

Natural Compounds with Anti-BACE1 Activity as Promising Therapeutic Drugs for Treating Alzheimer's Disease

Authors

Mehjabeen Naushad^{1*}, Siva Sundara Kumar Durairajan^{1*}, Amal Kanti Bera², Sanjib Senapati², Min Li³

Affiliations

- 1 Department of Microbiology, School of Life Sciences, Central University of Tamil Nadu, Tiruvarur, India
- 2 Department of Biotechnology, Indian Institute of Technology Madras, Chennai, India
- 3 Neuroscience Research Laboratory, Mr. & Mrs. Ko Chi-Ming Centre for Parkinson's Disease Research, School of Chinese Medicine, Hong Kong Baptist University, Hong Kong SAR, China

Key words

Alzheimer's disease, BACE1, BACE1 inhibitors, flavonoids, phenolics, alkaloids, terpenes

received April 8, 2019

revised September 24, 2019

accepted September 26, 2019

Bibliography

DOI <https://doi.org/10.1055/a-1019-9819>

Published online October 16, 2019 | *Planta Med* 2019; 85: 1316–1325 © Georg Thieme Verlag KG Stuttgart · New York | ISSN 0032-0943

Correspondence

Dr. D. Siva Sundara Kumar
Associate Professor and Head, Division of Mycobiology and Neurodegenerative Disease Research, Department of Microbiology, School of Life Sciences, Central University of Tamil Nadu
Tiruvarur, Pin 610005, India
Phone: + 91 43 66 27 72 07
d.sivasundarakumar@cutn.ac.in

Correspondence

Prof. Min Li, Professor
Neuroscience Research Laboratory, Mr. & Mrs. Ko Chi-Ming Centre for Parkinson's Disease Research, School of Chinese Medicine, Hong Kong Baptist University
Kowloon Tong, Hong Kong SAR, China
Phone: + 85 2 34 11 29 19, Fax: + 85 2 34 11 24 61
limin@hkbu.edu.hk

ABSTRACT

Alzheimer's disease is a neurodegenerative disease that leads to irreversible neuronal damage. Senile plaques, composed of amyloid beta peptide, is the principal abnormal characteristic of the disease. Among the factors involved, the secretase enzymes, namely, α secretase, beta-site amyloid precursor protein-cleaving enzyme, β secretase, and γ secretase, hold consequential importance. Beta-site amyloid precursor protein-cleaving enzyme 1 is considered to be the rate-limiting factor in the production of amyloid beta peptide. Research supporting the concept of inhibition of beta-site amyloid precursor protein-cleaving enzyme activity as one of the effective therapeutic targets in the mitigation of Alzheimer's disease is well accepted. The identification of natural compounds, such as β -amyloid precursor protein-selective beta-site amyloid precursor protein-cleaving enzyme inhibitors, and the idea of compartmentalisation of the beta-site amyloid precursor protein-cleaving enzyme 1 action have caused a dire need to closely examine the natural compounds and their effectiveness in the disease mitigation. Many natural compounds have been reported to effectively modulate beta-site amyloid precursor protein-cleaving enzyme 1. At lower doses, compounds like 2,2',4'-trihydroxychalcone acid, quercetin, and myricetin have been shown to effectively reduce beta-site amyloid precursor protein-cleaving enzyme 1 activity. The currently used five drugs that are marketed and used for the management of Alzheimer's disease have an increased risk of toxicity and restricted therapeutic efficiency, hence, the search for new anti-Alzheimer's disease drugs is of primary concern. A variety of natural compounds having pure pharmacological moieties showing multitargeting activity and others exhibiting specific beta-site amyloid precursor protein-cleaving enzyme 1 inhibition as discussed below have superior biosafety. Many of these compounds, which are isolated from medicinal herbs and marine flora, have been long used for the treatment of various ailments since ancient times in the Chinese and Ayurvedic medical systems. The aim of this article is to review the available data on the selected natural compounds, giving emphasis to the inhibition of beta-site amyloid precursor protein-cleaving enzyme 1 activity as a mode of Alzheimer's disease treatment.

* These two authors contributed equally to this work.

Introduction

Alzheimer's disease (AD) is a prevalent neurodegenerative proteopathy that is currently incurable [1]. Patients suffering from the disease are characterised by reduced cognitive function, progressive deterioration of the memory and neuronal damage, and changes in mood and behaviour [2]. Living with this disease can be debilitating and ultimately fatal. The presence of amyloid plaques (composed mainly of $A\beta$ peptides) and neurofibrillary tangles (aggregates of hyperphosphorylated tau protein) is the main pathological characteristic of AD [3]. This is accompanied by microglial proliferation, neuropil threads, and associated astrogliosis [4]. These pathological processes lead to the deterioration of the brain and its activities.

Sequential processing of the β -amyloid precursor protein (β APP) by β secretase and γ secretase leads to the formation of the $A\beta$ peptides [5]. The γ secretase cleaves C99 after the action of beta-site amyloid precursor protein-cleaving enzyme 1 (BACE1), leading to the formation of $A\beta_{40}$ and $A\beta_{42}$ in most cases [6]. β Secretase or BACE1 also known as memapsin 2, and Asp 2) holds one of the pivotal roles in AD disease pathogenesis as its protein levels and/or enzyme activity are observed to be significantly elevated in the AD brain compared to the slight elevation seen in the normal aging brain [7–9]. Hence, BACE1 is of specific interest as a drug target since its inhibition is considered a potential treatment, if not the cure. Similar to BACE1, BACE2 is a type I transmembrane protein and is a close homologue [10, 11]. BACE2 has been reported to cleave β APP like BACE1, but the fragments produced have so far not been observed in senile plaques. Hence, its role in AD is questionable [11]. BACE2 is expressed highly in the peripheral tissues and in oligodendrocytes, astrocytes, and neuronal subsets [12], whereas BACE1 is highly concentrated only in the brain [10]. BACE1 is considered to be the rate-limiting factor involved in the formation of $A\beta$, and as such is a suitable target for drugs [13]. A detailed illustration of the β APP processing by the secretases is depicted in ► Fig. 1.

