;3: 839-915.
[5] Genestra M. Oxyl radicals, redox-sensitive signalling cascades and antioxidants. Cell Signal. 2007;19:1807-1819.
[6] Halliwell B. Free radicals, antioxidants, and human disease: curiosity, cause, or consequence? Lancet. 1994;344:721-724.
[7] Halliwell B. Biochemistry of oxidative stress. Biochem Soc Trans. 2007;35:1147-1150.
[8] Droge W. Free radicals in the physiological control of cell function. Physiological reviews. 2002;82:47-95.
[9] Young IS, Woodside JV. Antioxidants in health and disease. J Clin Pathol. 2001;54:176-186.
[10] Valko M, Rhodes CJ, Moncol J, et al. Free radicals, metals and antioxidants in oxidative stress-induced cancer. Chemico-biological interactions. 2006;160:1-40.
[11] Valko M, Morris H, Cronin MT. Metals, toxicity and oxidative stress. Curr Med Chem. 2005;12:1161-1208.
[12] Mashima R, Witting PK, Stocker R. Oxidants and antioxidants in atherosclerosis. Curr Opin Lipidol. 2001;12:411-418.
[13] Balaban RS, Nemoto S, Finkel T. Mitochondria, oxidants, and aging. Cell. 2005;120:483-495.
[14] Beckman KB, Ames BN. The free radical theory of aging matures. Physiol Rev. 1998;78:547-581.
[15] Bush AI. Metals and neuroscience. Curr Opin Chem Biol. 2000;4: 184-191.
[16] Demuro A, Mina E, Kayed R, et al. Calcium dysregulation and membrane disruption as a ubiquitous neurotoxic mechanism of soluble amyloid oligomers. J Biol Chem. 2005;280:17294-17300.
[17] Echtay KS, Roussel D, St-Pierre J, et al. Superoxide activates mitochondrial uncoupling proteins. Nature. 2002;415:96-99.
[18] Sundaresan M, Yu ZX, Ferrans VJ, et al. Requirement for generation of H2O2 for platelet-derived growth factor signal transduction. Science. 1995;270:296-299.
[19] Pacher P, Beckman JS, Liaudet L. Nitric oxide and peroxynitrite in health and disease. Physiol Rev. 2007;87:315-424.
[20] Kong Q, Lin CL. Oxidative damage to RNA: mechanisms, consequences, and diseases. Cell Mol Life Sci. 2010;67: 1817-1829.
[21] Navarro A, Boveris A. Brain mitochondrial dysfunction in aging, neurodegeneration, and Parkinson's disease. Front Aging Neurosci. 2010;2. pii: 34.
[22] Patten DA, Germain M, Kelly MA, et al. Reactive oxygen species: stuck in the middle of neurodegeneration. J Alzheimers Dis. 2010; 20 Suppl 2: S357-367.
[23] Higgins GC, Beart PM, Shin YS, et al. Oxidative stress: emerging mitochondrial and cellular themes and variations in neuronal injury. J Alzheimers Dis. 2010;20 Suppl 2:S453-473.
[24] Lassmann H. Mechanisms of neurodegeneration shared between multiple sclerosis and Alzheimer's disease. J Neural Transm. 2011;118:747-752.
[25] Wang X, Michaelis EK. Selective neuronal vulnerability to oxidative stress in the brain. Front Aging Neurosci. 2010;2:12.
[26] Dauer W, Przedborski S. Parkinson's disease: mechanisms and models. Neuron. 2003;39:889-909.
[27] Rowland LP, Shneider NA. Amyotrophic lateral sclerosis. N Engl J Med. 2001;344:1688-1700.
[28] Dinkova-Kostova AT, Talalay P, Sharkey J, et al. An exceptionally potent inducer of cytoprotective enzymes: elucidation of the structural features that determine inducer potency and reactivity with Keap1. J Biol Chem. 2010;285:33747-33755.
[29] Finkel T, Holbrook NJ. Oxidants, oxidative stress and the biology of ageing. Nature. 2000;408:239-247.
[30] Adibhatla RM, Hatcher JF. Lipid oxidation and peroxidation in CNS health and disease: from molecular mechanisms to therapeutic opportunities. Antioxid Redox Signal. 2010;12: 125-169.
[31] Bochkov VN, Oskolkova OV, Birukov KG, et al. Generation and biological activities of oxidized phospholipids. Antioxid Redox Signal. 2010;12:1009-1059.
