**Review**

**Therapeutic effects of tea polyphenols on Alzheimer's disease**

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**Abstract**

Alzheimer's disease (AD) is a great health threat to the elderly, and it also brings a great burden to society and families. Although free radicals have important biological functions. However, their imbalancesare associated with the pathobiology of many human diseases, including AD. Cumulative evidence suggests that tea drinking is associated with a reduced risk of AD. Many human epidemiological and animal studies have shown that tea polyphenols can promote health, reduce disease occurrence, and possibly prevent AD. Oxidative stress and inflammation of reactive oxygen species (ROS) and reactive nitrogen species (RNS) play a key role in neurodegenerative diseases AD, supporting the clinical application of tea polyphenols may have preventive neuroprotective effects on brain aging, and neurodegenerative diseases such as AD. This article reviews the involvement of oxidative stress in the pathogenesis of AD especially the interaction between oxidative stress and other key mechanisms of AD, and summarizes the protective and therapeutic effects of tea polyphenols on AD in cells, animal models and clinics. It hopes to provide insights into novel prevention and therapeutic strategies for AD.

**Key words**: Oxidative stress; AD;reactive oxygen species; reactive nitrogen species; free radicals; natural antioxidants; tea polyphenols; green tea polyphenols; theaflavins.

**Introduction**

With the aging of the population, the incidence of AD has increased significantly worldwide, unless effective prevention and/or treatment strategies are developed, the incidence of neurodegenerative diseases such as AD will continue to rise. Many drugs have been approved for the treatment of AD, however, their effects are not ideal, and they may cause various side effects [1]. Therefore, there is an urgent need for new AD prevention and treatment strategies with better efficacy and fewer side effects. Oxidative stress is one of the earliest changes in the pathogenesis of AD, which is related to the occurrence of AD leading to synaptic dysfunction and neuronal loss stimulating oxidative stress and resulting in AD, thereby forming a vicious circle, promoting the occurrence and development of AD. The prevention of oxidative stress may delay the occurrence of AD and the disease progression in AD patients. Many studies have shown that tea polyphenols can reduce A -induced neurotoxicities such as oxidative stress, mitochondrial dysfunction, and apoptosis. Since the pathogenesis of AD involves a variety of molecular events, the neuroprotective and effects of tea polyphenols also involve multiple mechanisms. In addition to reducing oxidative stress, preventing apoptosis, and promoting neurogenesis, tea polyphenols can inhibit the accumulation of A, restore calcium homeostasis, and also reduce transition metal overload in the brain. The anti-inflammatory properties of tea polyphenols are also related to their neuroprotective mechanisms. Most evidence suggests that tea polyphenol supplementation or tea consumption is beneficial in preventing the risk of AD without side effects [2]. Tea polyphenols not only have benefits for AD but also boost the patient's immunity, leading to better health, although they often contain multiple compounds or mixtures. For neurodegenerative diseases such as AD, tea polyphenols are suitable drugs because the pathogenesis of these diseases is complex and has many targets and pathways. These effects are more pronounced when clinical trials are used for long-term treatment. In this paper, the author will discuss the researches on the preventive and therapeutic effects of tea polyphenols including green tea polyphenols and theaflavins in neurodegenerative diseases AD, hoping to provide a new concept and approach for the prevention and treatment of AD [3, 4, 5; 6, 7].

**Oxidative stress and AD**

Oxidative stress plays an important role in the pathogenesis of AD. The brain is more susceptible to oxidative stress than other organs, and most components of neurons in AD may be oxidized due to mitochondrial dysfunction, elevated metal levels, inflammation, and β-amyloid (Aβ) peptides. The brain also is very rich in unsaturated fatty acids that are very easy to oxidize, and the concentration of antioxidants to prevent oxidation is very low. In addition, the brain's oxygen consumption is very large, which increases the chance of oxidative stress damage to the brain. Oxidative stress participates in the development of AD by promoting Aβ deposition, tau hyperphosphorylation, and subsequent synaptic and neuronal loss. The relationship between oxidative stress and AD suggests that oxidative stress is an essential part of the pathological process and that antioxidants may contribute to the treatment of AD. Many studies have shown that oxidative stress plays an important role in Aβ-induced cytotoxicity [8]. Aβ induces nerve cells to increase ROS and RNS production, leading to mitochondrial function degradation, reducing mitochondrial membrane voltage and activating caspase, ultimately leading to apoptosis of nerve cells [9]. Oxidative stress promotes the production of preamyloid Aβ. The expression of catalytic subunit progerin 1 by catabolic enzymes and  secretases accelerates the production of Aβ in precursor amyloid precursor protein (APP) and forms a vicious cycle [10, 11]. Studies have shown that genetic defects in the antioxidant system increase Aβ deposition in the brains of animals, while intake of antioxidants reduces Aβ deposition and improves the cognitive status of animals [12]. Extra free radicals can cause damage to the organism. Free radicals peroxidize membrane lipids [13] and oxidize proteins [14], leading to plasma membrane damage and cytoskeletal protein crosslinking. In addition, free radicals damage RNA [15], DNA [16, 17]. In the brain, high metabolic rates, low concentrations of glutathione and the antioxidant enzyme catalase, and a high proportion of polyunsaturated fatty acids make brain tissue particularly vulnerable to oxidative damage. Oxidative stress is a prooxidant/antioxidant homeostasis pro-oxidant-side imbalance with protein aggregation occurring in AD.

Oxidative stress is a state caused by an imbalance between oxidant production and the endogenous antioxidant defense system when oxidant production exceeds the clearance capacity of the antioxidant defense system in the AD brain. During oxidative stress, ROS and RNS react with proteins and lipids, disrupting their function, leading to progressive neuronal cell damage and ultimately brain cell death. The deletion of endothelial ROS and RNS in the human cerebral vascular endothelium and oxidative stress increase the expression of APP and enhance the production of Aβ peptide, indicating that the deletion of endothelial ROS and RNS contributes to AD pathology. Increased oxidative stress and ROS and RNS production in AD is based on protein oxidation, manifested by increased levels of protein carbonyl and 3-nitrotyrosine, as well as markers of oxidative damage to DNA and RNA, such as 8OHdG and 8-hydroxyguanosine prominent in AD brains [18,19]. Most studies have shown decreased activity of antioxidant enzymes in the brain of AD patients, but there is also an abnormal increase in SOD expression in brain neuropathy in AD patients, which may be an adaptive response to increased oxidative damage in these regions [20]. Differences in antioxidant enzyme expression and activity may reflect redistribution of antioxidant enzymes in neuropathy or enzyme inactivation caused by oxidation. Aβ peptides produce H2O2 through metal ion reduction and produce lipid peroxidation products. Oxidation of peptides leads to the formation of alkoxy radicals and hydroxylation of the peptide skeleton. An important oxidation process involves the irreversible nitrification of tyrosine residues by ONOO- produced by the reaction of ROS and RNS with NO. Understanding these mechanisms associated with ROS and RNS free radicals may provide new therapeutic targets for the prevention and treatment of AD.

