

Therapeutic potential of baicalein in Alzheimer's and Parkinson's disease

Yanwei Li¹, Jinying Zhao¹, Christian Hölscher²

1. Department of Human Anatomy, Medical College, Shaoyang University, Shaoyang, Hunan, PR China

2. Biomedical and Life Science, Faculty of Health and Medicine, Lancaster University, Lancaster LA1 4YQ, UK

Running title: *baicalein in central neurodegenerative diseases*

Corresponding author:

Yanwei Li, Department of Human Anatomy, Medical College, Shaoyang University, Shaoyang, Hunan 422000, PR China.

E-mail: liyanwei915@163.com

Abstract

Alzheimer's disease (AD) and Parkinson's disease (PD) are the two most common progressive central neurodegenerative diseases affecting the population over the age of 60 years. Apart from treatments that temporarily improve symptoms, there is no medicine currently available which can inhibit or reverse the progression of AD and PD. In Traditional Chinese Medicine (TCM), the root of *Scutellaria baicalensis Georgi* is a classic compatible component in decoction of herbal medicine used for treating central nervous system (CNS) diseases. Modern pharmacokinetic studies have confirmed that baicalein (5, 6, 7-trihydroxyflavone) is a major bioactive flavone constituent root of *Scutellaria baicalensis Georgi*. Studies showed that baicalein possesses a range of key pharmacological properties, such as reducing oxidative stress, anti-inflammatory properties, inhibiting aggregation of disease-specific amyloid proteins, inhibiting excitotoxicity, stimulating neurogenesis and differentiation action, and anti-apoptosis effect. Based on these, baicalein shows therapeutic potential for AD and PD. In the present review, we will summarize the pharmacological protective actions of baicalein that make it suitable for the treatment of AD and PD, and the potential mechanisms underlying the effects will also be discussed.

Key points:

1. No cures are available for inhibiting or reversing the progression of Alzheimer's disease (AD) and Parkinson's disease (PD).
2. Active ingredients of traditional Chinese medicine (TCM) show good therapeutic effects on age-related central nervous system (CNS) diseases.
3. Baicalein, a major bioactive flavone constituent of *Scutellaria baicalensis Georgi*, has been in the focus of research as a versatile therapeutic agent for AD and PD.

Introduction

Alzheimer's disease (AD) and Parkinson's disease (PD) are the most common two progressive central neurodegenerative diseases affecting the population over the age of 60 years. Pathologically, AD is characterized by extracellular β -amyloid ($A\beta$) plaques and intracellular neurofibrillary tangles in the brain. AD manifests itself in progressive memory loss and cognitive decline, accompanied with changes in emotion and even psychiatric symptoms (Cavallucci et al., 2012). The pathology of PD is characterized by a progressive loss of dopaminergic neurons and nerve fibers in the nigrostriatal system which leads to tremors, muscular rigidity, bradykinesia, and postural and gait abnormalities (Kowal et al., 2013). With the increasing aging population, the incidence of AD and PD is on the rise and is becoming a large economic burden to society and to patient families (Reitz et al., 2011). However, no cures are available for these central neurodegenerative diseases.

Currently, the main approach to the treatment of AD is trying to augment cholinergic neurotransmission in the brain. This involves the use of cholinesterase inhibitors such as tacrine, galantamine, rivastigmine, and donepezil (Kulshreshtha and Piplani, 2016). In PD, administering the dopamine precursor L-dopa remains the standard treatment for the symptoms of PD in the last 30 years (Olanow and Schapira, 2013). However, these medicines can only improve the symptoms temporarily; they cannot inhibit or reverse the progression of AD and PD. The long-term use of them produces various adverse effects and complications, and eventually fails to improve symptoms of patients any longer as the disease progresses (Kumar et al., 2015; Sharma et al., 2015). Therefore, there is a great need for the development of new anti-neurodegeneration drugs that can stop disease progression.

In oriental countries, a variety of traditional Chinese medicine (TCM) preparations contain different kinds of herbal medicines which have proven to have good therapeutic effects on age-related central nervous system (CNS) diseases, including dementia, cerebral stroke and paralysis, in clinical practice for centuries (Li et al., 2013b; More et al., 2013). However, the molecular mechanisms involved in TCM remain unclear; they are therefore not readily accepted by practitioners of modern Western medicine. The systematic identification of these mechanisms with modern pharmacological and biochemical techniques can be challenging (Wang et al., 2016b). Interestingly, the root of *Scutellaria baicalensis Georgi* is a classic compatible component in decoction of herbal medicine used for treating CNS diseases, and there are reports that elevated consumption of dietary supplements containing the herbal medicine is associated with a lower incidence of dementia and other age-related neurological disorders (Gasiorowski et al., 2011). Nevertheless, more research is required to uncover its molecular actions.

Scutellaria baicalensis Georgi (Chinese herbal name: Huang qin) is extensively cultivated in China, Siberia, Mongolia, the Russian Far East, Korea and Japan. As a Chinese traditional medicine, Huang qin usually refers to the dried root of *Scutellaria baicalensis Georgi*. Modern pharmacokinetic studies have confirmed that baicalein (5, 6, 7-trihydroxyflavone) is a major bioactive flavone constituent of *Scutellaria baicalensis Georgi* (Liu et al., 2017). Baicalein has been well known for its broad pharmacological activities, including antibacterial, antiviral,

anticarcinogenic and anti-inflammatory activities in TCM (Dinda et al., 2017; Huang et al., 2005; Ji et al., 2015; Li et al., 2011). Baicalein has a wide safety margin and appears to be able to permeate the blood-brain barrier (BBB)(Chen et al., 2016; Liu et al., 2010; Tsai et al., 2002). Several *in vitro* studies have shown that baicalein is effective in preventing neurotoxicity induced by a variety of neurotoxins and stress inducers, including A β (Lin et al., 2017; Wang et al., 2004), 6-hydroxydopamine (6-OHDA) (Lee et al., 2005), rotenone (Li et al., 2012), H₂O₂(Zhang et al., 2010), and glutamate (Lee et al., 2003), and others. In recent studies, moreover, baicalein has been shown to possess neuroprotective properties in several animal models of AD and PD (Cheng et al., 2008; Gao et al., 2015; Gu et al., 2016; Zhou et al., 2016). Thus, baicalein has been in the focus of research as a versatile therapeutic agent for central neurodegenerative diseases (Fig. 1)(Gasiorowski et al., 2011). In the following paragraphs, we review the pharmacological actions of baicalein in the treatment of AD and PD, and highlight the potential underlying biochemical mechanisms of action.

Antioxidant properties of baicalein

Reactive oxygen species (ROS) are particularly active in the brain as the excitatory amino acids neurotransmitters (glutamate) and catecholamine neurotransmitters (dopamine), whose metabolism is a source of free radicals, can enhance oxidative stress (Miyazaki and Asanuma, 2009). Neural cells are considered to be more susceptible to oxidative damage as compared to other body tissues, because of limited antioxidant systems in place, higher level of iron in certain regions, and high level of unsaturated fatty acids which are more susceptible to peroxidation (Uttara et al., 2009).

A large body of evidence has suggested that higher level of markers of oxidative stress including protein nitrotyrosine, carbonyls in proteins, lipid oxidation products and oxidized DNA bases exist in the brains of AD (Aslan and Ozben, 2004; Korolainen et al., 2006) and PD patients (Sanders and Greenamyre, 2013). It is well known that oxidative stress is involved in the pathogenesis of neurodegenerative disorders (Khan et al., 2016). In the pathological process, ROS attack neurons and glial cells, contributing to DNA injury, proteins misfolding and aggregation, inflammation, tissue damage and subsequent cellular apoptosis, which are directly or indirectly linked to neuronal cell death. Contrary, antioxidants, including vitamin C, vitamin E, glutathione, coenzyme Q (CoQ), carotenoids, and melatonin, have been suggested to prevent or reduce the rate of progression of these diseases (Li et al., 2013a). A combination of neurotransmitter supplements and antioxidants has been recommended in the treatment of AD or PD. Therefore, development of antioxidants as neuroprotective drugs is a potentially beneficial strategy for clinical therapy.

Recently, baicalein has been shown to have beneficial protective effect on the oxidative stress-related injury in several *in vitro* studies. In PC12 cells, baicalein inhibited H₂O₂-induced cell viability loss, intracellular ROS generation, and lipid peroxidation in a dose-dependent manner (Zhang et al., 2010). In rotenone-damaged PC12 cells, similarly, baicalein also inhibited the accumulation of ROS, ATP deficiency, and mitochondrial membrane potential dissipation (Li et al., 2012). Baicalein potently reduced A β -induced neurotoxicity in PC12 cells, possibly by a reduction of oxidative stress (Heo et al., 2004). Moreover, baicalein protected rat cortical neurons from

A β -induced toxicity by its inhibition of oxidative stress. In addition, baicalein effectively protected SH-SY5Y cells, a dopaminergic neuroblastoma cell line, from 6-OHDA-induced damage by the attenuation of ROS and mitochondrial dysfunction (Lee et al., 2005). Baicalein can protect mitochondrial membranes against oxidative damage induced by the four different agents (gamma-radiation, peroxy radicals, ascorbate-Fe²⁺ and peroxynitrite) (Adhikari et al., 2011). Therefore, these findings suggested that baicalein exerts an anti-oxidant effect, which may be beneficial for treatment of oxidative stress-related neurodegenerative diseases.

Mechanism of antioxidant stress of baicalein

Scavenging free radicals

During oxidative stress, free radicals are directly or indirectly linked to the induced neuronal cell death. Free radical scavengers also are beneficial for reducing or preventing progression of neurodegenerative diseases (Li et al., 2013a). It is generally accepted that baicalein exerts an anti-oxidant effect mainly mediated by scavenging primary and secondary free radicals, including superoxide, H₂O₂, and hydroxyl radicals, during oxidative stress (Hamada et al., 1993). Baicalein belongs to the groups of flavonoids. In general, flavonoids contain multiple hydroxyl substitutions that exhibit radical scavenging activity. Baicalein has the 5,6,7-trihydroxy structure on ring-A. The reported antioxidant properties of baicalein are attributed to these structural features. In contrast, baicalin, a glycosylation product of baicalein, has less anti-oxidative activity compared with baicalein (Gao et al., 1999; Wozniak et al., 2015). In addition, it has been shown that the suppressive effect of baicalein on hydrogen peroxide-induced injury in SH-SY5Y cells was greater than that of baicalin (Gao et al., 2001). These findings suggest that antioxidant properties of baicalein is primarily dependent on the 5,6,7-trihydroxy structure on ring-A.

Recovering or upregulating antioxidant enzymes

In general, there is significant endogenous antioxidant depletion in the brain of PD and AD, which may aggravate oxidative stress (Mazzetti et al., 2015). Activation of endogenous antioxidants, including catalase, superoxide dismutase, and glutathione reductase enzymes, which reduce the cellular concentration of free radicals, is a common therapeutic strategy against neurodegeneration (Chan and Chan, 2015).