[32] Sedelnikova OA, Redon CE, Dickey JS, et al. Role of oxidatively induced DNA lesions in human pathogenesis. Mutat Res. 2010;704:152-159.
[33] Ferrari R, Guardigli G, Mele D, et al. Oxidative stress during myocardial ischaemia and heart failure. Curr Pharm Des. 2004; 10:1699-1711.
[34] Mark RJ, Lovell MA, Markesbery WR, et al. A role for 4-hydroxynonenal, an aldehydic product of lipid peroxidation, in disruption of ion homeostasis and neuronal death induced by amyloid beta-peptide. J Neurochem. 1997;68:255-264.
[35] Markesbery WR, Lovell MA. Four-hydroxynonenal, a product of lipid peroxidation, is increased in the brain in Alzheimer's disease. Neurobiol Aging. 1998;19:33-36.
[36] Arlt S, Beisiegel U, Kontush A. Lipid peroxidation in neurodegeneration: new insights into Alzheimer's disease. Curr Opin Lipidol. 2002;13:289-294.
[37] Selley ML, Close DR, Stern SE. The effect of increased concentrations of homocysteine on the concentration of (E)-4-hydroxy-2-nonenal in the plasma and cerebrospinal fluid of patients with Alzheimer's disease. Neurobiol Aging. 2002;23: 383-388.
[38] Keller JN, Pang Z, Geddes JW, et al. Impairment of glucose and glutamate transport and induction of mitochondrial oxidative stress and dysfunction in synaptosomes by amyloid beta-peptide: role of the lipid peroxidation product 4-hydroxynonenal. J Neurochem. 1997;69:273-284.
[39] Mattson MP, Chan SL. Neuronal and glial calcium signaling in Alzheimer's disease. Cell Calcium. 2003;34:385-397.
[40] Tamagno E, Robino G, Obbili A, et al. H_2O_2 and 4-hydroxynonenal mediate amyloid beta-induced neuronal apoptosis by activating JNKs and p38MAPK. Exp Neurol. 2003;180:144-155.
[41] Bruce-Keller AJ, Li YJ, Lovell MA, et al. 4-Hydroxynonenal, a product of lipid peroxidation, damages cholinergic neurons and impairs visuospatial memory in rats. J Neuropathol Exp Neurol. 1998;57:257-267.
[42] Davies MJ. The oxidative environment and protein damage. Biochim Biophys Acta. 2005;1703:93-109.
[43] Berlett BS, Stadtman ER. Protein oxidation in aging, disease, and oxidative stress. J Biol Chem. 1997;272:20313-20316.
[44] Dalle-Donne I, Aldini G, Carini M, et al. Protein carbonylation, cellular dysfunction, and disease progression. J Cell Mol Med. 2006;10:389-406.
[45] Levine RL, Williams JA, Stadtman ER, et al. Carbonyl assays for determination of oxidatively modified proteins. Methods Enzymol. 1994;233:346-357.
[46] Smith MA, Sayre LM, Anderson VE, et al. Cytochemical demonstration of oxidative damage in Alzheimer disease by immunochemical enhancement of the carbonyl reaction with 2, 4-dinitrophenylhydrazine. J Histochem Cytochem. 1998;46: 731-735.
[47] Korolainen MA, Nyman TA, Nyyssonen P, et al. Multiplexed proteomic analysis of oxidation and concentrations of cerebrospinal fluid proteins in Alzheimer disease. Clin Chem. 2007;53:657-665.
[48] Lovell MA, Markesbery WR. Oxidative DNA damage in mild cognitive impairment and late-stage Alzheimer's disease. Nucleic Acids Res. 2007;35:7497-7504.

[49] Gabbita SP, Lovell MA, Markesbery WR. Increased nuclear DNA oxidation in the brain in Alzheimer's disease. J Neurochem. 1998; 71:2034-2040.
[50] Collins AR, Dusinska M, Gedik CM, et al. Oxidative damage to DNA: do we have a reliable biomarker? Environ Health Perspect. 1996;104 Suppl 3:465-469.
[51] Nunomura A, Perry G, Pappolla MA, et al. RNA oxidation is a prominent feature of vulnerable neurons in Alzheimer's disease. J Neurosci. 1999;19:1959-1964.
[52] Nunomura A, Perry G, Aliev G, et al. Oxidative damage is the earliest event in Alzheimer disease. J Neuropathol Exp Neurol. 2001;60:759-767.