Many studies have shown a close relationship between the destruction of metals and AD. One mechanism of Aβ accumulation may be due to disturbances in metal homeostasis in the AD brain. We have studied steady-state failure, oxidative stress, Aβ, APP, ferroregulatory protein (IRP), and divalent metal transporter 1 (DMT1) of iron and copper. It was found that iron-copper overload may be closely related to oxidative stress damage in the late stage of AD, and iron-copper deficiency may be closely related to early onset of AD. Destruction of metal homeostasis directly causes oxidative stress. Antioxidants can protect AD by regulating iron and copper homeostasis [7]. In SH-SY5Y cells overexpressing Swedish mutant human β-amyloid precursor protein (APPsw) and in strain CL2006, and Caenorhabditis elegans. An increase in iron content and oxidative stress levels was observed. Iron and calcium levels, ROS and RNS production within APPsw cells were significantly increased compared to control cells. The SOD activity and antioxidant levels of APPsw cells were significantly lower than those of control cells. In addition, iron treatment reduces cell viability and mitochondrial membrane potential, exacerbates oxidative stress damage, and releases Aβ1–40 from APPSW cells. Iron homeostasis disruption in APPsw cells may be associated with elevated expression of ferric transporter divalent metal transporter 1. Furthermore, nematodes with Aβ expression increase iron accumulation [21]. We also found that APPsw reduced iron content in neuroblastoma SH-SY5Y cells and increased oxidative stress. The activity of APPsw catalase was significantly lower than that of control cells. ROS and RNS production and calcium levels within APPsw cells were significantly increased compared to control cells. The mitochondrial membrane potential of APPsw cells was significantly lower than that of control cells. In addition, iron treatment reduces ROS and calcium levels in APPsw cells and improves cell viability. Iron deficiency in APPsw cells may be associated with the pathogenesis of AD [22]. Overall, these results suggest that the accumulation of Aβ in neuronal cells is associated with disruption of neuronal iron homeostasis and may be related to the pathogenesis of AD.

We investigated the effect of antioxidant nicotine on hippocampal and cortical metal homeostasis in APP mutant transgenic mice. After antioxidant nicotine treatment, the metal content of copper and zinc in age spots and nerve fibers was significantly reduced. The distribution density of copper and zinc in the CA1 region of the hippocampus also decreased. In addition, copper and zinc levels in nicotine-treated mice in age spots and nerve endings were significantly reduced by approximately 10-20% compared to the sucrose-treated control group. At the same time, the distribution of copper and zinc in the CA1 region of the hippocampus was observed. Copper is enriched in the pyramidal neuronal layer, while zinc is relatively dispersed. After treatment with antioxidant nicotine, the metal content of copper or zinc is significantly reduced, especially in the pyramidal neuronal layer [23]. ROS are mainly catalyzed by transition metals, and oxidative stress plays a key role in the pathogenesis of AD. We found increased expression levels of DMT1 IRE and DMT1 NEAGER in APPsw cells. Endogenous DMT1 is silenced by RNA interference, thereby reducing divalent ion influx, resulting in decreased APP expression and Aβ secretion. These findings suggest that DMT1 plays a key role in ion-mediated neuropathy of AD [24]. We also investigated the interaction and toxicity of Aβ1-42 and copper in the Aβ1-42 transgenic Caenorhabditis elegans model CL2006. The data show that CL2006 worms significantly worsen paralytic behavior after exposure to 10% copper ions. Exogenous copper treatment also partially alters the steady-state balance of zinc, manganese and iron. Aβ and copper-induced ROS production appears to be achieved by the SOD-1, PRDX-2, SKN-1, HSP-60, and HSP-16.2 genes [25].

The mitochondrial respiratory chain is the main site for intracellular production of ROS and RNS [26]. Oxidative modification of cellular components can disrupt membrane integrity, alter the function of essential proteins, lead to disruption of ionic homeostasis, mitochondrial dysfunction, and ultimately activation of apoptotic pathways and neuronal cell death [27]. In isolated mitochondria, Aβ can lead to mitochondrial membrane oxidative damage, disrupt lipid polarity and protein fluidity, and inhibit key enzymes of the mitochondrial respiratory chain, resulting in increased mitochondrial membrane permeability and cytochrome c release [28]. Data from transgenic mice consistently indicate that the presence of Aβ in mitochondria is associated with impaired mitochondrial metabolism and increased production of mitochondrial more ROS. We found that antioxidant nicotine inhibits MPP+ and calcium-induced hyperlitude mitochondrial swelling and intact mitochondrial release cytochrome C. The redox state within mitochondria is also maintained by antioxidant nicotine, which may be attributed to a weakening of mitochondrial permeability switching [29].

Numerous studies have linked oxidative stress to A-induced neurotoxicity. A treatment can increase levels of hydrogen peroxide and lipid peroxides. Possible role of soluble A oligomers as proximal neurotoxins in hippocampal neuronal cells, and oxidative stress involved in soluble A oligomer-induced synaptic damage and neuronal loss [30]. In various AD transgenic mouse models carrying APP and PS-1 mutants, increased H2O2 and NO production and increased oxidative modifications of proteins and lipids were associated with age-related A accumulation, further confirming that A promotes oxidative stress [31]. Increased oxidative stress in AD brains may initiate activation of a cascade of redox-sensitive cell signaling pathways, including JNK, which promotes the expression of BACE1 and PS1, ultimately increasing A production as well as deterioration of cognitive function. Since JNK is also associated with A-induced apoptosis, pharmacological inhibition of redox-sensitive signaling pathways such as JNK may reduce the accumulation of A and inhibit neuronal apoptosis [32].

The most common feature of AD is neurofibrillary tangles (NFTs) composed of the Tau protein. Neurofibrillary tangles and neuronal degeneration associated with increased oxidative stress. Oxidative stress-mediated abnormal Tau hyperphosphorylation dynamically, N-methyl-D-aspartate (NMDA) receptor (NMDAR) activation and Aβ toxicity alter synaptic function, which also associated with protein phosphatase inhibition and Tau hyperphosphorylation. NMDAR maintains neuronal excitability, Ca2+ influx, and memory formation through synaptic plasticity mechanisms. The synaptic redox stress mechanism associated with NMDARs alters expression [33].

**The structure and antioxidant properties of tea polyphenols**

Green tea mainly contains green tea polyphenols (GTP), and black tea mainly contains polymerized tea polyphenols theaflavins. Both are very good antioxidants, GTP have a stronger antioxidant capacity than theaflavins, but it is easy to oxidize at room temperature and difficult to preserve, theaflavins are relatively stable at room temperature and easy to preserve. Both have health implications for the body.

1. **The structure and antioxidant properties of green tea polyphenols**

The structure of GTP mainly contained in green tea is a general term for monomeric catechins, flavonoids and flavonols, anthocyanins and phenolic acid condensation phenolic acids. In addition to phenolic acids and condensed phenolic acids, other phenolic acids have the basic skeleton of the C6-C3-C6 configuration, which basically consists of two aromatic rings A and B, connected by a 3-carbon bridge, usually in the form of heterocyclic C. Four major catechins were isolated and identified in GTP: epicatechin (EC), epicatechin phenolate (EGC), epicatechin gallate (ECG), epicatechin gallate (EGCG) (fig.1).



Figure1. The structures of the green tea polyphenols and main theaflavins derivatives.

A large number of studies have shown that GTP can scavenge oxygen free radicals produced by different systems．The oxygen radicals produced by polymorphonuclear leukocytes (PMNs) are commonly used to test the scavenging effect of GTP on oxygen radicals produced by cellular systems. The results showed that GTP almost completely eliminated the oxygen free radicals produced by this system and were more effective than vitamin C and other antioxidants. In the superoxide anion radical system produced by the riboflavin/EDTA system under light, the scavenging rate of superoxide anion radicals by GTP is similar to that of vitamin C. The maximum clearance of EGCG to superoxide anion radicals is approximately 98%. We calculated the stoichiometric factor for the reaction of EGCG with oxygen radicals. The reaction rate constant k of EGCG and superoxide anion radicals was determined as 7.71x10-6 mMS-1, and the stoichiometric factor was 5.98. Studies have shown that each EGCG molecule captures and scavenges 6 superoxide anion radicals [34, 35, 36, 37].