Interestingly, aside from scavenging free radicals, baicalein is able to increase antioxidant capabilities by recovering activities of endogenous antioxidant enzymes and upregulating their gene expression. Some studies have indicated that formation of dopamine (DA) quinone, a DAergic neuron-specific oxidative stressor, plays an important role in the pathogenesis of PD (Asanuma et al., 2004; Asanuma et al., 2003; Miyazaki and Asanuma, 2009). However, baicalein can prevent the formation of DA semiquinone radicals from DA as shown in an in vitro cell-free system. Furthermore, baicalein has neuroprotective properties against excess L-DOPA-induced DA neurotoxicity through the suppression of DA quinone formation. Furthermore, the long-term treatment of baicalein upregulates the intracellular glutathione (GSH) contents (Takeshima et al., 2011). One of the mechanisms that neuronal cells have adapted to protect themselves against oxidative stress and other insults is the nuclear factor-erythroid 2-related factor 2 (Nrf2) pathway and the binding of this master transcriptional regulator with antioxidant response element (ARE) in the regulatory region of many genes, which leads to the expression of several enzymes with

antioxidant and detoxification capacities. Notably, manganese superoxide dismutase (MnSOD) is an important antioxidant enzyme in mitochondria against oxidative stress. It was found that baicalein restored both MnSOD protein expression and its activity, and furthermore restored nuclear Nrf2 protein expression and its ARE binding activity (Lee et al., 2011). In addition, baicalein protects C6 glial cells against hydrogen peroxide-induced oxidative stress and apoptosis through regulation of the Nrf2 signaling pathway (Choi et al., 2016). Hence, these studies demonstrate that baicalein attenuates mitochondrial oxidative stress by activating Nrf2-mediated MnSOD induction. It can be predicted that the Nrf2 signaling pathway can also be upregulated by baicalein in neurons, but further research is needed to confirm this.

Inhibiting lipoxygenase

It is generally believed that oxidative stress-related genes such as the lipoxygenase (LOX) by oxidizing polyunsaturated fatty acids synthesize hydroperoxyacids, which are potent pro-oxidant mediators. Especially 12/15-lipoxygenase (12/15-LOX, encoded by the ALOX15 gene), the dominant isoform in the brain, is known for its ability to damage mitochondria (Joshi et al., 2015). It was found that 12/15-LOX contributes to brain damage after middle cerebral artery occlusion (van Leyen et al., 2006). Importantly, the amount of 12/15-LOX is increased in affected frontal and temporal regions of AD brains (Pratico et al., 2004), and cerebrospinal fluid levels of both of its metabolic products, 12-HETE and 15-HETE, are elevated in individuals with a clinical diagnosis of AD (Yao et al., 2005). Moreover, overexpression of 12/15-LOX leads to increased levels of A β and deposition, phosphorylated tau at Ser 202/Thr 205 and Ser 396 and a worsening of memory deficits in AD transgenic mice (Chu et al., 2012; Giannopoulos et al., 2013). On the contrary, pharmacologic block and genetic deletion of 12/15-LOX in various AD models improves synaptic integrity, and ameliorates A β and tau neuropathology, and cognitive impairments (Chu et al., 2015; Yang et al., 2010). Therefore, the 12/15-LOX is showing promise as an emerging therapeutic target for AD (Joshi et al., 2015).

Besides 12/15-LOX, 5-LOX, another lipoxygenase that is expressed in the CNS, may be involved in the process of oxidative stress of PD. 5-LOX was found to be over-expressed in the nigrostriatal system after the injection of MPTP into C57BL6 mice. 5-LOX is also involved in rotenone-induced damage in PC-12 cells (Zhang et al., 2011). It has also been shown that the over-activation of the 5-LOX pathway may lead to neurodegeneration by causing lipid peroxidation. In contrast, inhibitors of 5-LOX showed neuroprotective effects on (MPTP)/MPP(+)-induced dopaminergic neuronal death in midbrain neuron-glia co-cultures and *in vivo* in mice (Kang et al., 2013). Similarly, the 5-LOX inhibitor phenidone attenuated LPS-induced oxidative stress and dopaminergic neurodegeneration (Li et al., 2008). In addition, the 5-LOX inhibitor zileuton can reduce rotenone-induced cell injury (Zhang et al., 2011).

Recent studies showed that the anti-oxidant effect of baicalein may be mediated by inhibiting these oxidative stress-related genes. Baicalein has been regarded as the 12/15-LOX inhibitor and has the potential to inhibit the 12/15-LOX pathway (Deschamps et al., 2006; Sadik et al., 2003; Sekiya and Okuda, 1982). Furthermore, long-term oral administration of baicalein inhibits 12/15-LOX enzymatic activity in APP/PS1 mice (Gu et al., 2016). It has been hypothesized that the mechanism of baicalein inhibition of 12/15-LOX is by reducing target molecules such as iron,

which molecular modeling suggests is through direct binding of the catecholic moiety of baicalein to the iron. Though there is no information on whether baicalein can inhibit 5-LOX enzymatic activity, baicalein reduces oxidative stress in rotenone or 6-OHDA -induced PD cellular models (Lee et al., 2005; Li et al., 2012). According to bioactivity and structural characteristics of baicalein, it is speculated that baicalein has an inhibitory effect on 5-LOX. However, this requires further research to confirm this.

Chelating excessive iron

Brain iron accumulation in the specific brain regions affected by the disease is a common pathological feature of neurodegenerative diseases including AD and PD (Raven et al., 2013). Mutations or dysregulation of several proteins related to iron transport leads to abnormal brain iron accumulation and have been linked to pathogenesis of AD and PD (Schneider, 2016). The elevated iron levels observed in dopaminergic neurons of the substantia nigra of PD subjects have been suggested to incite intracellular α -synuclein aggregation (Song et al., 2007; Weinreb et al., 2013). Multiple forms of iron accumulation in the brain also demonstrated tau or A β pathology, suggesting parallels with AD (Kruer, 2013). The most likely mechanism by which A β or α -synuclein (α -Syn) aggregation may increase oxidative stress *in vitro* refers to its ability to bind iron. Like most transition metals, an excess of intracellular iron is toxic. Through the Fenton reaction, iron can react with endogenous hydrogen peroxide to produce the short-lived and highly reactive hydroxyl radical (OH \cdot), and further lipid peroxidation and oxidative stress related cell death. Recently, the iron-dependent cell death pathway was defined as ferroptosis, which was characterized in neurotoxin-induced animal and cellular models of AD or PD (Guiney et al., 2017; Hambright et al., 2017). Therefore, ferroptosis may also be an important neurodegenerative mechanism in diseases such as AD and PD. In neurodegenerative diseases, iron chelators may be strong drug candidates to pharmacologically modulate the ferroptotic signaling cascade (Do Van et al., 2016; Dusek et al., 2016).

Experiments have shown that baicalein can chelate iron and inhibit iron-induced lipid peroxidation and oxidative stress damage. As early as the 1980s, baicalein was observed to inhibit iron-induced lipid peroxidation in rat liver (Kimuya et al., 1981). In addition, baicalein has also been shown to strongly inhibit iron-dependent lipid peroxidation in microsomes and mitochondria (Gao et al., 1995; Gao et al., 1998). Baicalein can protect neurons from FeCl $_3$ -induced epilepsy (Hamada et al., 1993) and can control iron overload and reduce iron overload-induced liver damage in mouse models (Zhang et al., 2006; Zhao et al., 2005). Moreover, baicalein prevented ROS-mediated damage of human dermal fibroblast cells better than the iron chelator deferoxamine (Gao et al., 1998). The most effective iron binding site of flavonoids is the 6,7-dihydroxy structure. This site is incorporated in the baicalein molecule and forms, similarly to deferoxamine, complexes with iron in the stoichiometry 1:1 and was superior to deferoxamine (Mladenka et al., 2011). Baicalein appears to bind the ferrous ion more strongly than ferrozine, a well-known iron(II) chelator. Using ^1H nuclear magnetic resonance (NMR) and Zn $^{2+}$ and Ga $^{3+}$ as probes, the iron-binding site on baicalein was elucidated to be at the O6/O7 oxygen atoms of the A-ring. No binding was observed for baicalin under the same NMR conditions. Furthermore, baicalein strongly inhibits the Fe-promoted Fenton chemistry via a combination of chelation and radical scavenging mechanisms while baicalin can provide only partial protection against radical

damage. These results indicate that baicalein is a strong iron chelator under physiological conditions (Perez et al., 2009). In addition, in a natural product library screening for ferroptosis inhibitors, it was found that baicalein is a potent inhibitor of erastin-induced ferroptosis in pancreatic cancer cells (Xie et al., 2016). Baicalein exhibits remarkable anti-ferroptosis activity compared with well-known ferroptosis inhibitors such as ferrostatin-1, liproxstatin-1, deferoxamine mesylate, and β -mercaptoethanol (Xie et al., 2016). Thus, based on these findings, baicalein may have the potential to treat neurodegenerative diseases by chelating excessive iron.

In conclusion, these above findings suggest that baicalein has antioxidant stress effects by scavenging free radicals, upregulating antioxidant enzymes, inhibiting lipoxygenase, and chelating excessive iron. Hence, baicalein may be an effective antioxidant stress agent for neurodegenerative diseases.

Baicalein inhibits aggregation of disease-specific amyloid proteins

Neurodegenerative diseases share a similarity in aggregation of disease-specific amyloid proteins in the brain. Amyloids are β -sheet rich fibrillar aggregates formed by self-assembly of different proteins including A β and α -Syn. Accumulation of abnormally folded A β plaque occurs in the brain of AD patients, and abnormally folded α -Syn is the major component of Lewy bodies, which are intracellular inclusions present in the dopaminergic neurons of PD patients' brains (Spillantini et al., 1997). Mutations of β -amyloid precursor protein (APP) or α -Syn are linked with familial early-onset AD or PD (Goedert, 2015). Thus, these disease-specific amyloid proteins (A β and α -Syn) are considered to be the key molecule to be targeted for AD or PD therapy. It has been suggested that the precursors of fibrils, oligomeric α -Syn or A β , might be more toxic than mature fibrils, are mediating α -Syn and A β neurotoxicity (Glabe and Kaye, 2006; Kaye et al., 2003; Volles and Lansbury, 2003). Though the molecular mechanisms underlying amyloid proteins aggregation remain unknown, compounds that can slow and/or prevent the fibrillation and aggregation process of α -Syn or A β , could lead to a new therapeutic strategy for treating PD and AD.