[53] Lovell MA, Gabbita SP, Markesbery WR. Increased DNA oxidation and decreased levels of repair products in Alzheimer's disease ventricular CSF. J Neurochem. 1999;72:771-776.
[54] Smith MA, Perry G, Richey PL, et al. Oxidative damage in Alzheimer's. Nature. 1996;382:120-121.
[55] Munch G, Cunningham AM, Riederer P, et al. Advanced glycation endproducts are associated with Hirano bodies in Alzheimer's disease. Brain Res. 1998;796:307-310.
[56] Tyedmers J, Mogk A, Bukau B. Cellular strategies for controlling protein aggregation. Nat Rev Mol Cell Biol. 2010;11:777-788.
[57] Klein WL, Stine WB Jr, Teplow DB. Small assemblies of unmodified amyloid beta-protein are the proximate neurotoxin in Alzheimer's disease. Neurobiol Aging. 2004;25:569-580.
[58] Roy S, Zhang B, Lee VM, et al. Axonal transport defects: a common theme in neurodegenerative diseases. Acta Neuropathol. 2005;109:5-13.
[59] Williams A. Defining neurodegenerative diseases. Bmj. 2002; 324: 1465-1466.
[60] Lee BH, Lee MJ, Park S, et al. Enhancement of proteasome activity by a small-molecule inhibitor of USP14. Nature. 2010;467: 179-184.
[61] Jung T, Catalgol B, Grune T. The proteasomal system. Mol Aspects Med. 2009;30:191-296.
[62] Seifert U, Bialy LP, Ebstein F, et al. Immunoproteasomes preserve protein homeostasis upon interferon-induced oxidative stress. Cell. 2010;142:613-624.
[63] Ciechanover A, Brundin P. The ubiquitin proteasome system in neurodegenerative diseases: sometimes the chicken, sometimes the egg. Neuron. 2003;40:427-446.
[64] Dahlmann B. Role of proteasomes in disease. BMC Biochem. 2007;8 Suppl 1:S3.
[65] Bertini I, Cavallaro G. Metals in the "omics" world: copper homeostasis and cytochrome c oxidase assembly in a new light. J Biol Inorg Chem. 2008;13:3-14.
[66] Mills E, Dong XP, Wang F, et al. Mechanisms of brain iron transport: insight into neurodegeneration and CNS disorders. Future Med Chem. 2010;2:51-64.
[67] Barnham KJ, Bush AI. Metals in Alzheimer's and Parkinson's diseases. Curr Opin Chem Biol. 2008;12:222-228.
[68] Halliwell B. The wanderings of a free radical. Free Radic Biol Med. 2009;46:531-542.
[69] Halliwell B. Role of free radicals in the neurodegenerative diseases: therapeutic implications for antioxidant treatment. Drugs Aging. 2001;18:685-716.
[70] Ke Y, Qian ZM. Brain iron metabolism: neurobiology and neurochemistry. Prog Neurobiol. 2007;83:149-173.
[71] Zecca L, Youdim MB, Riederer P, et al. Iron, brain ageing and neurodegenerative disorders. Nat Rev Neurosci. 2004;5:863-873.
[72] Salvador GA, Uranga RM, Giusto NM. Iron and mechanisms of neurotoxicity. Int J Alzheimers Dis. 2010;2011:720658.
[73] Bertram L, Tanzi RE. Thirty years of Alzheimer's disease genetics: the implications of systematic meta-analyses. Nat Rev Neurosci. 2008;9:768-778.
[74] Christen Y. Oxidative stress and Alzheimer disease. Am J Clin Nutr. 2000;71:621S-629S.
[75] Querfurth HW, LaFerla FM. Alzheimer's disease. N Engl J Med. 2010;362:329-344.
[76] Pratico D. Oxidative stress hypothesis in Alzheimer's disease: a reappraisal. Trends Pharmacol Sci. 2008;29:609-615.
[77] Montine KS, Reich E, Neely MD, et al. Distribution of reducible 4-hydroxynonenal adduct immunoreactivity in Alzheimer disease is associated with APOE genotype. J Neuropathol Exp Neurol. 1998;57:415-425.
[78] Wang J, Xiong S, Xie C, et al. Increased oxidative damage in nuclear and mitochondrial DNA in Alzheimer's disease. J Neurochem. 2005;93:953-962.