Lipid peroxidation can eventually lead to cell metabolism, dysfunction, and even death. In the field of central nervous system, the relationship between oxygen radical production, lipid peroxidation and central nervous system damage has received more and more attention. We studied and compared the inhibitory effect of four tea polyphenol monomers on iron-induced lipid peroxidation of brain synaptosomes by spin capture. The results showed that EGCG, ECG, EGC and EC had inhibitory effects on lipid peroxidative damage to brain synaptosomes [38,39]．

We measured the inhibitory effect of four GTP monomers on TBA reactants formed during lipid peroxidation of brain synaptosomes. With the increase of the concentration of EGCG, ECG, EGC and EC, the formation amount of TBA reactants gradually decreased, and the inhibition rate gradually increased, which had a good dose-response relationship. The IC50 were 0.35mM/L, 0.24mM/L, 0.19mmol/L and 0.11mM/L, respectively, indicating that its inhibitory effect on iron-induced lipid peroxidative damage of brain synaptosomes was enhanced sequentially: EGCG>ECG>GTP>EGC>EC [38]. GTP with different structures not only have different antioxidant properties and scavenging effects on different free radicals, but also GTP isomers with different structures and phases may have different scavenging effects on free radicals. We found that three pairs of tea polyphenol isomers with the same structure were studied and found that they did have different antioxidant properties and different scavenging effects on different free radicals [39].

One study compared the scavenging ability of GTP on free radicals in mouse liver mitochondria. Result shown that GTP have a significant protective effect against mitochondrial oxidative damage. In addition, the antioxidant effect of GTP on zebrafish embryos was studied. It was found that zebrafish embryos showed significantly higher survival and heart rate in the face of oxidative stress after 1 day of GTP pretreatment. This is accompanied by decreased lipid and protein oxidation, as well as increased antioxidant defenses associated with markers of oxidative stress 4-hydroxynonenal (4-HNE), antihexyl lysine, dibromotyrosine, and 8-hydroxy2-deoxyguanosine (8-OHdG). [40].

The antioxidant effect of GTP and their oxidation products mainly refers to their role in scavenging free radicals. Free radicals can be produced by automatic oxidation of certain proteins, lipids, and low-molecular compounds, as well as redox of peroxides and certain metal ions. Free radicals in living organisms are in equilibrium between bio-generative systems and bio-protective systems. Once the balance is disrupted, it endangers the body and leads to disease. It needs exogenous antioxidants to scavenge free radicals and protect the normal functioning of the body. GTP are easily oxidize and provide protons, have the permeability of phenolic antioxidants, provide H+ bound to free radicals, can be reduced to inert compounds or more stable free radicals, thereby directly scavenging free radicals and avoiding oxidative damage. In addition, GTP can also act on related enzymes that produce free radicals, complexe with metal ions, thereby indirectly scavenging free radicals, thus playing a dual role in preventing and breaking chains, and their effects and mechanisms have been extensively studied [41]. GTP are rich in active hydroxyl groups and can bind to biological macromolecules such as proteins through hydrogen bonding, thus affecting many physiological processes. It can affect the enzymatic activity. For example, it enhances the activity of antioxidant enzymes and phase II enzymes and inhibits the activity of various enzymes, thereby exerting a significant inhibitory effect on many pathological processes.

1. **The structure and antioxidant properties of theaflavins**

Theaflavins are a class of substances formed by oxidative condensation of GTP that can be dissolved in ethyl acetate and appear yellow. Theaflavins not only play an important role in the soup color and taste of black tea, such as antioxidant, anticancer, anti-inflammatory, anti-cardiovascular and cerebrovascular diseases. It is the soul of black tea, and has a variety of health functions. Theaflavins are found in all fermented and semi-fermented tea categories, such as black tea, oolong tea, yellow tea, etc. Theaflavins are known as "soft gold". By consuming molecular oxygen catalyzed the oxidation of catechins to quinones, the oxidized quinones from cyclo-dihydroxylated catechins condense with the quinones from cyclotrihydroxylated catechins to produce different theaflavins. The structure of the main theaflavins are shown in figure 1.

　　Theaflavin has a strong scavenging ability against ROS such as superoxide radicals, hydroxyl radicals, singlet oxygen, and hydrogen peroxide. Moreover, theaflavins can protect cells from oxidative damage. It has a significant protective effect against H2O2-damaged cells, and the survival rate is increased. Compared with EGCG, theaflavins showed poor protection at concentrations of 2.87-11.5 lg/ml. However, at 0.72-1.43 lg/ml, theaflavins are more effective than EGCG. Protective effect of theaflavin derivatives against hydroxyl radical-induced DNA damage. In this study, the effect of theaflavins on hydroxyl radical-induced DNA oxidative damage was systematically analyzed. The results showed that theaflavins have good antioxidants for scavenging ROS and preventing hydroxyl radical-induced DNA damage in vitro compared to (EGCG) [42]. Theaflavins were found to regulate heterologous metabolic enzymes, oxidative stress and adduct formation in rats. Theaflavins can direct scavenge ROS and RNS, chelate transition metals and inhibit lipid, protein and DNA oxidation. In animal models and humans, theaflavins can reduce body weight, relieve metabolic syndrome, and prevent diabetes and cardiovascular disease. Theaflavins inhibit the occurrence of various cancers by regulating oxidative damage of biomolecules, endogenous antioxidants, and antioxidant gene transcription [43-46].

**Preventive and therapeutic effects of green tea polyphenols on AD**

Many studies have shown that GTP can reduce A-induced neurotoxicities such as oxidative stress, mitochondrial dysfunction, and apoptosis. Since the pathogenesis of AD involves multiple molecular events, the neuroprotective effects of GTP also appear to involve multiple mechanisms. In addition to reducing oxidative stress, preventing apoptosis, and promoting neurogenesis, GTP can inhibit the accumulation of A, restore calcium homeostasis, and reduce transition metal overload in the brain. The anti-inflammatory properties of GTP are also related to their neuroprotective mechanisms. Supplementation with GTP is beneficial for older adults and AD patients at risk of AD with no or minimal side effects. A lot of research on the role of GTP in the prevention of neurodegenerative diseases AD in cell and animal system [3, 4, 5, 47, 48, 49].

PubMed databases were searched for articles on tea and cognition registered. Cohort studies and three cross-sectional studies support the positive effects of green tea intake, supporting the hypothesis that GTP intake may reduce the risk of dementia, AD, mild cognitive impairment, or cognitive impairment [50]. A study conducted in Japan, among 1003 Japanese aged > 70 years, the prevalence of cognitive impairment was significantly reduced in those who drank more green tea [51]. A same trend has emerged in tea drinking among older Chinese [52]. Over the past 55 years or more, green tea consumption has been inversely correlated with the incidence of cognitive impairment [53]. A study included more than 7,000 older Chinese adults over the age of 80, including a 7-year follow-up. Tea drinkers had higher cognitive function than non-tea drinkers at all time points [54]. Among older people living in the Chinese community living in Singapore, tea drinking was also associated with better cognitive performance [55]. Other studies in Norway have demonstrated a similar association. One study showed that a significant correlation was observed only in men who drank green tea, but not in women [56]. A study conducted in the United States also showed sex differences [57]. When stratified by tea type, only green tea was found to correlate because green tea contains a higher EGCG content [58, 59].