Interestingly, studies have shown that baicalein can inhibit the aggregation of disease-specific amyloid proteins. Low micromolar concentrations of baicalein, and especially its oxidized forms, inhibit the formation of α -Syn fibrils. In addition, existing fibrils of α -Syn are disaggregated by baicalein (Zhu et al., 2004). Subsequent biophysical studies and *in vitro/vivo* studies demonstrated that baicalein is capable of mitigating α -Syn aggregation and reducing cytotoxicity (Caruana et al., 2012; Hu et al., 2016; Jiang et al., 2010; Lu et al., 2011). Baicalein is a potent inhibitor of α -Syn oligomerisation both in cell-free and cellular systems, and is also an effective inhibitor of α -Syn fibrillation in cell-free systems. Baicalein inhibited the formation of α -Syn oligomers in SH-SY5Y and HeLa cells, and protected SH-SY5Y cells from α -Syn -oligomer-induced toxicity (Lu et al., 2011). Besides inhibition of wild-type α -Syn aggregation and cytotoxicity, several researchers found that baicalein also reduced mutant (A30P, A53T and E46K) α -Syn aggregation *in vitro* and plays a protective role in N2A cell models of familial Parkinsonism (Caruana et al., 2012; Jiang et al., 2010). Moreover, Baicalein attenuated α -Syn aggregation in the MPP⁺-treated nigrostriatal dopaminergic system of the rat brain (Hung et al., 2016). In a chronic

PD mouse model created by continuous intragastric administration of rotenone, baicalein did not decrease α -Syn mRNA expression, but α -Syn oligomers were significantly decreased in the ileum, thoracic spinal cord, and midbrain. Taken together, these results suggest that baicalein could prevent the progression of α -Syn accumulation in PD partly by inhibiting formation and aggregation of the α -Syn oligomers (Hu et al., 2016).

In addition, baicalein can also inhibit A β fibrillation and oligomerisation, disaggregate pre-formed A β amyloid fibrils and prevent A β fibril-induced toxicity in PC12 cells (Lu et al., 2011). In cell cultures, baicalein significantly reduced the production of A β by increasing APP α -secretase processing. AD transgenic mice treated daily with baicalein for 8 weeks showed enhanced APP α -secretase processing and reduced A β production (Zhang et al., 2013).

Though the exact molecular mechanism of inhibition of aggregation of amyloid proteins of baicalein is not clear, the underlying mechanism was thoroughly examined by various biochemical and biophysical approaches, and a mechanism based on special molecular structure characteristic of baicalein has been proposed. In fact, it has long been known that polyphenols, such as baicalein and other flavonoids, are reductive substances that are readily oxidized to quinones by oxygen, while quinones are very reactive and can react with the side-chain amino groups of proteins. Oxidized forms of baicalein, baicalein quinones, play a critical role in the inhibition reaction, and the product of the inhibition reaction is predominantly a soluble oligomer of α -Syn, in which the protein molecules have been covalently modified by baicalein quinone to form a Schiff base with a lysine side chain in α -Syn, and Tyr is involved in the interaction of α -Syn with baicalein (Zhu et al., 2004). Further structure-activity analysis showed that vicinal dihydroxyphenyl moieties of baicalein are required for quinone formation and attachment to α -Syn, and baicalein with three vicinal hydroxyl groups exhibited the enhanced inhibitory effects on α -Syn fibrillation compared to baicalin with two vicinal hydroxyl groups (Caruana et al., 2012). The antioxidant activities of baicalein is correlated with their *in vitro* inhibitory effects on α -Syn fibrillation (Meng et al., 2009). Interestingly, a recent study indicated that baicalein-stabilized α -Syn oligomers are able to inhibit fibrillation of non-baicalein-treated α -Syn. These highly stable α -Syn oligomers that inhibit fibrillation do not disrupt the integrity of the biological membrane, suggesting that some forms of soluble oligomer formation can be beneficial (Hong et al., 2008). Therefore, these results suggest that baicalein has potential as a therapeutic agent for the treatment of neurodegenerative diseases such as AD, PD and tauopathies by inhibition of aggregation of disease-specific amyloid proteins.

Anti-inflammatory properties of baicalein

A key element of disease progression in neurodegenerative diseases is the development of a chronic neuroinflammation response in the brain (Ferrari and Tarelli, 2011; Heneka et al., 2015a). Clinical studies observed significant microglial and astrocyte activation in the affected brain regions of patients with PD or AD. High concentrations of proinflammatory cytokines such as interleukin (IL)-1 β , IL-6, and tumor necrosis factor α (TNF- α) have been found in the brain, cerebral spinal fluid, and blood of AD and PD patients (Chen et al., 2008). Neuroinflammatory processes contribute to the cascade of events culminating in neuronal damage. Elevated

production of proinflammatory cytokines by activated microglia and astrocyte can lead to synapse dysfunction and ultimately synapse loss (Herrero et al., 2015). Moreover, the chronic neuroinflammation response is considered to play a role in promoting the formation of pathological plaque including senile plaque (Heneka et al., 2015b) and Lewy's body (Garcia-Esparcia et al., 2014). Therefore, anti-inflammatory medication always is an effective therapeutic or preventive strategy for CNS neurodegenerative diseases (Hirohata et al., 2008).

In oriental countries, the root of *Scutellaria baicalensis Georgi* has been widely used against various inflammatory diseases, and infections of the respiratory and gastrointestinal tract for centuries. As the major active component of the herbal medicine, baicalein has also been reported to exhibit potent anti-inflammatory effects in several topical and systematic inflammatory diseases (Dinda et al., 2017). As early as 1984, baicalein was found to inhibit the acetic acid-induced increase in vascular permeability in mice and to reduce acute paw edema in the rats (Kubo et al., 1984), and baicalein also suppressed the development of secondary lesions in adjuvant-induced arthritis in rats (Kubo et al., 1984). Baicalein also attenuates inflammatory responses in LPS-induced mastitis in mice (He et al., 2015). Moreover, a Chinese patent medicine of baicalein, the capsule preparation of baicalein aluminum, is approved for treatment of enteritis and dysentery in clinic (Wang et al., 2016a).

A growing number of studies have showed that baicalein is capable of inhibiting neuroinflammation in the CNS, and anti-inflammation has been proposed as one of the major mechanisms underlying baicalein-induced neuroprotection. In primary midbrain neuron-glia cultures from E-14 rat embryos, it was found that baicalein exerts potent neuroprotective effect on LPS-induced injury of dopaminergic neurons (Li et al., 2005). Baicalein almost completely blocked LPS-induced activation of microglia, and the excessive production of TNF- α by LPS stimulation was also attenuated by baicalein in a concentration-dependent pattern (Li et al., 2005). In ischemic brain injury, baicalein exhibits its anti-inflammatory activity by inhibiting the infiltration of neutrophil cells (Hwang et al., 2002). Baicalein also exerts a protective effect on BBB disruption in the rat model of intracerebral hemorrhage (ICH). Additionally, baicalein attenuates astroglial activation and inflammasome activation in the MPTP-induced PD model (Cheng et al., 2008; Lee et al., 2014). Moreover, Baicalein alleviates early brain injury after experimental subarachnoid hemorrhage in rats by inhibiting inflammatory pathways (Wang et al., 2015a). Baicalein prevents neuroinflammation and memory impairments in accelerated senescence mice (Jeong et al., 2011). In conclusion, these findings show that baicalein has the potential to inhibit neuroinflammation through attenuation of glial activation and associated cytokine release in CNS neurodegenerative diseases.

Data suggest that the likely mechanism by which baicalein inhibits inflammation is related to the inhibition of the nuclear factor- κ B (NF- κ B) signaling pathways (Chen et al., 2016; Spencer et al., 2012). NF- κ B is a multi-subunit nuclear transcription factor rapidly activating the transcription of various cytokines and chemokines. The expression of many inflammation-related genes is controlled through the NF- κ B signaling pathway (Park and Hong, 2016). Baicalein can inhibit NF- κ B activation and inhibitor of NF- κ B (I κ B) degradation, and significantly inhibits the phosphorylation of p38, extracellular signal-regulated kinase (ERK) and c-Jun N-terminal kinase

(JNK) activated by LPS-induced mouse mastitis (He et al., 2015). In experimental subarachnoid hemorrhage of rats, baicalein also alleviates early brain injury via the TLR4/NF- κ B-mediated inflammatory pathway (Wang et al., 2015a). In the MPTP-induced PD model, baicalein attenuates astroglial activation by downregulating the activations of NF- κ B, ERK, and JNK (Lee et al., 2014).

Besides inhibition of NF- κ B signaling pathways, baicalein may have the potential to inhibit neuroinflammation through the attenuation of iNOS expression and nitric oxide (NO) production. It was shown that baicalein inhibits LPS-induced NO production in a concentration-dependent manner in RAW 264.7 macrophages, which was in parallel with the inhibition of iNOS gene expression (Chen et al., 2001). Moreover, baicalein also inhibited iNOS expression in microglia (Li et al., 2005). Accumulating evidence indicated that iNOS was the most important contributor to NO production in the brain after inflammatory assault, compared with the other isoforms of NOS, i.e., eNOS and nNOS (Iravani et al., 2002). Excessive accumulation of NO has long been known to be toxic to neurons (Steinert et al., 2010). When NO meets with superoxide, peroxynitrite is formed, that is a potent oxidant and nitrating agent capable of attacking and modifying proteins, lipids and DNA as well as depleting antioxidant defenses (Radi, 2013). Some authors think that the powerful neuroprotective effect of baicalein is derived from its dual functions in inhibiting both superoxide and NO production and consequent formation of lethal peroxynitrite (Li et al., 2005). In addition, baicalein may be anti-inflammatory by interacting Nrf2 to regulate the redox balance (Choi et al., 2016) (Lee et al., 2011).

In summary, a significant body of studies has focused on the anti-inflammatory mechanisms underlying baicalein-induced neuroprotection. Therefore, baicalein's anti-inflammatory properties add to its value as a therapy for neurodegenerative diseases.

Baicalein inhibits excitotoxicity

Excitotoxicity induces neuronal degeneration, triggered by the over- or prolonged activation of glutamate receptors in the CNS by excitatory amino acids. Glutamate is a major excitatory neurotransmitter and plays an important role in the mammalian CNS. However, excess glutamate is highly toxic to neurons (Lewerenz and Maher, 2015). Substantial experimental and clinical studies show that glutamate excitotoxicity pathway may play a substantial role in the pathogenesis of neuronal death in central neurodegenerative diseases such as AD (Esposito et al., 2013; Tannenberg et al., 2004) and PD (Blandini et al., 2001). The clinical use of the noncompetitive N-methyl-d-aspartate (NMDA) antagonist memantine in monotherapy or combination therapy with cholinesterase inhibitors improved cognition, behavior, and activities of daily living of AD patients (Matsunaga et al., 2015; Owen, 2016) and PD dementia patients (Olivares et al., 2012; Wang et al., 2015b). Therefore, inhibition of glutamate release or antagonism of glutamate receptors is a potentially valuable therapeutic strategy for treating these diseases.