[79] Ahmed N, Ahmed U, Thornalley PJ, et al. Protein glycation, oxidatio n and nitration adduct residues and free adducts of cerebrospinal fluid in Alzheimer's disease and link to cognitive impairment. J Neurochem. 2005;92:255-263.
[80] Choi J, Rees HD, Weintraub ST, et al. Oxidative modifications and aggregation of Cu,Zn-superoxide dismutase associated with Alzheimer and Parkinson diseases. J Biol Chem. 2005;280: 11648-11655.
[81] Wong A, Luth HJ, Deuther-Conrad W, et al. Advanced glycation endproducts co-localize with inducible nitric oxide synthase in Alzheimer's disease. Brain Res. 2001;920:32-40.
[82] Poppek D, Keck S, Ermak G, et al. Phosphorylation inhibits turnover of the tau protein by the proteasome: influence of RCAN1 and oxidative stress. Biochem J. 2006;400:511-520.
[83] Keck S, Nitsch R, Grune T, et al. Proteasome inhibition by paired helical filament-tau in brains of patients with Alzheimer's disease. J Neurochem. 2003;85:115-522.
[84] Bonda DJ, Lee HG, Blair JA, et al. Role of metal dyshomeostasis in Alzheimer's disease. Metallomics. 2011;3:267-270.
[85] Zhang L, Zhao B, Yew DT, et al. Processing of Alzheimer's amyloid precursor protein during H2O2-induced apoptosis in human neuronal cells. Biochem Biophys Res Commun. 1997;235:845-848.
[86] Atwood CS, Moir RD, Huang X, et al. Dramatic aggregation of Alzheimer abeta by Cu(II) is induced by conditions representing physiological acidosis. J Biol Chem. 1998;273:12817-12826.
[87] Atwood CS, Scarpa RC, Huang X, et al. Characterization of copper interactions with alzheimer amyloid beta peptides: identification of an attomolar-affinity copper binding site on amyloid beta1-42. J Neurochem. 2000;75:1219-1233.
[88] Atwood CS, Obrenovich ME, Liu T, et al. Amyloid-beta: a chameleon walking in two worlds: a review of the trophic and toxic properties of amyloid-beta. Brain Res Brain Res Rev. 2003;43: 1-16.
[89] Cherny RA, Barnham KJ, Lynch T, et al. Chelation and intercalation: complementary properties in a compound for the treatment of Alzheimer's disease. J Struct Biol. 2000;130: 209-216.
[90] Thomas B, Beal MF. Parkinson's disease. Hum Mol Genet. 2007;16 Spec No. 2:R183-194.
[91] Baloyannis SJ, Costa V, Baloyannis IS. Morphological alterations of the synapses in the locus coeruleus in Parkinson's disease. J Neurol Sci. 2006;248:35-41.
[92] Wakabayashi K, Tanji K, Mori F, et al. The Lewy body in Parkinson's disease: molecules implicated in the formation and degradation of alpha-synuclein aggregates. Neuropathology. 2007;27:494-506.
[93] Chu Y, Dodiya H, Aebischer P, et al. Alterations in lysosomal and proteasomal markers in Parkinson's disease: relationship to alpha-synuclein inclusions. Neurobiol Dis. 2009;35:385-398.
[94] Surendran S, Rajasankar S. Parkinson's disease: oxidative stress and therapeutic approaches. Neurol Sci. 2010;31:531-540.
[95] Hodara R, Norris EH, Giasson BI, et al. Functional consequences of alpha-synuclein tyrosine nitration: diminished binding to lipid vesicles and increased fibril formation. J Biol Chem. 2004;279: 47746-47753.
[96] Betarbet R, Sherer TB, MacKenzie G, et al. Chronic systemic pesticide exposure reproduces features of Parkinson's disease. Nat Neurosci. 2000;3:1301-1306.

[97] Manning-Bog AB, McCormack AL, Li J, et al. The herbicide paraquat causes up-regulation and aggregation of alpha-synuclein in mice: paraquat and alpha-synuclein. J Biol Chem. 2002;277:1641-1644.
[98] Gille G, Reichmann H. Iron-dependent functions of mitochondria-relation to neurodegeneration. J Neural Transm. 2011;118: 349-359.
[99] Sian-Hulsmann J, Mandel S, Youdim MB, et al. The relevance of iron in the pathogenesis of Parkinson's disease. J Neurochem. 2011;118:939-957.