**Preventive and therapeutic effect of theaflavin on AD**

　Epidemiological investigations report that drinking black tea can reduce the risk of dementia and depression. Theaflavins are known to have antioxidant and anti-inflammatory effects, contribute to the anti-aggregation and neuroprotective effects against Aβ [60]. Intraventricular LPS injections induced neuroinflammation, indicating impaired spatial memory and depressive-like behavior. Oral administration with theaflavins prevents these LPS-induced depressive changes. Theaflavins also prevent dendritic atrophy and spinal loss in the brain. Theaflavins have stronger anti-inflammatory effects than other polyphenols such as catechins, chlorogenic acid, and caffeic acid. It has been shown that theaflavins can inhibit neuroinflammation and prevent symptoms of inflammation-related brain diseases [61,62]. One study shownthat the neuromuscular blocking effect of theaflavins infusion on tetanus toxin in mouse phrenic diaphragm preparations, as well as the effect of binding this toxin to rat cerebral cortical synaptic membrane preparations. Also theaflavin mixed with tetanus toxin partially blocks the inhibition of the toxin. Theaflavins inhibit the specific binding of tetanus toxin to synaptic membrane preparations, and the effect is dose-dependent [63]. These findings suggest that the theaflavins protect neuron　against tetanus toxin's effects by binding to tetanus toxin.

Kinetics showed that theaflavin was a competitive inhibitor of porcine pancreatic α-amylase, while theaflavin was a hybrid inhibitor with competitive inhibitory properties. The competition inhibition constant of theaflavins was lower than that of catechins, indicating that theaflavin bound more closely to free porcine pancreatic α-amylase than to porcine pancreatic α-amylase-starch complexes. It was consistently found that the theaflavin increased the inhibitory effect on porcine pancreatic α-amylase by enhancing the association with the enzyme activation site [64]. Theaflavins have inhibitory effects on Aβ aggregation, but they exhibit different inhibitory abilities in different mechanistic steps of the Aβ aggregation pathway compared to catechins. Catechins only affect the later stages of aggregation, where catechins may bind to specific structures present in the aggregate. In contrast, theaflavins shows inhibition at each stage of polymerization, implying sequence-specific recognition [65].

Theaflavins protect cells from glutamate-induced oxidative stress, partially restore cell viability, and prevent apoptosis through bcl-2 and bax regulation, and attenuate intercellular ROS and RNS production. The protective effect is mediated by a variety of factors, including decrease in intracellular Ca2+ concentration, increased glutathione levels and associated enzyme activity. Theaflavins protect cells from glutamate-induced damage through indirect regulation of apoptosis-associated proteins and cellular energy enzymes. Theaflavins provide a potential therapeutic agent for glutamate accumulation and toxicity-related diseases AD [66].The results showed that theaflavins inhibited the formation of Aβ aggregates, protects nerves, contributes to the anti-aggregation and neuroprotective effect of Aβ [67].

**Molecular mechanism of tea polyphenols in the prevention and treatment of AD**

There have been many reports about the mechanism of AD prevention by tea drinking, most of which focus on the antioxidant effects of tea polyphenols (GTP and theaflavins), scavenging free radicals, iron chelating properties, inhibition of inflammation, regulation of cell survival/death genes, and induction of neuronal activity through mitochondrial function. Not only animal experiments, but also molecular experiments studies on the molecular mechanism of the effect of tea polyphenols on AD have been carried out in vivo and in vitro [68].

**1. Tea polyphenols have antioxidant effect and remove free radicals in AD**

We investigated the pathogenesis of iron in AD and the regulatory effect of the tea polyphenol on ROS free radicals. The results showed that tea polyphenols reduced the content of A ROS and RNS and intracellular calcium ions, and increased mitochondrial membrane potential in APPW cells [21,22]. Related effects of tea polyphenols, one study evaluated a similar role of tea polyphenols in streptozotocin-induced dementia in rats. One month after oral administration of tea polyphenols significant reductions in ROS and RNS levels production [69]. Tea polyphenols reduced the oxidized lipid peroxides, proteins, and DNA are increased in AD patients, and the antioxidant effects of tea polyphenols may contribute to the prevention of AD [70, 71, 72]. The tea polyphenol may have preventive and therapeutic effects on AD by scavenging ROS and RNS free radicals.

**2. Regulating effect of tea polyphenols on AD iron imbalance**

Iron imbalance in the body is closely related to the occurrence of AD, and important, iron overload can lead to nerve cell damage and AD-related diseases. In addition, if iron deficiency leads to anemia and hypoxia, it is also associated with AD. We investigated the pathogenic mechanism of iron in AD and the regulatory effect of the tea polyphenol on iron imbalance. The results showed that tea polyphenol reduced the oxidative damage of AD cells by complexing with iron, thereby protecting AD cells. The APPsw cell treated with tea polyphenols at different concentrations for 48 h to determine the iron content in the cellular iron pool, and the results showed a significant decrease in the iron pool of the APPsw cell [73]. These studies suggest that the tea polyphenol may reduce oxidative stress in peripheral and brain tissue by chelating excess iron in iron pools, have preventive and therapeutic effects on AD, and may inhibit behavioral changes associated with cognitive impairment.

**3. The inhibitory effect of tea polyphenols on A accumulation in AD brain**

Studies have shown that tea polyphenols may be used in the prevention and treatment of AD by inhibiting the accumulation of Ain AD brain. Long-term administration of tea polyphenol preparations or EGCG has been shown to improve spatial cognitive learning in rats and reduce cerebral amyloidosis in AD transgenic mice, respectively [74]. Tea polyphenol reduced the production of A in mouse neuron-like cells (N2a) transfected with APP and in primary neurons of mice. Tea polyphenols significantly promoted the lysis of APP α-C-terminal fragments and improved the soluble APPPα of N-terminal APP lysate. To validate these findings in vivo, treatment of excessive A-producing APPsw transgenic mice with tea polyphenol found the levels and plaques associated with promoting the α-secretase proteolysis pathway of non-amyloid production [75]. Tea polyphenols reduce A by promoting non-amyloid production α-secretase proteolytic pathway. EGCG reduced Ain mouse neuron-like cells (N2a) transfected with human APP and primary neurons from mice overexpressed by the Swedish mutant APP. Consistent with these observations, tea polyphenol was found to significantly promote cell division-C-terminal fragments of APP, and increased the solubility of N-terminal APP lysate [76].

EGCG treated SweAPP N2a cells, with elevated active-secretase, a-deintegratin metalloproteinase protein and active a-deintegratin metalloproteinase protein 10 and cleavage and sAPP-[77]. A-deintegratin metalloproteinase protein 10 activation is necessary for EGCG to promote non-amyloidosis APP treatment. Therefore, a-deintegratin metalloproteinase protein 10 is an important drug target for the treatment of cerebral A in AD. EGCG therapy has been reported to inhibit brain A deposition in Tg2576 mice. Even comparable effective doses of EGCG in humans may exceed clinical convenience, studies offer a solution through treatment of EGCG [5].

**4. Tea polyphenols have a preventive effect on AD by inhibiting AD-related diseases**

Although diabetes is considered to be two different diseases with AD, it has been recognized as an important factor in dementia risk. Drinking tea the oxidative stress and inflammation can be mitigated diabetes. In addition, the elimination of inflammation should reduce the incidence of immune reactions. The tea polyphenols provide potential benefits for reducing the risk of diabetes and AD by targeting common risk factors, such as obesity, hyperlipidemia, hypertension, cardiovascular disease, and stroke. Tea polyphenols have important antioxidant properties and natural properties that regulate intracellular neuronal signaling pathways. Diabetes, a disease caused by insulin resistance in the brain. AD and diabetes have a common underlying pathological process and important intervention pathways [78]. We found out that after 4 weeks of feeding tea polyphenols, the random blood glucose levels decreased fasting blood glucose levels about 30.4% (low concentration) and 2-hour blood glucose content about 51.2% (high concentration), 31.6% (low concentration) and 43.3% (high concentration), and fasting blood glucose content about low concentration 26.5% (low concentration) and 49.7% (high concentration). The mean random blood glucose and 2-hour blood glucose in the tea polyphenols group were 16.2 mm and 13.5 mm, respectively, both close to normal standards [79]. Our experiments have also proven that tea polyphenols can lose weight. Rats fed high fat gained significant weight compared to rats fed control rations. Weight loss was significantly reduced with feedings control and GTC. In the high-fat and control groups, feeding rats GTP resulted in significant weight loss within 30 days (approximately 9.4% and 6.3%, respectively, compared to the corresponding controls). This effect became more pronounced after 45 days of feeding GTC (about 11.8% and 8.2%) [80].Tea polyphenols offer potential benefits for reducing diabetes and AD risk by targeting common risk factors, including obesity, hyperlipidemia, hypertension, cardiovascular disease, and stroke.