Recently, some authors demonstrated that baicalein can inhibit excitotoxicity in several animal models of neurodegeneration. It was found that baicalein can protect neurons from a variety of stresses, including those exerted by glutamate (Lee et al., 2003). Baicalein also attenuates

methamphetamine-induced loss of dopamine transporter in mouse striatum (Wu et al., 2006). In the PD rat model induced by 6-OHDA-medial forebrain bundle (MFB) injection, baicalein plays a neuromodulatory role in balancing gamma aminobutyric acid (GABA) and glutamate neurotransmitter levels in the basal ganglia (Yu et al., 2012). In rat primary mesencephalic cultures, baicalein inhibited the glutamate -induced intracellular calcium ion increase. Baicalein also showed neuroprotective effects on excitotoxic neuronal cell death in primary rat cortical cell cultures (Yang et al., 2014). Interestingly, an improved drugs screening system revealed that baicalein not only ameliorates the A β -induced depolarization of neurons, but also possibly functions as an antagonist of AMPA and NMDA receptors (Lin et al., 2017). In rat hippocampal nerve terminals (synaptosomes), baicalein inhibits depolarization-induced glutamate release, while in slice preparations whole cell patch-clamp experiments revealed that baicalein reduced the frequency of miniature excitatory postsynaptic currents without affecting their amplitude. Further studies showed that inhibiting excitotoxicity of baicalein was mediated through the inhibition of NMDA receptor function by interacting with the glycine binding site of the NMDA receptor. In animal models of neurodegeneration, excitotoxicity is commonly induced experimentally by kainic acid (Mohd Sairazi et al., 2015). In a kainic acid rat model, intraperitoneally administering baicalein substantially attenuated kainic acid-induced neuronal cell death, c-Fos expression, and the activation of the mammalian target of rapamycin in the hippocampus (Chang et al., 2016). Therefore, these findings suggest that baicalein is valuable for treating brain disorders related to glutamate excitotoxicity.

Neurogenesis and cell differentiation action of baicalein

Neuronal loss in the disease-specific brain regions following neurotransmitter decrease and synapse loss are the main cause of the typical cognitive and motor symptoms of neurodegenerative diseases. In AD, the basal forebrain, the hippocampus and its neighboring cortical structures within the temporal lobe lose a large number of cholinergic neurons, and acetylcholine neurotransmission is lost, which impairs the cognitive ability of AD patients (Lombardo and Maskos, 2015). In PD, there is loss of dopaminergic neurons in the substantia nigra, and loss of dopaminergic projections to the basal ganglia, which declines motor ability of PD patients (Gaig and Tolosa, 2009). It is now widely accepted that lack of endogenous neurotrophic factors, such as glial cell line-derived neurotrophic factor (GDNF), nerve growth factor (NGF) and brain-derived neurotrophic factor (BDNF), is involved in several neurodegenerative diseases (Allen et al., 2013; Holscher, 2014; Lee and Son, 2009). Therefore, any therapeutics that can promote neurotrophic and neurogenesis to remedy neuronal loss will be a promising therapeutic strategy for neurodegenerative diseases.

Recently, both neurotrophic and neurogenesis action of baicalein has been verified in a series of studies, in which the molecular mechanisms of the neurogenetic effect of baicalein have been studied. Baicalein can attenuate the irradiation-induced impairment of hippocampal neurogenesis by modulating oxidative stress and elevating BDNF-pCREB signaling (Oh et al., 2013). Moreover, baicalein might also promote neurogenesis in MPTP-treated mice by up-regulation of a series of genes such as SRY (sex-determining region Y)-box 8 (SOX8) and SRY (sex-determining region Y)-box 11 (SOX11), genes that can promote endogenous neurogenesis

(Gao et al., 2015). In addition, baicalein enhanced the survival of HiB5, a hippocampal progenitor cell line, and its differentiation to ChAT immunoreactive cells (Heo et al., 2009). Baicalein also induced cortical neurogenesis after focal cerebral ischemia, and baicalein treatment was found to promote differentiation of cortical precursor cells into neuronal but not glial cells, which may be attributable to the regulation of AKT, GSK-3 β mRNA and Ang-1 protein levels (Zou et al., 2016). Therefore, these findings suggest that baicalein most likely acts as a neurotrophic and neurogenetic drug that may be useful for treating neurodegenerative diseases.

Anti-apoptotic effects of baicalein

During development and maturation of the CNS, neuronal apoptosis is a physiological process that is an integral part of neurogenesis. However, aberrant apoptosis has been implicated in the pathogenesis of neurodegeneration (Okouchi et al., 2007). Evidence of increased neuronal apoptosis, such as DNA damage, apoptotic bodies, chromatin condensation, and the induction of select genes that take part in apoptosis including bax and caspases, have been observed in affected brain regions (the hippocampus or the substantia nigra) of the AD or PD brain (Rohn and Head, 2009; Schulz, 2006). These neuropathological processes were found in cell culture and animal models of neurodegeneration induced by neurotoxins (Bove and Perier, 2012; Cotman and Su, 1996). Based on the available data in animal models and studies involving postmortem patients, therefore, several potential candidates of the apoptotic machinery have been uncovered as possible drug targets for the treatment of AD or PD (Rohn and Head, 2009).

Several studies in cellular and animal models have shown that baicalein may have anti-apoptotic effects in neurons. Baicalein can block 6-OHDA-induced SH-SY5Y cell apoptosis (Mu et al., 2009). Furthermore, the mitochondrial pathway associated with cell apoptosis including membrane potential loss, the release of cytochrome c, the downregulation of Bcl-2, upregulation of Bax induced by H₂O₂ was also abrogated in the presence of baicalein (Zhang et al., 2010). Moreover, baicalein suppressed rotenone-induced PC12 cells apoptosis, and inhibited the accumulation of ROS, ATP deficiency, mitochondrial membrane potential dissipation, and caspase-3/7 activation in a concentration-dependent manner (Li et al., 2012). In addition, baicalein inhibited MPP⁺-induced apoptosis and the activation of caspases 9 and 12 in the rat nigrostriatal dopaminergic system (Hung et al., 2016). Baicalin effectively prevented A β -induced mitochondrial membrane potential decrease, Bax/Bcl-2 ratio increase, cytochrome c release, and caspase-9/-3 activation. In primary cultured cortical neurons stressed by oxygen and glucose deprivation, similarly, baicalein increased Bcl-2/Bcl-xL-associated protein phosphorylation and maintained the protein levels of Bcl-2 in mitochondria, which subsequently reduced cytochrome c release into the cytosol (Liu et al., 2010). Baicalein protected HT22 murine hippocampal neuronal cells against endoplasmic reticulum stress-induced apoptosis via inhibition of ROS production and CHOP induction (Choi et al., 2010). Furthermore, baicalein also markedly reduced apoptosis in the penumbra of transient middle cerebral artery occlusion rats (Liu et al., 2010). These results showed that baicalein-induced attenuation of neurotoxins -induced apoptosis is mediated via inhibiting mitochondrial and ER stress pathways. Thus, baicalein may be a beneficial neuroprotective drug by inhibiting neuronal apoptosis.

Clinical trials of baicalein

Baicalein has potential as a novel neuroprotective agent for the treatment of neurodegenerative diseases. Hence, clinical trials of baicalein for treating neurodegenerative diseases are necessary. Notably, two Phase I clinical trials of baicalein chewable tablets in healthy Chinese adult volunteers have been completed in China.

In 2014, a Phase I, randomized, double-blind, single-dose trial of baicalein (100-2800 mg) in 72 healthy Chinese adults investigated the pharmacokinetic (PK) properties of baicalein and its main metabolite, baicalin. Samples of blood, urine and feces were collected at regular intervals up to 48 h after administration of the drug. The drug concentration was then analyzed using liquid chromatography-tandem mass spectrometry (LC/MS/MS). Physical examinations, vital signs, ECG findings, hematology, and urinalysis were monitored before and at regular intervals after administration of the drug. The PK profile of baicalein and baicalin was characterized by a median T_{max} of 0.75-3.5 h and 0.5-3 h, respectively, followed by a multiphasic profile with a $t_{1/2}$ of 1.90-15.01 h and 4.22-10.80 h, respectively. The estimates of the proportionality coefficient (90% CI) for C_{max} , AUC_{0-t} and $AUC_{0-\infty}$ were 0.83 (0.70-0.96), 0.91 (0.81-1.00) and 0.92 (0.82-1.02), respectively. All values overlapped within the pre-specified range of (0.89-1.11), (0.93-1.07), and (0.93-1.07), respectively. The total urinary clearance of baicalein and baicalin was <1%. And approximately 27% of baicalein was eliminated as unchanged drug in feces. Moreover, baicalein was well tolerated. Eleven treatment-related adverse events (hyperactive bowel sounds, abdominal distention, constipation, dizziness, somnolence, blurred vision, plasma fibrinogen decreased, blood leukocyte decreased, etc.) were observed in the higher dose group, and all were rated as "mild" and resolved without further treatment. There were no clinically relevant changes in blood pressure or ECG in individuals during the study. No serious adverse events occurred, and clinical laboratory assessments showed no signs of toxicity in the liver or kidney. (Li et al., 2014)

In 2016, another single-center, double-blind, placebo-controlled, parallel-group study investigated the PK, safety and tolerability of baicalein after a multiple-ascending-dose protocol in healthy Chinese volunteers. In this clinical trial, participants were randomized to receive baicalein (n = 8 per dose regimen) or placebo (n = 2 per dose regimen). Dosing regimens were 200, 400, and 800 mg once daily on days 1 and 10, twice daily on days 3-9. Plasma, urine, and feces samples were assayed for baicalein and its predominant metabolite baicalin using validated HPLC-MS/MS methods. Thirty-three of 36 enrolled participants completed the study. A total of 44 adverse events occurred in 23 participants. A steady-state concentration of analytes in plasma was achieved on day 8 after repeated dosing. Analytes concentrations and exposure increased with increasing dose. The dose proportionality constant (β) for AUC_{ss} of baicalein and baicalin was 0.922 (90 % confidence interval, 0.650-1.195) and 0.942 (90 % confidence interval, 0.539-1.345), respectively. The accumulation index varied from 1.66 to 2.07 for baicalein and from 1.68 to 2.45 for baicalin. In conclusion, in dose range of 200-800 mg, multiple-dose oral baicalein administration was safe and well tolerated, dose proportionality was inconclusive, and no serious accumulation of baicalein was observed (Pang et al., 2016).

Although two Phase I clinical trials have indicated that oral baicalein is safe and well tolerated, , clinical trials for other dosages for baicalein are not reported, and further phase II and III clinical trials of baicalein in AD and PD patients will have to be conducted to prove its efficiency.