[100] Asano K, Chee CB, Gaston B, et al. Constitutive and inducible nitric oxide synthase gene expression, regulation, and activity in human lung epithelial cells. Proc Natl Acad Sci U S A. 1994;91: 10089-10093.
[101] Wakamatsu K, Fujikawa K, Zucca FA, et al. The structure of neuromelanin as studied by chemical degradative methods. J Neurochem. 2003;86:1015-1023.
[102] Lotharius J, Brundin P. Impaired dopamine storage resulting from alpha-synuclein mutations may contribute to the pathogenesis of Parkinson's disease. Hum Mol Genet. 2002;11:2395-2407.
[103] Lepoivre M, Flaman JM, Bobe P, et al. Quenching of the tyrosyl free radical of ribonucleotide reductase by nitric oxide. Relationship to cytostasis induced in tumor cells by cytotoxic macrophages. J Biol Chem. 1994;269:21891-21897.
[104] Reisberg B, Doody R, Stoffler A, et al. Memantine in moderate-to-severe Alzheimer's disease. N Engl J Med. 2003;348:1333-1341.
[105] Janus C, Pearson J, McLaurin J, et al. A beta peptide immunization reduces behavioural impairment and plaques in a model of Alzheimer's disease. Nature. 2000;408:979-982.
[106] Yao Z, Drieu K, Papadopoulos V. The Ginkgo biloba extract EGb 761 rescues the PC12 neuronal cells from beta-amyloid-induced cell death by inhibiting the formation of beta-amyloid-derived diffusible neurotoxic ligands. Brain Res. 2001;889:181-190.
[107] Kenche VB, Barnham KJ. Alzheimer's disease & metals: therapeutic opportunities. Br J Pharmacol. 2011;163:211-219.
[108] Cherny RA, Atwood CS, Xilinas ME, et al. Treatment with a copper-zinc chelator markedly and rapidly inhibits beta-amyloid accumulation in Alzheimer's disease transgenic mice. Neuron. 2001;30:665-676.
[109] Ritchie CW, Bush AI, Mackinnon A, et al. Metal-protein attenuation with iodochlorhydroxyquin (clioquinol) targeting Abeta amyloid deposition and toxicity in Alzheimer disease: a pilot phase 2 clinical trial. Arch Neurol. 2003;60:1685-1691.
[110] Adlard PA, Cherny RA, Finkelstein DI, et al. Rapid restoration of cognition in Alzheimer's transgenic mice with 8-hydroxy quinoline analogs is associated with decreased interstitial Abeta. Neuron. 2008;59:43-55.
[111] Faux NG, Ritchie CW, Gunn A, et al. PBT2 rapidly improves cognition in Alzheimer's Disease: additional phase II analyses. J Alzheimers Dis. 2010;20:509-516.
[112] Dinkova-Kostova AT, Talalay P. Direct and indirect antioxidant properties of inducers of cytoprotective proteins. Mol Nutr Food Res. 2008;52 Suppl 1:S128-138.
[113] Jung KA, Kwak MK. The Nrf2 system as a potential target for the development of indirect antioxidants. Molecules. 2010;15: 7266-7291.
[114] Epifano F, Curini M, Menghini L, et al. Natural coumarins as a novel class of neuroprotective agents. Mini Rev Med Chem. 2009; 9:1262-1271.
[115] Singh S, Vrishni S, Singh BK, et al. Nrf2-ARE stress response mechanism: a control point in oxidative stress-mediated dysfunctions and chronic inflammatory diseases. Free Radic Res. 2010;44:1267-1288.
[116] Ramsey CP, Glass CA, Montgomery MB, et al. Expression of Nrf2 in neurodegenerative diseases. J Neuropathol Exp Neurol. 2007; 66:75-85.
[117] Calkins MJ, Johnson DA, Townsend JA, et al. The Nrf2/ARE pathway as a potential therapeutic target in neurodegenerative disease. Antioxid Redox Signal. 2009;11:497-508.
[118] Vargas MR, Johnson DA, Sirkis DW, et al. Nrf2 activation in astrocytes protects against neurodegeneration in mouse models of familial amyotrophic lateral sclerosis. J Neurosci. 2008;28: 13574-13581.
[119] Ghosh N, Ghosh R, Mandal SC. Antioxidant protection: A promising therapeutic intervention in neurodegenerative disease. Free Radic Res. 2011;45:888-905.

(Edited by Bai H, Gong QH/Yang Y/Song LP)