**5. Tea polyphenols have a preventive effect on AD by inhibiting inflammation**

Studies have found that nerve damage caused by inflammation is an important factor in AD. Elimination of inflammation should reduce the incidence of immune reactions. The inflammatory process produces large amounts of ROS and RNS free radicals, resulting in oxidative stress damage. The oxidative stress and inflammation can be mitigated by tea polyphenols, thereby limiting their devastating effects on the patient's organism. Tea polyphenols also have anti-inflammatory properties, which is the basis of the mechanism of action in AD [82,83]. In a study conducted using injected mice of lipopolysaccharides, prior administration of tea polyphenols shown to prevent lipopolysaccharide-induced memory impairment and inhibit the increase in cytokines and inflammatory proteins [84]. Another in vitro study on BV-2 microglia showed that tea polyphenols inhibited the lipopolysaccharide-induced inflammation-related responses, including ROS and RNS production, cyclooxygenase-2 expression, and inducible nitric oxide synthase expression.

**6. Preventive effect of tea polyphenols on AD through signal transduction pathway**

The factors that cause AD activate multiple signaling molecules, such as protein kinase C (PKC) signaling pathways [85]. In vitro and in vivo studies have shown that low concentrations of tea polyphenols stimulate the production of soluble non-toxic Aβ in human neuroblastoma and PC12 cells. Oral administration of tea polyphenols for 2 weeks increases PKCα and ε in the hippocampus of mice compared to control animals [86]. The learning and memory ability of elderly Wister rats treated with tea polyphenols for 8 weeks was significantly improved compared with control young rats, and acetylcholinesterase activity was reduced in the brains of rats [87]. There was also a study in which scopolamine-induced amnesia could be reversed by giving mice a diet of tea polyphenols. Tea polyphenols significantly inhibit acetylcholinesterase activity [87]. It was also found that tea polyphenols dose-dependently attenuated A(25-35)-induced cell death, intracellular ROS levels and 8-oxodG formation, as well as p53, Bax, and caspase-3 expression, but upregulated Bcl-2. In addition, tea polyphenols prevented activation of A (25-25)-induced NF-B and ERK and p38 MAP kinase pathways [89].

The above discussion shows that the preventive and therapeutic effects of tea polyphenols on AD are multi-targeted, including antioxidant effects, scavenging of free radicals, iron chelating properties, inhibition of hydroxydopamine, signal transduction pathways, regulation of cell survival/death genes, and induction of neuronal activity through mitochondrial function. Studies have also shown that tea polyphenols can prevent Aβ and plaque formation and enhance cognitive function. Therefore, the use of tea polyphenols as multi-target drugs is of great significance for the prevention and treatment of AD.

**Conclusion**

Given the multietiological nature of AD, the current use of drugs targeting a single molecular target has limited ability to alter the course of the disease, so it has only partial benefits for AD patients, even side effects can occur. According to this concept, new strategies include the use of mixtures of multiple drugs and/or the development of a single molecule with two or more active neuroprotective neurorescue moieties, while manipulating multiple targets associated with AD pathology. Natural tea polyphenols (GTP and theaflavins) can enter the brain and have multifunctional activities such as metal chelating, free radical scavenging, anti-inflammatory and neuroprotective. (Mandel et al., 2007). In summary, the pathway and mechanism of tea polyphenols in the prevention and treatment of AD are shown in Figure 2.

Even tea polyphenols are used as clinical drugs for the treatment of hyperlipidemia, improve atherosclerosis and immunity. But it has not been used to treat AD [90]. In the future, more efforts should be made to evaluate the efficacy of tea polyphenols in the prevention and intervention of AD using clinical trials. Due to the complex composition of tea polyphenols, standardized formulations can be used for individual tea polyphenol component. To avoid differences between different trials, a well-characterized research group is also needed, and in order to determine effective doses and facilitate the evaluation of clinical trial efficacy. Separation and purification can further understand the protective mechanism of tea polyphenols against AD. Concerning the use of tea polyphenol therapies for AD, results from most clinical trials have been disappointing, despite the positive effects observed in epidemiological investigation and preclinical studies. The first thing need to know is that AD is a complex multifactorial disease. So far there is no drug can achieve satisfactory therapeutic results in the clinic. By the way, to achieve satisfactory therapeutic results in the clinic, it needs the tea polphenols must be considered in the context of species, time, place, level, and target. To achieve this situation requires a very long period of clinical trials, which in turn requires a lot of manpower, material resources and time. Even so, it is worth conducting more rigorous clinical trials in the future to find the satisfactory therapeutic results for AD, saving more patients for the benefit of humanity. Recently, A team proposed the 5R principle of antioxidants, "right species, right place, right time, right level, and right target", which is worth referring to in future about tea polyphenol therapies on AD [91].

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Figure 2. The pathways and mechanisms of tea polyphenols in the prevention and treatment of AD. GTP: Green tea polyphenols.