Conclusion

The neuroprotective effects of baicalein in AD and PD have been widely investigated in *in vitro/vivo* studies. To summarize, baicalein shows antioxidant effects, has anti-inflammatory properties, inhibits the aggregation of disease-specific amyloid proteins, reduces excitotoxicity, enhances neurogenesis and cell differentiation, and has anti-apoptotic effects. This forms a solid base to propose baicalein as a treatment for neurodegenerative diseases (Fig. 1). Importantly, two phase I clinical trials of baicalein in healthy Chinese adult volunteers have been completed (Li et al., 2014; Pang et al., 2016) and confirmed that baicalein is safe and well tolerated by healthy subjects. Thus, all of these properties indicate that the therapeutic implications of baicalein may prove successful in slowing or even halting further progression of CNS neurodegenerative diseases. Baicalein has the potential to be used alone or in combination with existing drugs for AD and PD patients. As the next step, clinical trials testing the effects in patients with neurodegenerative disorders will have to be initiated to test what the actual benefits will be.

Compliance with Ethical Standards

Funding

This work was supported by research project funding of the Education Department of Hunan Province in China (NO: 14C1039).

Conflicts of Interest

Yanwei Li, Jinying Zhao, and Christian Hölscher declare no conflicts of interest.

References

- Adhikari, S., Tilak, J. C., Devasagayam, T. P., 2011. Free radical reactions of a naturally occurring flavone baicalein and possible mechanisms towards its membrane protective properties. *Indian J Biochem Biophys* 48, 275-282.
- Allen, S. J., Watson, J. J., Shoemark, D. K., Barua, N. U., Patel, N. K., 2013. GDNF, NGF and BDNF as therapeutic options for neurodegeneration. *Pharmacol Ther* 138, 155-175.
- Asanuma, M., Miyazaki, I., Diaz-Corrales, F. J., Ogawa, N., 2004. Quinone formation as dopaminergic neuron-specific oxidative stress in the pathogenesis of sporadic Parkinson's disease and neurotoxin-induced parkinsonism. *Acta Med Okayama* 58, 221-233.
- Asanuma, M., Miyazaki, I., Ogawa, N., 2003. Dopamine- or L-DOPA-induced neurotoxicity: the role of dopamine quinone formation and tyrosinase in a model of Parkinson's disease. *Neurotox Res* 5, 165-176.
- Aslan, M., Ozben, T., 2004. Reactive oxygen and nitrogen species in Alzheimer's disease. *Curr Alzheimer Res* 1, 111-119.
- Blandini, F., Greenamyre, J. T., Fancellu, R., Nappi, G., 2001. Blockade of subthalamic glutamatergic activity corrects changes in neuronal metabolism and motor behavior in rats with nigrostriatal lesions. *Neurol Sci* 22, 49-50.

Bove, J., Perier, C., 2012. Neurotoxin-based models of Parkinson's disease. *Neuroscience* 211, 51-76.

Caruana, M., Neuner, J., Hogen, T., Schmidt, F., Kamp, F., Scerri, C., Giese, A., Vassallo, N., 2012. Polyphenolic compounds are novel protective agents against lipid membrane damage by alpha-synuclein aggregates in vitro. *Biochim Biophys Acta* 1818, 2502-2510.

Cavallucci, V., D'Amelio, M., Cecconi, F., 2012. Abeta toxicity in Alzheimer's disease. *Mol Neurobiol* 45, 366-378.

Chan, J. Y., Chan, S. H., 2015. Activation of endogenous antioxidants as a common therapeutic strategy against cancer, neurodegeneration and cardiovascular diseases: A lesson learnt from DJ-1. *Pharmacol Ther* 156, 69-74.

Chang, Y., Lu, C. W., Lin, T. Y., Huang, S. K., Wang, S. J., 2016. Baicalein, a Constituent of *Scutellaria baicalensis*, Reduces Glutamate Release and Protects Neuronal Cell Against Kainic Acid-Induced Excitotoxicity in Rats. *Am J Chin Med* 44, 943-962.

Chen, H., O'Reilly, E. J., Schwarzschild, M. A., Ascherio, A., 2008. Peripheral inflammatory biomarkers and risk of Parkinson's disease. *Am J Epidemiol* 167, 90-95.

Chen, M., Lai, L., Li, X., Zhang, X., He, X., Liu, W., Li, R., Ke, X., Fu, C., Huang, Z., Duan, C., 2016. Baicalein Attenuates Neurological Deficits and Preserves Blood-Brain Barrier Integrity in a Rat Model of Intracerebral Hemorrhage. *Neurochem Res* 41, 3095-3102.

Chen, Y. C., Shen, S. C., Chen, L. G., Lee, T. J., Yang, L. L., 2001. Wogonin, baicalin, and baicalein inhibition of inducible nitric oxide synthase and cyclooxygenase-2 gene expressions induced by nitric oxide synthase inhibitors and lipopolysaccharide. *Biochem Pharmacol* 61, 1417-1427.

Cheng, Y., He, G., Mu, X., Zhang, T., Li, X., Hu, J., Xu, B., Du, G., 2008. Neuroprotective effect of baicalein against MPTP neurotoxicity: behavioral, biochemical and immunohistochemical profile. *Neurosci Lett* 441, 16-20.

Choi, E. O., Jeong, J. W., Park, C., Hong, S. H., Kim, G. Y., Hwang, H. J., Cho, E. J., Choi, Y. H., 2016. Baicalein protects C6 glial cells against hydrogen peroxide-induced oxidative stress and apoptosis through regulation of the Nrf2 signaling pathway. *Int J Mol Med* 37, 798-806.

Choi, J. H., Choi, A. Y., Yoon, H., Choe, W., Yoon, K. S., Ha, J., Yeo, E. J., Kang, I., 2010. Baicalein protects HT22 murine hippocampal neuronal cells against endoplasmic reticulum stress-induced apoptosis through inhibition of reactive oxygen species production and CHOP induction. *Exp Mol Med* 42, 811-822.

Chu, J., Li, J. G., Giannopoulos, P. F., Blass, B. E., Childers, W., Abou-Gharbia, M., Pratico, D., 2015. Pharmacologic blockade of 12/15-lipoxygenase ameliorates memory deficits, Abeta and tau neuropathology in the triple-transgenic mice. *Mol Psychiatry* 20, 1329-1338.

Chu, J., Zhuo, J. M., Pratico, D., 2012. Transcriptional regulation of beta-secretase-1 by 12/15-lipoxygenase results in enhanced amyloidogenesis and cognitive impairments. *Ann Neurol* 71, 57-67.

Cotman, C. W., Su, J. H., 1996. Mechanisms of neuronal death in Alzheimer's disease. *Brain Pathol* 6, 493-506.

Deschamps, J. D., Kenyon, V. A., Holman, T. R., 2006. Baicalein is a potent in vitro inhibitor against both reticulocyte 15-human and platelet 12-human lipoxygenases. *Bioorg Med Chem* 14, 4295-4301.

Dinda, B., Dinda, S., DasSharma, S., Banik, R., Chakraborty, A., Dinda, M., 2017. Therapeutic potentials of baicalin and its aglycone, baicalein against inflammatory disorders. *Eur J Med Chem* 131, 68-80.

Do Van, B., Gouel, F., Jonneaux, A., Timmerman, K., Gele, P., Petrault, M., Bastide, M., Laloux, C., Moreau, C., Bordet, R., Devos, D., Devedjian, J. C., 2016. Ferroptosis, a newly characterized form of cell

death in Parkinson's disease that is regulated by PKC. *Neurobiol Dis* 94, 169-178.

Dusek, P., Schneider, S. A., Aaseth, J., 2016. Iron chelation in the treatment of neurodegenerative diseases. *J Trace Elem Med Biol* 38, 81-92.

Esposito, Z., Belli, L., Toniolo, S., Sancesario, G., Bianconi, C., Martorana, A., 2013. Amyloid beta, glutamate, excitotoxicity in Alzheimer's disease: are we on the right track? *CNS Neurosci Ther* 19, 549-555.

Ferrari, C. C., Tarelli, R., 2011. Parkinson's disease and systemic inflammation. *Parkinsons Dis* 2011, 436813.

Gaig, C., Tolosa, E., 2009. When does Parkinson's disease begin? *Mov Disord* 24 Suppl 2, S656-664.

Gao, D., Sakurai, K., Chen, J., Ogiso, T., 1995. Protection by baicalein against ascorbic acid-induced lipid peroxidation of rat liver microsomes. *Res Commun Mol Pathol Pharmacol* 90, 103-114.

Gao, D., Tawa, R., Masaki, H., Okano, Y., Sakurai, H., 1998. Protective effects of baicalein against cell damage by reactive oxygen species. *Chem Pharm Bull (Tokyo)* 46, 1383-1387.

Gao, L., Li, C., Yang, R. Y., Lian, W. W., Fang, J. S., Pang, X. C., Qin, X. M., Liu, A. L., Du, G. H., 2015. Ameliorative effects of baicalein in MPTP-induced mouse model of Parkinson's disease: A microarray study. *Pharmacol Biochem Behav* 133, 155-163.

Gao, Z., Huang, K., Xu, H., 2001. Protective effects of flavonoids in the roots of *Scutellaria baicalensis* Georgi against hydrogen peroxide-induced oxidative stress in HS-SY5Y cells. *Pharmacol Res* 43, 173-178.

Gao, Z., Huang, K., Yang, X., Xu, H., 1999. Free radical scavenging and antioxidant activities of flavonoids extracted from the radix of *Scutellaria baicalensis* Georgi. *Biochim Biophys Acta* 1472, 643-650.

Garcia-Esparcia, P., Llorens, F., Carmona, M., Ferrer, I., 2014. Complex deregulation and expression of cytokines and mediators of the immune response in Parkinson's disease brain is region dependent. *Brain Pathol* 24, 584-598.

Gasiorowski, K., Lamer-Zarawska, E., Leszek, J., Parvathaneni, K., Yendluri, B. B., Blach-Olszewska, Z., Aliev, G., 2011. Flavones from root of *Scutellaria baicalensis* Georgi: drugs of the future in neurodegeneration? *CNS Neurol Disord Drug Targets* 10, 184-191.

Giannopoulos, P. F., Joshi, Y. B., Chu, J., Pratico, D., 2013. The 12-15-lipoxygenase is a modulator of Alzheimer's-related tau pathology in vivo. *Aging Cell* 12, 1082-1090.

Glabe, C. G., Kaye, R., 2006. Common structure and toxic function of amyloid oligomers implies a common mechanism of pathogenesis. *Neurology* 66, S74-78.

Goedert, M., 2015. NEURODEGENERATION. Alzheimer's and Parkinson's diseases: The prion concept in relation to assembled A β , tau, and alpha-synuclein. *Science* 349, 1255555.

Gu, X. H., Xu, L. J., Liu, Z. Q., Wei, B., Yang, Y. J., Xu, G. G., Yin, X. P., Wang, W., 2016. The flavonoid baicalein rescues synaptic plasticity and memory deficits in a mouse model of Alzheimer's disease. *Behav Brain Res* 311, 309-321.

Guiney, S. J., Adlard, P. A., Bush, A. I., Finkelstein, D. I., Ayton, S., 2017. Ferroptosis and cell death mechanisms in Parkinson's disease. *Neurochem Int* 104, 34-48.

Hamada, H., Hiramatsu, M., Edamatsu, R., Mori, A., 1993. Free radical scavenging action of baicalein. *Arch Biochem Biophys* 306, 261-266.

Hambright, W. S., Fonseca, R. S., Chen, L., Na, R., Ran, Q., 2017. Ablation of ferroptosis regulator glutathione peroxidase 4 in forebrain neurons promotes cognitive impairment and neurodegeneration. *Redox Biol* 12, 8-17.

He, X., Wei, Z., Zhou, E., Chen, L., Kou, J., Wang, J., Yang, Z., 2015. Baicalein attenuates inflammatory responses by suppressing TLR4 mediated NF-kappaB and MAPK signaling pathways in LPS-induced mastitis in mice. *Int Immunopharmacol* 28, 470-476.

Heneka, M. T., Carson, M. J., El Khoury, J., Landreth, G. E., Brosseron, F., Feinstein, D. L., Jacobs, A. H., Wyss-Coray, T., Vitorica, J., Ransohoff, R. M., Herrup, K., Frautschy, S. A., Finsen, B., Brown, G. C., Verkhratsky, A., Yamanaka, K., Koistinaho, J., Latz, E., Halle, A., Petzold, G. C., Town, T., Morgan, D., Shinohara, M. L., Perry, V. H., Holmes, C., Bazan, N. G., Brooks, D. J., Hunot, S., Joseph, B., Deigendesch, N., Garaschuk, O., Boddeke, E., Dinarello, C. A., Breitner, J. C., Cole, G. M., Golenbock, D. T., Kummer, M. P., 2015a. Neuroinflammation in Alzheimer's disease. *Lancet Neurol* 14, 388-405.

Heneka, M. T., Golenbock, D. T., Latz, E., 2015b. Innate immunity in Alzheimer's disease. *Nat Immunol* 16, 229-236.

Heo, H., Shin, Y., Cho, W., Choi, Y., Kim, H., Kwon, Y. K., 2009. Memory improvement in ibotenic acid induced model rats by extracts of *Scutellaria baicalensis*. *J Ethnopharmacol* 122, 20-27.

Heo, H. J., Kim, D. O., Choi, S. J., Shin, D. H., Lee, C. Y., 2004. Potent Inhibitory effect of flavonoids in *Scutellaria baicalensis* on amyloid beta protein-induced neurotoxicity. *J Agric Food Chem* 52, 4128-4132.

Herrero, M. T., Estrada, C., Maatouk, L., Vyas, S., 2015. Inflammation in Parkinson's disease: role of glucocorticoids. *Front Neuroanat* 9, 32.

Hirohata, M., Ono, K., Morinaga, A., Yamada, M., 2008. Non-steroidal anti-inflammatory drugs have potent anti-fibrillogenic and fibril-destabilizing effects for alpha-synuclein fibrils in vitro. *Neuropharmacology* 54, 620-627.

Hong, D. P., Fink, A. L., Uversky, V. N., 2008. Structural characteristics of alpha-synuclein oligomers stabilized by the flavonoid baicalein. *J Mol Biol* 383, 214-223.

Hu, Q., Uversky, V. N., Huang, M., Kang, H., Xu, F., Liu, X., Lian, L., Liang, Q., Jiang, H., Liu, A., Zhang, C., Pan-Montojo, F., Zhu, S., 2016. Baicalein inhibits alpha-synuclein oligomer formation and prevents progression of alpha-synuclein accumulation in a rotenone mouse model of Parkinson's disease. *Biochim Biophys Acta* 1862, 1883-1890.

Huang, Y., Tsang, S. Y., Yao, X., Chen, Z. Y., 2005. Biological properties of baicalein in cardiovascular system. *Curr Drug Targets Cardiovasc Haematol Disord* 5, 177-184.

Hung, K. C., Huang, H. J., Wang, Y. T., Lin, A. M., 2016. Baicalein attenuates alpha-synuclein aggregation, inflammasome activation and autophagy in the MPP+-treated nigrostriatal dopaminergic system in vivo. *J Ethnopharmacol* 194, 522-529.

Hwang, Y. S., Shin, C. Y., Huh, Y., Ryu, J. H., 2002. Hwangryun-Hae-Dok-tang (Huanglian-Jie-Du-Tang) extract and its constituents reduce ischemia-reperfusion brain injury and neutrophil infiltration in rats. *Life Sci* 71, 2105-2117.

Iravani, M. M., Kashafi, K., Mander, P., Rose, S., Jenner, P., 2002. Involvement of inducible nitric oxide synthase in inflammation-induced dopaminergic neurodegeneration. *Neuroscience* 110, 49-58.

Jeong, K., Shin, Y. C., Park, S., Park, J. S., Kim, N., Um, J. Y., Go, H., Sun, S., Lee, S., Park, W., Choi, Y., Song, Y., Kim, G., Jeon, C., Park, J., Lee, K., Bang, O., Ko, S. G., 2011. Ethanol extract of *Scutellaria baicalensis* Georgi prevents oxidative damage and neuroinflammation and memorial impairments in artificial senescence mice. *J Biomed Sci* 18, 14.

Ji, S., Li, R., Wang, Q., Miao, W. J., Li, Z. W., Si, L. L., Qiao, X., Yu, S. W., Zhou, D. M., Ye, M., 2015. Anti-H1N1 virus, cytotoxic and Nrf2 activation activities of chemical constituents from *Scutellaria baicalensis*. *J Ethnopharmacol* 176, 475-484.

Jiang, M., Porat-Shliom, Y., Pei, Z., Cheng, Y., Xiang, L., Sommers, K., Li, Q., Gillardon, F., Hengerer, B., Berlinicke, C., Smith, W. W., Zack, D. J., Poirier, M. A., Ross, C. A., Duan, W., 2010. Baicalein reduces E46K alpha-synuclein aggregation in vitro and protects cells against E46K alpha-synuclein toxicity in cell models of familiar Parkinsonism. *J Neurochem* 114, 419-429.

Joshi, Y. B., Giannopoulos, P. F., Pratico, D., 2015. The 12/15-lipoxygenase as an emerging therapeutic target for Alzheimer's disease. *Trends Pharmacol Sci* 36, 181-186.

Kang, K. H., Liou, H. H., Hour, M. J., Liou, H. C., Fu, W. M., 2013. Protection of dopaminergic neurons by 5-lipoxygenase inhibitor. *Neuropharmacology* 73, 380-387.

Kayed, R., Head, E., Thompson, J. L., McIntire, T. M., Milton, S. C., Cotman, C. W., Glabe, C. G., 2003. Common structure of soluble amyloid oligomers implies common mechanism of pathogenesis. *Science* 300, 486-489.

Khan, T. A., Hassan, I., Ahmad, A., Perveen, A., Aman, S., Quddusi, S., Alhazza, I. M., Ashraf, G. M., Aliev, G., 2016. Recent Updates on the Dynamic Association Between Oxidative Stress and Neurodegenerative Disorders. *CNS Neurol Disord Drug Targets* 15, 310-320.

Kimuya, Y., Kubo, M., Tani, T., Arichi, S., Okuda, H., 1981. Studies on *Scutellariae Radix*. IV. Effects on lipid peroxidation in rat liver. *Chem Pharm Bull (Tokyo)* 29, 2610-2617.

Korolainen, M. A., Goldsteins, G., Nyman, T. A., Alafuzoff, I., Koistinaho, J., Pirttila, T., 2006. Oxidative modification of proteins in the frontal cortex of Alzheimer's disease brain. *Neurobiol Aging* 27, 42-53.

Kowal, S. L., Dall, T. M., Chakrabarti, R., Storm, M. V., Jain, A., 2013. The current and projected economic burden of Parkinson's disease in the United States. *Mov Disord* 28, 311-318.

Kruer, M. C., 2013. The neuropathology of neurodegeneration with brain iron accumulation. *Int Rev Neurobiol* 110, 165-194.

Kubo, M., Matsuda, H., Tanaka, M., Kimura, Y., Okuda, H., Higashino, M., Tani, T., Namba, K., Arichi, S., 1984. Studies on *Scutellariae radix*. VII. Anti-arthritic and anti-inflammatory actions of methanolic extract and flavonoid components from *Scutellariae radix*. *Chem Pharm Bull (Tokyo)* 32, 2724-2729.

Kulshreshtha, A., Piplani, P., 2016. Current pharmacotherapy and putative disease-modifying therapy for Alzheimer's disease. *Neurol Sci* 37, 1403-1435.

Kumar, A., Singh, A., Ekavali, 2015. A review on Alzheimer's disease pathophysiology and its management: an update. *Pharmacol Rep* 67, 195-203.

Lee, E., Park, H. R., Ji, S. T., Lee, Y., Lee, J., 2014. Baicalein attenuates astroglial activation in the 1-methyl-4-phenyl-1,2,3,4-tetrahydropyridine-induced Parkinson's disease model by downregulating the activations of nuclear factor-kappaB, ERK, and JNK. *J Neurosci Res* 92, 130-139.

Lee, E., Son, H., 2009. Adult hippocampal neurogenesis and related neurotrophic factors. *BMB Rep* 42, 239-244.

Lee, H. H., Yang, L. L., Wang, C. C., Hu, S. Y., Chang, S. F., Lee, Y. H., 2003. Differential effects of natural polyphenols on neuronal survival in primary cultured central neurons against glutamate- and glucose deprivation-induced neuronal death. *Brain Res* 986, 103-113.

Lee, H. J., Noh, Y. H., Lee, D. Y., Kim, Y. S., Kim, K. Y., Chung, Y. H., Lee, W. B., Kim, S. S., 2005. Baicalein attenuates 6-hydroxydopamine-induced neurotoxicity in SH-SY5Y cells. *Eur J Cell Biol* 84, 897-905.

Lee, I. K., Kang, K. A., Zhang, R., Kim, B. J., Kang, S. S., Hyun, J. W., 2011. Mitochondria protection of baicalein against oxidative damage via induction of manganese superoxide dismutase. *Environ Toxicol Pharmacol* 31, 233-241.

Lewerenz, J., Maher, P., 2015. Chronic Glutamate Toxicity in Neurodegenerative Diseases-What is the Evidence? *Front Neurosci* 9, 469.

Li, C., Lin, G., Zuo, Z., 2011. Pharmacological effects and pharmacokinetics properties of Radix Scutellariae and its bioactive flavones. *Biopharm Drug Dispos* 32, 427-445.

Li, F. Q., Wang, T., Pei, Z., Liu, B., Hong, J. S., 2005. Inhibition of microglial activation by the herbal flavonoid baicalein attenuates inflammation-mediated degeneration of dopaminergic neurons. *J Neural Transm (Vienna)* 112, 331-347.