**References**

1. Eskelinen M.H., Ngandu T., Tuomilehto J., Soininen H., Kivipelto M. Midlife coffee and tea drinking and the risk of late-life dementia: A population-based CAIDE study. *J. Alzheimers Dis.*2009;16:85–91.
2. Nasim Rezaee, W.M.A.D. Binosha Fernando, Eugene Hone, Hamid R. Sohrabi, Stuart K. Johnson, Stuart Gunzburg, Ralph N. Martins [Potential of Sorghum Polyphenols to Prevent and Treat Alzheimer’s Disease: A Review Article](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8527926/).Front Aging Neurosci. 2021; 13: 729949.
3. Zhao B-L.(2005)Natural antioxidant for neurodegenerative diseases. Mol Neurobiol, 31, 283-293.
4. Zhao B-L. (2009). Natural Antioxidants Protect Neurons in Alzheimer’s Disease and Parkinson’s Disease. Neurochem Res 34:630–638，
5. Zhao B-L. 2012.Natural Antioxidant green tea polyphenols and health。ACTA BIOPHYSICA SINICA. 28,26-36.
6. Zhao Y. and Zhao B-L. (2012). Oxidative Stress, Natural antioxidants protect neurons against Alzheimer’s Disease. Front Biosci. 15, 454-461.
7. Zhao B-L and Wan L(2012). Imbalance of metal ion metabolism and early pathogenesis of Alzheimer disease. Progressin Biochemistry and Biophysics, 39，735-743.
8. Smith M., K. Hirai, K. Hsiao, M. Pappolla, P. Harris, S. Siedlak, M. Tabaton, and G. Perry(1998).Amyloid-beta deposition in Alzheimer transgenic mice is associated with oxidative stress. J Neurochem 70, (5) 2212-5.
9. Smith M. A., C. A. Rottkamp, A. Nunomura, A. K. Raina, and G. Perry (2000).Oxidative stress in Alzheimer's disease*.* *Biochim Biophys Acta* 1502,(1) 139-44
10. Oda A., A. Tamaoka, and W. (2009). Araki: Oxidative stress up-regulates presenilin 1 in lipid rafts in neuronal cells*.* *J Neurosci Res* 88,(5) 1137-45.
11. Ansari M. A. and Scheff S. W. (2010) Oxidative stress in the progression of Alzheimer disease in the frontal cortex. J Neuropathol Exp Neurol 69, (2) 155-6.
12. Dumont M., E. Wille, C. Stack, N. Calingasan, M. Beal, and M. Lin(2009).Reduction of oxidative stress, amyloid deposition, and memory deficit by manganese superoxide dismutase overexpression in a transgenic mouse model of Alzheimer's disease. Faseb J 23, (8) 2459-66.
13. Butterfield D. A. and J. Kanski(2001).Brain protein oxidation in age-related neurodegenerative disorders that are associated with aggregated proteins*.* *Mech Ageing Dev* 122,(9) 945-62.
14. Stadtman E.R. (1990) Metal ion-catalyzed oxidation of proteins: biochemical mechanism and biological consequences. Free Radical Biol. Med. 9, 315–325.
15. Nurk E., Refsum H., Drevon C.A., Tell G.S., Nygaard H.A., Engedal K., Smith A.D. (2009).Intake of flavonoid-rich wine, tea, and chocolate by elderly men and women is associated with better cognitive test performance. *J. Nutr.*139:120–127.
16. Gabbita S.P., Lovell M.A., and Markesbery W.R. (1998) Increased nuclear DNA oxidation in the brain in Alzheimer’s disease. J. Neurochem. 71, 2034–2040.
17. Mecocci P.L., MacGarvey U., and Beal M.F. (1994) Oxidative damage to mitochondrial DNA is increased in Alzheimer’s disease. Ann. Neurol. 36, 747–750.
18. Pratico D. and S. Sung (2004).Lipid peroxidation and oxidative imbalance: early functional events in Alzheimer's disease*.* *J Alzheimers Dis* 6,(2) 171-5.
19. Hareram Birla, Tarun Minocha, Gaurav Kumar, Anamika Misra, Sandeep Kumar Singh.[Role of Oxidative Stress and Metal Toxicity in the Progression of Alzheimer’s Disease](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7457422/).Curr Neuropharmacol. 2020; 18(7): 552–562.
20. Padurariu M., A. Ciobica, L. Hritcu, B. Stoica, W. Bild, and C. Stefanescu(2010).Changes of some oxidative stress markers in the serum of patients with mild cognitive impairment and Alzheimer's disease*.* *Neurosci Lett* 469,(1) 6-10.
21. Wan L, Guangjun Nie , Jie Zhang, Yunfeng Luo, Peng Zhang, Zhiyong Zhang, Baolu Zhao(2011) <Beta>-amyloid peptide increases levels of iron content and oxidative stress in human cell and C. elegans models of Alzheimer’s disease. F*ree Rad Biol Med* 50，,122–129.
22. Wan L, Nie G, Zhang J, Zhao B (2012) Overexpression of human wild-type amyloid- protein precursor decreases the iron content and increases the oxidative stress of neuroblastoma SH-SY5Y cells. Journal of Alzheimer’s Disease 30: 523-530.
23. Zhang J, Liu Q, Liu N-Q, Li F-L, Chen Q, Qin C, Zhu H， Huang Y-Y, He W, Zhao B-L.Nicotine reduces β-amyloidosis by regulating metal homeostasis. FASEB J 20，1212-1214，2006.
24. Zheng W, Na Xin，Zhi-Hong Chi, Bo-Lu Zhao, Jie Zhang, Jia-Yi Li, Zhan-You Wang.(2009). Divalent metal transporter 1 is involved in amyloid precursor protein processing and A generation.*FASEB J。* 23，4207-4217.
25. Luo Y–F, Zhang J, Liu N-Q, Luo Y, Zhao B-L.( 2011)Copper ions influence the toxicity of β-amyloid(1-42) in a concentration-dependent manner in a *Caenorhabditis elegans* model of Alzheimer’s disease. Sci China Life Sci,, 54: 1–8.
26. Caspersen C., N. Wang, J. Yao, A. Sosunov, X. Chen, J. W. Lustbader, H. W. Xu, D. Stern, G. McKhann, and S. D. Yan(2005).Mitochondrial Abeta: a potential focal point for neuronal metabolic dysfunction in Alzheimer's disease*.* *Faseb J* 19,(14) 2040-1.
27. Mattson M. P. (1997).Cellular actions of beta-amyloid precursor protein and its soluble and fibrillogenic derivatives*.* *Physiol Rev* 77,(4) 1081-132.
28. Casley C. S., L. Canevari, J. M. Land, J. B. Clark, and M. A. Sharpe(2002). Beta-amyloid inhibits integrated mitochondrial respiration and key enzyme activities*.* *J Neurochem* 80,(1) 91-100.
29. Xie Y-X, Bezard E, Zhao Bo-L.(2005). Unraveling the receptor- independent neuroprotective mechanism in mitochondria. J. Biol Chem. 37，32405-32412.
30. De Felice F. G., P. T. Velasco, M. P. Lambert, K. Viola, S. J. Fernandez, S. T. Ferreira, and W. L. Klein (2007).Abeta oligomers induce neuronal oxidative stress through an N-methyl-D-aspartate receptor-dependent mechanism that is blocked by the Alzheimer drug memantine*.* *J Biol Chem* 282,(15) 11590-601.
31. Manczak M., T. S. Anekonda, E. Henson, B. S. Park, J. Quinn, and P. H. Reddy(2006). Mitochondria are a direct site of A beta accumulation in Alzheimer's disease neurons: implications for free radical generation and oxidative damage in disease progression*.* *Hum Mol Genet* 15,(9) 1437-49.
32. Yao M., T. V. Nguyen, and C. J. Pike(2005).Beta-amyloid-induced neuronal apoptosis involves c-Jun N-terminal kinase-dependent downregulation of Bcl-w*.* *J Neurosci* 25,(5) 1149-58.
33. Christina M. Moloney, Val J. Lowe, Melissa E. Murray. [Visualization of neurofibrillary tangle maturity in Alzheimer's disease: A clinicopathologic perspective for biomarker research](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8478697/).Alzheimers Dement. 2021; 17(9): 1554–1574.
34. Shen S, Yang X, Yang F, Zhao B-L, Xin W: (1993)synergistic enhancement effect of catechins on antioxidation. Tea science, 13 (2), 141-146