Li, J., O, W., Li, W., Jiang, Z. G., Ghanbari, H. A., 2013a. Oxidative stress and neurodegenerative disorders. *Int J Mol Sci* 14, 24438-24475.

Li, M., Shi, A., Pang, H., Xue, W., Li, Y., Cao, G., Yan, B., Dong, F., Li, K., Xiao, W., He, G., Du, G., Hu, X., 2014. Safety, tolerability, and pharmacokinetics of a single ascending dose of baicalein chewable tablets in healthy subjects. *J Ethnopharmacol* 156, 210-215.

Li, X. X., He, G. R., Mu, X., Xu, B., Tian, S., Yu, X., Meng, F. R., Xuan, Z. H., Du, G. H., 2012. Protective effects of baicalein against rotenone-induced neurotoxicity in PC12 cells and isolated rat brain mitochondria. *Eur J Pharmacol* 674, 227-233.

Li, X. Z., Zhang, S. N., Liu, S. M., Lu, F., 2013b. Recent advances in herbal medicines treating Parkinson's disease. *Fitoterapia* 84, 273-285.

Li, Z., Choi, D. Y., Shin, E. J., Hunter, R. L., Jin, C. H., Wie, M. B., Kim, M. S., Park, S. J., Bing, G., Kim, H. C., 2008. Phendone protects the nigral dopaminergic neurons from LPS-induced neurotoxicity. *Neurosci Lett* 445, 1-6.

Lin, T. S., Tsai, H. J., Lee, C. H., Song, Y. Q., Huang, R. S., Hsieh-Li, H. M., Liang, M. C., Lin, Y., 2017. An Improved Drugs Screening System Reveals that Baicalein Ameliorates the Abeta/AMPA/NMDA-Induced Depolarization of Neurons. *J Alzheimers Dis* 56, 959-976.

Liu, C., Wu, J., Xu, K., Cai, F., Gu, J., Ma, L., Chen, J., 2010. Neuroprotection by baicalein in ischemic brain injury involves PTEN/AKT pathway. *J Neurochem* 112, 1500-1512.

Liu, R. X., Song, G. H., Wu, P. G., Zhang, X. W., Hu, H. J., Liu, J., Miao, X. S., Hou, Z. Y., Wang, W. Q., Wei, S. L., 2017. Distribution patterns of the contents of five biologically activate ingredients in the root of *Scutellaria baicalensis*. *Chin J Nat Med* 15, 152-160.

Lombardo, S., Maskos, U., 2015. Role of the nicotinic acetylcholine receptor in Alzheimer's disease pathology and treatment. *Neuropharmacology* 96, 255-262.

Lu, J. H., Ardah, M. T., Durairajan, S. S., Liu, L. F., Xie, L. X., Fong, W. F., Hasan, M. Y., Huang, J. D., El-Agnaf, O. M., Li, M., 2011. Baicalein inhibits formation of alpha-synuclein oligomers within living cells and prevents Abeta peptide fibrillation and oligomerisation. *Chembiochem* 12, 615-624.

Matsunaga, S., Kishi, T., Iwata, N., 2015. Memantine monotherapy for Alzheimer's disease: a systematic review and meta-analysis. *PLoS One* 10, e0123289.

Mazzetti, A. P., Fiorile, M. C., Primavera, A., Lo Bello, M., 2015. Glutathione transferases and neurodegenerative diseases. *Neurochem Int* 82, 10-18.

Meng, X., Munishkina, L. A., Fink, A. L., Uversky, V. N., 2009. Molecular mechanisms underlying the flavonoid-induced inhibition of alpha-synuclein fibrillation. *Biochemistry* 48, 8206-8224.

Miyazaki, I., Asanuma, M., 2009. Approaches to prevent dopamine quinone-induced neurotoxicity. *Neurochem Res* 34, 698-706.

Mladenka, P., Macakova, K., Filipicky, T., Zatloukalova, L., Jahodar, L., Bovicelli, P., Silvestri, I. P., Hrdina, R., Saso, L., 2011. In vitro analysis of iron chelating activity of flavonoids. *J Inorg Biochem* 105, 693-701.

Mohd Sairazi, N. S., Sirajudeen, K. N., Asari, M. A., Muzaimi, M., Mummedy, S., Sulaiman, S. A., 2015. Kainic Acid-Induced Excitotoxicity Experimental Model: Protective Merits of Natural Products and

Plant Extracts. *Evid Based Complement Alternat Med* 2015, 972623.

More, S. V., Kumar, H., Kang, S. M., Song, S. Y., Lee, K., Choi, D. K., 2013. Advances in neuroprotective ingredients of medicinal herbs by using cellular and animal models of Parkinson's disease. *Evid Based Complement Alternat Med* 2013, 957875.

Mu, X., He, G., Cheng, Y., Li, X., Xu, B., Du, G., 2009. Baicalein exerts neuroprotective effects in 6-hydroxydopamine-induced experimental parkinsonism in vivo and in vitro. *Pharmacol Biochem Behav* 92, 642-648.

Oh, S. B., Park, H. R., Jang, Y. J., Choi, S. Y., Son, T. G., Lee, J., 2013. Baicalein attenuates impaired hippocampal neurogenesis and the neurocognitive deficits induced by gamma-ray radiation. *Br J Pharmacol* 168, 421-431.

Okouchi, M., Ekshyyan, O., Maracine, M., Aw, T. Y., 2007. Neuronal apoptosis in neurodegeneration. *Antioxid Redox Signal* 9, 1059-1096.

Olanow, C. W., Schapira, A. H., 2013. Therapeutic prospects for Parkinson disease. *Ann Neurol* 74, 337-347.

Olivares, D., Deshpande, V. K., Shi, Y., Lahiri, D. K., Greig, N. H., Rogers, J. T., Huang, X., 2012. N-methyl D-aspartate (NMDA) receptor antagonists and memantine treatment for Alzheimer's disease, vascular dementia and Parkinson's disease. *Curr Alzheimer Res* 9, 746-758.

Owen, R. T., 2016. Memantine and donepezil: a fixed drug combination for the treatment of moderate to severe Alzheimer's dementia. *Drugs Today (Barc)* 52, 239-248.

Pang, H., Xue, W., Shi, A., Li, M., Li, Y., Cao, G., Yan, B., Dong, F., Xiao, W., He, G., Du, G., Hu, X., Cheng, G., 2016. Multiple-Ascending-Dose Pharmacokinetics and Safety Evaluation of Baicalein Chewable Tablets in Healthy Chinese Volunteers. *Clin Drug Investig* 36, 713-724.

Park, M. H., Hong, J. T., 2016. Roles of NF-kappaB in Cancer and Inflammatory Diseases and Their Therapeutic Approaches. *Cells* 5.

Perez, C. A., Wei, Y., Guo, M., 2009. Iron-binding and anti-Fenton properties of baicalein and baicalin. *J Inorg Biochem* 103, 326-332.

Pratico, D., Zhukareva, V., Yao, Y., Uryu, K., Funk, C. D., Lawson, J. A., Trojanowski, J. Q., Lee, V. M., 2004. 12/15-lipoxygenase is increased in Alzheimer's disease: possible involvement in brain oxidative stress. *Am J Pathol* 164, 1655-1662.

Radi, R., 2013. Peroxynitrite, a stealthy biological oxidant. *J Biol Chem* 288, 26464-26472.

Raven, E. P., Lu, P. H., Tishler, T. A., Heydari, P., Bartzokis, G., 2013. Increased iron levels and decreased tissue integrity in hippocampus of Alzheimer's disease detected in vivo with magnetic resonance imaging. *J Alzheimers Dis* 37, 127-136.

Reitz, C., Brayne, C., Mayeux, R., 2011. Epidemiology of Alzheimer disease. *Nat Rev Neurol* 7, 137-152.

Rohn, T. T., Head, E., 2009. Caspases as therapeutic targets in Alzheimer's disease: is it time to "cut" to the chase? *Int J Clin Exp Pathol* 2, 108-118.

Sadik, C. D., Sies, H., Schewe, T., 2003. Inhibition of 15-lipoxygenases by flavonoids: structure-activity relations and mode of action. *Biochem Pharmacol* 65, 773-781.

Sanders, L. H., Greenamyre, J. T., 2013. Oxidative damage to macromolecules in human Parkinson disease and the rotenone model. *Free Radic Biol Med* 62, 111-120.

Schneider, S. A., 2016. Neurodegeneration with Brain Iron Accumulation. *Curr Neurol Neurosci Rep* 16, 9.

Schulz, J. B., 2006. Anti-apoptotic gene therapy in Parkinson's disease. *J Neural Transm Suppl*, 467-476.

Sekiya, K., Okuda, H., 1982. Selective inhibition of platelet lipoxygenase by baicalein. *Biochem Biophys Res Commun* 105, 1090-1095.

Sharma, S., Singh, S., Sharma, V., Singh, V. P., Deshmukh, R., 2015. Neurobiology of L-DOPA induced dyskinesia and the novel therapeutic strategies. *Biomed Pharmacother* 70, 283-293.

Song, N., Jiang, H., Wang, J., Xie, J. X., 2007. Divalent metal transporter 1 up-regulation is involved in the 6-hydroxydopamine-induced ferrous iron influx. *J Neurosci Res* 85, 3118-3126.

Spencer, J. P., Vafeiadou, K., Williams, R. J., Vauzour, D., 2012. Neuroinflammation: modulation by flavonoids and mechanisms of action. *Mol Aspects Med* 33, 83-97.

Spillantini, M. G., Schmidt, M. L., Lee, V. M., Trojanowski, J. Q., Jakes, R., Goedert, M., 1997. Alpha-synuclein in Lewy bodies. *Nature* 388, 839-840.

Steinert, J. R., Chernova, T., Forsythe, I. D., 2010. Nitric oxide signaling in brain function, dysfunction, and dementia. *Neuroscientist* 16, 435-452.

Takeshima, M., Murata, M., Urasoe, N., Murakami, S., Miyazaki, I., Asanuma, M., Kita, T., 2011. Protective effects of baicalein against excess L-DOPA-induced dopamine quinone neurotoxicity. *Neurol Res* 33, 1050-1056.

Tannenberg, R. K., Scott, H. L., Westphalen, R. I., Dodd, P. R., 2004. The identification and characterization of excitotoxic nerve-endings in Alzheimer disease. *Curr Alzheimer Res* 1, 11-25.

Tsai, T. H., Liu, S. C., Tsai, P. L., Ho, L. K., Shum, A. Y., Chen, C. F., 2002. The effects of the cyclosporin A, a P-glycoprotein inhibitor, on the pharmacokinetics of baicalein in the rat: a microdialysis study. *Br J Pharmacol* 137, 1314-1320.