35. Yang FJ, Zhao,BL,Xin,WJ.(1992): ESR spin trapping method was used to study the effect of liposomes treated with smoking smoke on O2 - production by rat granulocytes. Journal of Biophysics，8,1992,659-663.
36. Yang FJ, Zhao,BL, Ren X-J, Xin,WJ.(1993): ESR study of tea polyphenols inhibiting lipid free radical production in rat liver microsomes stimulated by smoking gaseous substances. Journal of Biophysics，9, 468-471.
37. Zhao BL, Li,XJ, He, RG, Cheng, SJ, Xin,WJ: (1989)Scavenging effect of extracts of green tea and natural antioxidants on active oxygen radicals. Cell Biophys，14, 175-184.
38. Guo Q., Zhao B.-L., Li M.-F., Shen S.-R., and Xin W.-J. Studies on protective mechanisms of four components of green tea polyphenols (GTP) against lipid peroxidation in synaptosomes. Biochem. Biophys. Acta 1996,1304, 210–222.
39. Guo Q, Zhao B-L, Hou J-W, Xin W-J. ESR study on the structure-antioxidant activiity relationship of tea catechins and their epimers. *Bichim Biophys Acta* 1999,1427,13-23
40. Gao T, Shi Y, Xue Y, Yan F, Huang D, Wu Y, Weng Z.(2020)[Polyphenol extract from superheated steam processed tea waste attenuates the oxidative damage in vivo and in vitro.](https://pubmed.ncbi.nlm.nih.gov/31693210/) J Food Biochem. 44(1):e13096.
41. Ying L, Kong D, Gao Y, et al. In vitro antioxidant activity of phenolic-enriched extracts from Zhangping Narcissus tea cake and their inhibition on growth and metastatic capacity of 4T1 murine breast cancer cells[J]. Journal of Zhejiang University-SCIENCE B, 2018, 19(3): 199-210.
42. Yuan-yuan Wu, Wei Li, Yi Xu, En-hui Jin, You-ying Tu. [Evaluation of the antioxidant effects of four main theaflavin derivatives through chemiluminescence and DNA damage analyses](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3167908/).J Zhejiang Univ Sci B. 2011 Sep; 12(9): 744–751.
43. Huang F, Zheng X, Ma X, Jiang R, Zhou W, Zhou S, Zhang Y, Lei S, Wang S, Kuang J, Han X, Wei M, You Y, Li M, Li Y, Liang D, Liu J, Chen T, Yan C, Wei R, Rajani C, Shen C, Xie G, Bian Z, Li H, Zhao A, Jia W.( 2019)[Theabrownin from Pu-erh tea attenuates hypercholesterolemia via modulation of gut microbiota and bile acid metabolism.](https://pubmed.ncbi.nlm.nih.gov/31672964/)Nat Commun. 10(1):4971.
44. Piotr Olcha, Anna Winiarska-Mieczan, Małgorzata Kwiecień, Łukasz Nowakowski, Andrzej Miturski, Andrzej Semczuk, Bożena Kiczorowska, Krzysztof Gałczyński
45. [Antioxidative, Anti-Inflammatory, Anti-Obesogenic, and Antidiabetic Properties of Tea Polyphenols—The Positive Impact of Regular Tea Consumption as an Element of Prophylaxis and Pharmacotherapy Support in Endometrial Cancer](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC9224362/).Int J Mol Sci. 2022; 23(12): 6703.
46. [Murugan RS, Uchida K, Hara Y, Nagini S.(2008)Black tea polyphenols modulate xenobiotic-metabolizing enzymes, oxidative stress and adduct formation in a rat hepatocarcinogenesis model.](https://pubmed.ncbi.nlm.nih.gov/18985486/)Free Radic Res. 2008 Oct;42(10):873-84.
47. Farida El Gaamouch, Kalena Liu, Hsiao-yun Lin, Clark Wu, Jun Wang. [Development of grape polyphenols as multi-targeting strategies for Alzheimer’s disease](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8178246/) Neurochem Int.  2021; 147: 105046.
48. Manuela Leri, Maria Scuto, Maria Laura Ontario, Vittorio Calabrese, Edward J. Calabrese, Monica Bucciantini, Massimo Stefani. [Healthy Effects of Plant Polyphenols: Molecular Mechanisms](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7072974/).Int J Mol Sci. 2020; 21(4): 1250. Published online 2020 Feb 13.
49. Johant Lakey-Beitia, Andrea M. Burillo, Giovanni La Penna, Muralidhar L. Hegde, K.S. Rao.[Polyphenols as Potential Metal Chelation Compounds Against Alzheimer’s Disease](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7809605/).J Alzheimers Dis. 2021; 82(Suppl 1): S335–S357.
50. Kakuda T., A. Nozawa, A. Sugimoto, and H. Niino (2002). Inhibition by theanine of binding of [3H]AMPA, [3H]kainate, and [3H]MDL 105,519 to glutamate receptors*.* *Biosci Biotechnol Biochem* 66,(12) 2683-6.
51. Kuriyama S., Hozawa A., Ohmori K., Shimazu T., Matsui T., Ebihara S., Awata S., Nagatomi R., Arai H., Tsuji I. (2006).Green tea consumption and cognitive function: A cross-sectional study from the Tsurugaya Project 1. *Am. J. Clin. Nutr.*83:355–361.
52. Gu Y.J., He C.H., Li S., Zhang S.Y., Duan S.Y., Sun H.P., Shen Y.P., Xu Y., Yin J.Y., Pan C.W. (2017).Tea consumption is associated with cognitive impairment in older Chinese adults. *Aging Ment. Health.*1–7.
53. Ng T.P., Feng L., Niti M., Kua E.H., Yap K.B. (2008).Tea consumption and cognitive impairment and decline in older Chinese adults. *Am. J. Clin. Nutr.*88:224–231.
54. Feng L., Gwee X., Kua E.H., Ng T.P.) 2010). Cognitive function and tea consumption in community dwelling older Chinese in Singapore. *J. Nutr. Health Aging.*14:433–438.
55. Feng L., Li J., Ng T.P., Lee T.S., Kua E.H., Zeng Y(2012). Tea drinking and cognitive function in oldest-old Chinese. *J. Nutr. Health Aging.*16:754–758.
56. Huang C.Q., Dong B.R., Zhang Y.L., Wu H.M., Liu Q.X. (2009). Association of cognitive impairment with smoking, alcohol consumption, tea consumption, and exercise among Chinese nonagenarians/centenarians. *Cogn. Behav. Neurol.* 22:190–196.
57. Arab L., Biggs M.L., O’Meara E.S.,et al. (2011)Gender differences in tea, coffee, and cognitive decline in the elderly: The Cardiovascular Health Study. *J. Alzheimers Dis.* 27:553–566.
58. Liu Q, Jie Zhang, Hua Zhu, Chuan Qin, Qi Chen，Baolu Zhao.( 2007) Dissecting the Signalling Pathway of Nicotine-Mediated Neuroprotection in a Mouse Alzheimer Disease Model. FASEB J 21:61-73.
59. Zhao B-Lu (2020). The pros and cons of drinking tea. Traditional Medicine and Modern Medicine 3, 3, 163–174.
60. Xinlei Li, Scott D. Smid, Jun Lin, Zhihong Gong, Si Chen, Fangning You, Yan Zhang, Zhilong Hao, Hongzheng Lin, Xiaomin Yu, Xinyi Jin.[Neuroprotective and Anti-Amyloid β Effect and Main Chemical Profiles of White Tea: Comparison Against Green, Oolong and Black Tea](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6571989/).Molecules. 2019 May; 24(10): 1926.
61. Yasuhisa Ano, Rena Ohya, Masahiro Kita, Yoshimasa Taniguchi, Keiji Kondo.[Theaflavins Improve Memory Impairment and Depression-Like Behavior by Regulating Microglial Activation](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6384870/).Molecules. 2019; 24(3): 467.
62. Anna Winiarska-Mieczan, Ewa Tomaszewska, Karolina Jachimowicz. [Antioxidant, Anti-Inflammatory, and Immunomodulatory Properties of Tea—The Positive Impact of Tea Consumption on Patients with Autoimmune Diabetes](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8625657/).Nutrients. 2021 Nov; 13(11): 3972.
63. Satoh E, Ishii T, Shimizu Y, Sawamura S, Nishimura M. (2002)[A mechanism of the thearubigin fraction of black tea (Camellia sinensis) extract protecting against the effect of tetanus toxin.](https://pubmed.ncbi.nlm.nih.gov/12533914/)J Toxicol Sci. 27(5):441-7.
64. Sun L.J., Warren F.J., Netzel G., Gidley M.J. 3 or 3′-Galloyl substitution plays an important role in association of catechins and theaflavins with porcine pancreatic α-amylase: The kinetics of inhibition of α-amylase by tea polyphenols. *J. Funct. Foods.*2016;26:144–156.
65. Chastain S.E., Moss M. Green and black tea polyphenols mechanistically inhibit the aggregation of amyloid-β in Alzheimer’s disease. *Biophys. J.*2015;108:357a.
66. Jinting He, Lei Xu, Le Yang, Caixia Sun. [Anti-oxidative effects of catechins and theaflavins on glutamate-induced HT22 cell damage](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC9066190/).RSC Adv. 2019 Jul 5; 9(37): 21418–21428.
67. Xinlei Li, Scott D. Smid, Jun Lin, Zhihong Gong, Si Chen, Fangning You, Yan Zhang, Zhilong Hao, Hongzheng Lin, Xiaomin Yu, Xinyi Jin. [Neuroprotective and Anti-Amyloid β Effect and Main Chemical Profiles of White Tea: Comparison Against Green, Oolong and Black Tea](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6571989/).Molecules. 2019 May; 24(10): 1926.
68. Ali B., Jamal Q.M., Shams S., et al. In silico analysis of green tea polyphenols as inhibitors of AChE and BChE enzymes in Alzheimer’s disease treatment. *CNS Neurol. Disord. Drug Targets.*2016;15:624–628.
69. Biasibetti R., Tramontina A.C., Costa A.P., Dutra M.F., Quincozes-Santos A., Nardin P., Bernardi C.L., Wartchow K.M., Lunardi P.S., Goncalves C.A. Green tea (−)epigallocatechin-3-gallate reverses oxidative stress and reduces acetylcholinesterase activity in a streptozotocin-induced model of dementia. *Behav. Brain Res.*2013;236:186–193.
70. Praticò D. (2008) Evidence of oxidative stress in Alzheimer’s disease brain and antioxidant therapy. *Ann. N. Y. Acad. Sci.*1147:70–78.
71. Kaur T., Pathak C.M., Pandhi P., Khanduja K.L.( 2008) Effects of green tea extract on learning, memory, behavior and acetylcholinesterase activity in young and old male rats. *Brain Cogn.*67:25–30.
72. Sang S., Tian S., Wang H., Stark R.E., Rosen R.T., Yang C.S., Ho C.T.( 2003). Chemical studies of the antioxidant mechanism of tea catechins: Radical reaction products of epicatechin with peroxyl radicals. *Bioorg. Med. Chem.*11:3371–3378.
73. Ward R., Zucca F.A., Duyn J.H., Crichton R.R., Zecca L. (2014)The role of iron in brain ageing and neurodegenerative disorders. *Lancet Neurol.*13:1045–1060.
74. Haque A.M., Hashimoto M., Katakura M., Hara Y., Shido O. Green tea catechins prevent cognitive deficits caused by Aβ1–40 in rats. *J. Nutr. Biochem.*2008, 19:619–626.
75. Liu M, Chen F, Sha L, Wang S, Lin Tao, et al..[(−)-Epigallocatechin-3-Gallate Ameliorates Learning and Memory Deficits by Adjusting the Balance of TrkA/p75](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4012162/)[NTR](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4012162/)[Signaling in APP/PS1 Transgenic Mice](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4012162/).Mol Neurobiol. 2014; 49(3): 1350–1363.
76. Bae N, Byeon SE, Song J, Lee S-J, et al.. [Knock-down of protein L-isoaspartyl O-methyltransferase increases β-amyloid production by decreasing ADAM10 and ADAM17 levels](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4002779/).Acta Pharmacol Sin. 2011 Mar; 32(3): 288–294.
77. Ashley Payne, Samuel Nahashon, Equar Taka, Getinet M. Adinew, Karam F. A. Soliman.[Epigallocatechin-3-Gallate (EGCG): New Therapeutic Perspectives for Neuroprotection, Aging, and Neuroinflammation for the Modern Age](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8945730/).Biomolecules. 2022 Mar; 12(3): 371.
78. Fernando WMADB, Somaratne G, Goozee KG,et al(2017). [Diabetes and Alzheimer's Disease: Can Tea Phytochemicals Play a Role in Prevention?](https://pubmed.ncbi.nlm.nih.gov/28582855/) J Alzheimers Dis.59(2):481-501.
79. Yan J, Zhao Y, Suo S, Liu Y, Zhao B-L(2012)Green tea catechins ameliorate adipose insulin resistance by improving oxidative stress. Free Radical Biology & Medicine 52 , 1648–1657.
80. Yan J, Zhao Y, Zhao B(2013)Green tea catechins prevent obesity through modulation of peroxisome proliferator-activated receptors。Sci China，Life Sciences，56，804–810.
81. Lee Y.J., Choi D.Y., Yun Y.P., Han S.B., Oh K.W., Hong J.T.( 2013) Epigallocatechin-3-gallate prevents systemic inflammation-induced memory deficiency and amyloidogenesis via its anti-neuroinflammatory properties. *J. Nutr. Biochem.*24:298–310.
82. Winiarska-Mieczan A, Tomaszewska E, Jachimowicz K. [Antioxidant, Anti-Inflammatory, and Immunomodulatory Properties of Tea—The Positive Impact of Tea Consumption on Patients with Autoimmune Diabetes](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8625657/).Nutrients. 2021 Nov; 13(11): 3972.
83. Morales I., Guzman-Martinez L., Cerda-Troncoso C., Farias G.A., Maccioni R.B. (2014). Neuroinflammation in the pathogenesis of Alzheimer’s disease. A rational framework for the search of novel therapeutic approaches. *Front. Cell. Neurosci.*8:112.
84. Helieh S. Oz. [Chronic Inflammatory Diseases and Green Tea Polyphenols](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5490540/).Nutrients. 2017 Jun; 9(6): 561.
85. Afzal O, Hassan Dalhat M, Altamimi AS A., Rasool R et al..[Green Tea Catechins Attenuate Neurodegenerative Diseases and Cognitive Deficits](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC9655201/).Molecules. 2022 Nov; 27(21): 7604.
86. Levites Y., Amit T., Mandel S., Youdim M.B. (2003).Neuroprotection and neurorescue against Abeta toxicity and PKC-dependent release of nonamyloidogenic soluble precursor protein by green tea polyphenol (−)-epigallocatechin-3-gallate. *FASEB J.*17:952–954.
87. Kaur T., Pathak C.M., Pandhi P., Khanduja K.L.( 2008) Effects of green tea extract on learning, memory, behavior and acetylcholinesterase activity in young and old male rats. *Brain Cogn.*67:25–30.
88. Kim H.K., Kim M., Kim S., Kim M., Chung J.H. (2004).Effects of green tea polyphenol on cognitive and acetylcholinesterase activities. *Biosci. Biotechnol. Biochem.*68:1977–1979.
89. Lee SY, Lee JW, Lee H, Yoo HS, Yun YP, Oh KW, Ha TY, Hong JT.[Inhibitory effect of green tea extract on beta-amyloid-induced PC12 cell death by inhibition of the activation of NF-kappaB and ERK/p38 MAP kinase pathway through antioxidant mechanisms.](https://pubmed.ncbi.nlm.nih.gov/16153742/) Brain Res Mol Brain Res. 2005 Oct 31;140(1-2):45-54.
90. Jia B-Q,Lu Y-L. Diagnosis and therapy handbook. Beijing Medical University Press.2001. 367.
91. Meng J, Lv Z, Zhang Y, Wang Y, Qiao X, Sun C, Chen Y, Guo M, Han W, Ye A, Xie T, Chu B, Shi C, Yang S, Chen C.[Precision Redox: The Key for Antioxidant Pharmacology.](https://pubmed.ncbi.nlm.nih.gov/33270507/)Antioxid Redox Signal. 2021;34(14):1069-1082.