Uttara, B., Singh, A. V., Zamboni, P., Mahajan, R. T., 2009. Oxidative stress and neurodegenerative diseases: a review of upstream and downstream antioxidant therapeutic options. *Curr Neuropharmacol* 7, 65-74.

van Leyen, K., Kim, H. Y., Lee, S. R., Jin, G., Arai, K., Lo, E. H., 2006. Baicalein and 12/15-lipoxygenase in the ischemic brain. *Stroke* 37, 3014-3018.

Volles, M. J., Lansbury, P. T., Jr., 2003. Zeroing in on the pathogenic form of alpha-synuclein and its mechanism of neurotoxicity in Parkinson's disease. *Biochemistry* 42, 7871-7878.

Wang, C. X., Xie, G. B., Zhou, C. H., Zhang, X. S., Li, T., Xu, J. G., Li, N., Ding, K., Hang, C. H., Shi, J. X., Zhou, M. L., 2015a. Baicalein alleviates early brain injury after experimental subarachnoid hemorrhage in rats: possible involvement of TLR4/NF-kappaB-mediated inflammatory pathway. *Brain Res* 1594, 245-255.

Wang, H. F., Yu, J. T., Tang, S. W., Jiang, T., Tan, C. C., Meng, X. F., Wang, C., Tan, M. S., Tan, L., 2015b. Efficacy and safety of cholinesterase inhibitors and memantine in cognitive impairment in Parkinson's disease, Parkinson's disease dementia, and dementia with Lewy bodies: systematic review with meta-analysis and trial sequential analysis. *J Neurol Neurosurg Psychiatry* 86, 135-143.

Wang, J., Wang, Q., Wu, D., Yan, J., Wu, Y., Li, H., 2016a. Comparative studies on the interactions of baicalein and Al(III)-baicalein complex with human serum albumin. *Luminescence* 31, 54-62.

Wang, S. Y., Wang, H. H., Chi, C. W., Chen, C. F., Liao, J. F., 2004. Effects of baicalein on beta-amyloid peptide-(25-35)-induced amnesia in mice. *Eur J Pharmacol* 506, 55-61.

Wang, Z. Y., Liu, J. G., Li, H., Yang, H. M., 2016b. Pharmacological Effects of Active Components of Chinese Herbal Medicine in the Treatment of Alzheimer's Disease: A Review. *Am J Chin Med* 44, 1525-1541.

Weinreb, O., Mandel, S., Youdim, M. B., Amit, T., 2013. Targeting dysregulation of brain iron homeostasis in Parkinson's disease by iron chelators. *Free Radic Biol Med* 62, 52-64.

Wozniak, D., Drys, A., Matkowski, A., 2015. Antiradical and antioxidant activity of flavones from *Scutellariae baicalensis radix*. *Nat Prod Res* 29, 1567-1570.

Wu, P. H., Shen, Y. C., Wang, Y. H., Chi, C. W., Yen, J. C., 2006. Baicalein attenuates methamphetamine-induced loss of dopamine transporter in mouse striatum. *Toxicology* 226, 238-245.

Xie, Y., Song, X., Sun, X., Huang, J., Zhong, M., Lotze, M. T., Zeh, H. J., 3rd, Kang, R., Tang, D., 2016. Identification of baicalein as a ferroptosis inhibitor by natural product library screening. *Biochem Biophys Res Commun* 473, 775-780.

Yang, H., Zhuo, J. M., Chu, J., Chinnici, C., Pratico, D., 2010. Amelioration of the Alzheimer's disease phenotype by absence of 12/15-lipoxygenase. *Biol Psychiatry* 68, 922-929.

Yang, J., Wu, X., Yu, H., Liao, X., Teng, L., 2014. NMDA receptor-mediated neuroprotective effect of the *Scutellaria baicalensis* Georgi extract on the excitotoxic neuronal cell death in primary rat cortical cell cultures. *ScientificWorldJournal* 2014, 459549.

Yao, Y., Clark, C. M., Trojanowski, J. Q., Lee, V. M., Pratico, D., 2005. Elevation of 12/15 lipoxygenase products in AD and mild cognitive impairment. *Ann Neurol* 58, 623-626.

Yu, X., He, G. R., Sun, L., Lan, X., Shi, L. L., Xuan, Z. H., Du, G. H., 2012. Assessment of the treatment effect of baicalein on a model of Parkinsonian tremor and elucidation of the mechanism. *Life Sci* 91, 5-13.

Zhang, S., Ye, J., Dong, G., 2010. Neuroprotective effect of baicalein on hydrogen peroxide-mediated oxidative stress and mitochondrial dysfunction in PC12 cells. *J Mol Neurosci* 40, 311-320.

Zhang, S. Q., Obregon, D., Ehrhart, J., Deng, J., Tian, J., Hou, H., Giunta, B., Sawmiller, D., Tan, J., 2013. Baicalein reduces beta-amyloid and promotes nonamyloidogenic amyloid precursor protein processing in an Alzheimer's disease transgenic mouse model. *J Neurosci Res* 91, 1239-1246.

Zhang, X. Y., Zhang, L. H., Li, C. T., Chen, W. J., Zhao, J. B., Wei, E. Q., 2011. [5-lipoxygenase is involved in rotenone-induced injury in PC12 cells]. *Zhejiang Da Xue Xue Bao Yi Xue Ban* 40, 150-155.

Zhang, Y., Li, H., Zhao, Y., Gao, Z., 2006. Dietary supplementation of baicalin and quercetin attenuates iron overload induced mouse liver injury. *Eur J Pharmacol* 535, 263-269.

Zhao, Y., Li, H., Gao, Z., Xu, H., 2005. Effects of dietary baicalin supplementation on iron overload-induced mouse liver oxidative injury. *Eur J Pharmacol* 509, 195-200.

Zhou, L., Tan, S., Shan, Y. L., Wang, Y. G., Cai, W., Huang, X. H., Liao, X. Y., Li, H. Y., Zhang, L., Zhang, B. J., Lu, Z. Q., 2016. Baicalein improves behavioral dysfunction induced by Alzheimer's disease in rats. *Neuropsychiatr Dis Treat* 12, 3145-3152.

Zhu, M., Rajamani, S., Kaylor, J., Han, S., Zhou, F., Fink, A. L., 2004. The flavonoid baicalein inhibits fibrillation of alpha-synuclein and disaggregates existing fibrils. *J Biol Chem* 279, 26846-26857.

Zou, H., Long, J., Zhang, Q., Zhao, H., Bian, B., Wang, Y., Zhang, J., Wang, L., 2016. Induced cortical neurogenesis after focal cerebral ischemia--Three active components from Huang-Lian-Jie-Du Decoction. *J Ethnopharmacol* 178, 115-124.

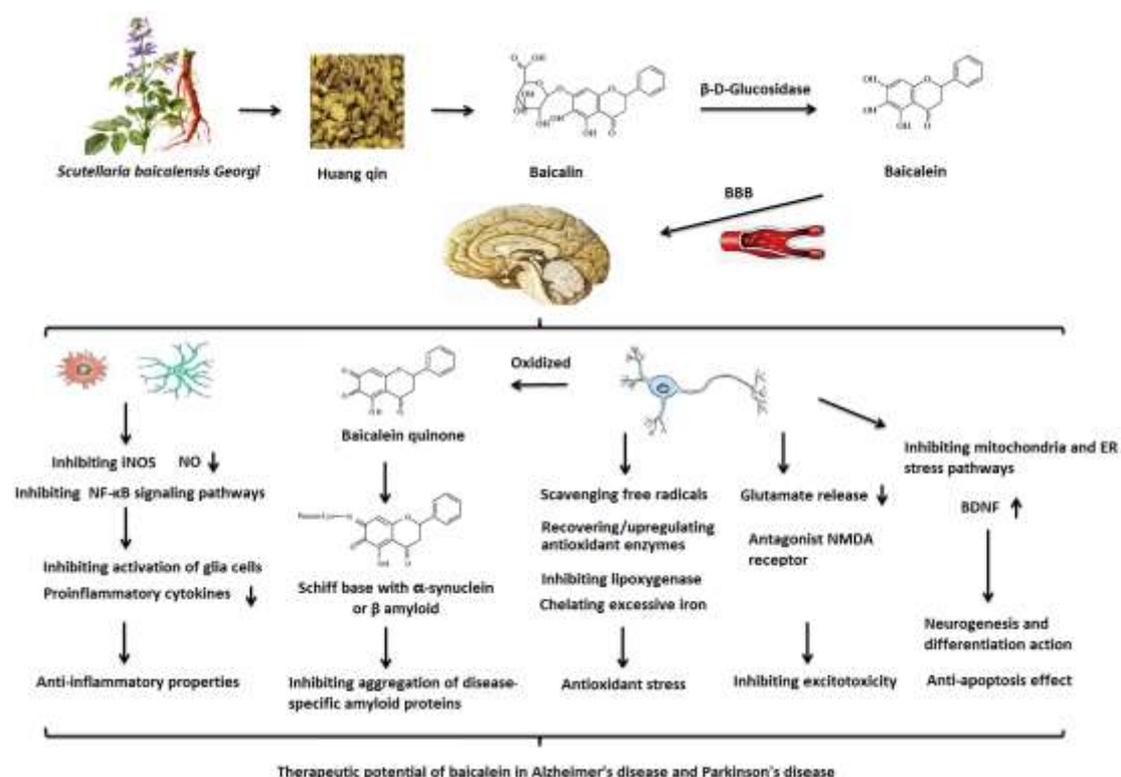


Fig. 1: The pharmacological effects of baicalein on neurodegenerative diseases

In Chinese traditional medicine, Huang qin usually refers to the dried root of *Scutellaria baicalensis Georgi*. Baicalin is a major hydrosoluble constituent in the decoction of Huang qin. After entering the human body, baicalin is hydrolyzed into baicalein by β -D-glucosidases, and baicalein can cross the BBB into the brain. Baicalein has antioxidant effects in the CNS by scavenging free radicals, by upregulating antioxidant enzymes, inhibiting lipoxygenase, and chelating excessive iron. Baicalein, especially baicalein quinone (its oxidized forms), might inhibit aggregation of disease-specific amyloid proteins ($A\beta$ and α -Syn) by formation of a Schiff base with $A\beta$ or α -Syn. The vicinal dihydroxyphenyl moieties of baicalein are required for quinone formation and attachment to amyloid proteins. Baicalein can reduce the chronic neuroinflammation response and production of proinflammatory cytokines by inhibiting the expression of iNOS and the production of NO, as well as by the inhibition of the NF- κ B signaling pathways in glial cells. Baicalein may inhibit excitotoxicity by reducing glutamate release and antagonizing NMDA receptor. Baicalein may promote neurogenesis and neural cells differentiation by upregulating BDNF signaling pathway. Baicalein also has anti-apoptotic effects by inhibiting mitochondrial and ER stress signaling pathways. Based on these multiple pharmacological effects, baicalein shows a therapeutic potential for treating AD and PD.