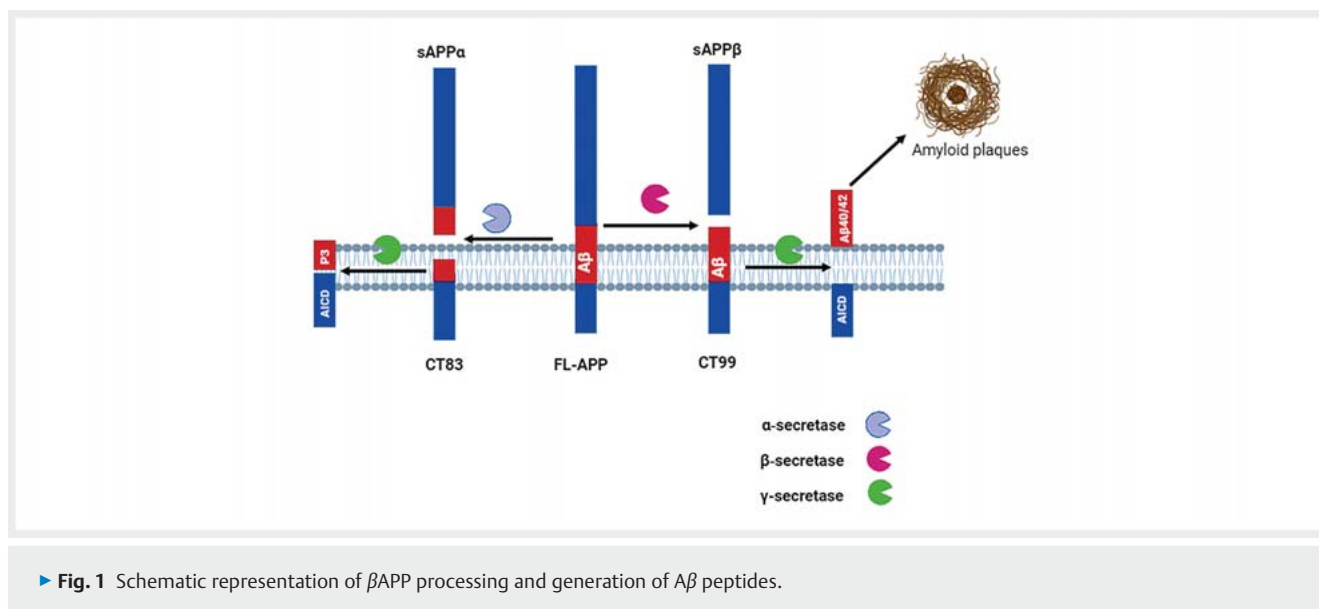
Inhibition of BACE1 activity can be an effective therapeutic target for treating AD. Complete BACE1 inhibition was considered to be desirable after Roberds et al. and other independent studies reported that BACE1^{-/-} mice failed to exhibit excessive $A\beta$ deposition. Luo et al. also evidenced that BACE1 knockout fully prevents $A\beta$ production while displaying a normal phenotype in mice [14, 15]. This gave rise to the idea that therapeutic inhibition BACE1 in humans, similar to BACE1-null mice, may be free of mechanism-based toxicity and thus an effective method in Alzheimer's disease treatment. These analyses boosted the drug discoveries targeting complete BACE1 inhibition and many clinical trials which resulted in mild cognitive impairment. This leads to a need for further research and subsequent studies on complete BACE1 inhibition that found BACE1-null mice showed increased instances of seizures, schizophrenia-like phenotypes, demyelination, axonal misguidance, and high offspring mortality rates in contrast to BACE1^{+/+} mice (i.e., heterozygous mice with a genetically decreased level of BACE1). BACE1^{+/+} mice failed to exhibit any side effects and phenotypic changes but showed potential $A\beta$ reduction. Recent studies have shown that the A673T mutation of APP impairs cleavage by BACE1, resulting in protection against AD, thus fur-

ther supporting BACE1 inhibition [16]. From the prior studies conducted [14–16] and the critical role played in amyloidogenic processing, BACE1 can thus be considered a probable target. Four therapeutic agents approved by the FDA (donepezil, rivastigmine, galantamine, and memantine) are currently being used to mitigate AD symptoms, but these drugs have not been able to prevent or reverse disease progression [17, 18]. As of 2018, 112 anti-AD agents are under investigation. Approximately 27% of these agents now in phase III clinical trials and ~ 5% in phase II clinical trials are BACE1 inhibitors [19]. As of 2019, the BACE1 inhibitors LY-3314814, MK-8931 (verubecestat), and LY-3202626 have been dropped from clinical trials. Some of BACE1 inhibitors like elenbecestat is progressing through the clinical trials [20, 21]. Every year several drugs are being developed to treat AD in hopes of satisfactory results, but most of them fail at the preclinical stage, even before entering clinical trials. There are different approaches for targeting BACE1, like inhibition of activity and suppression of BACE1 expression [22, 23]. As of 2019, less than 2% of the drugs in studies act by inhibition of BACE1 activity. Inhibition of BACE1 by natural products has rendered promising results in AD therapeutics as exemplified by flavonoids (galangin, myricetin, baicalein), alkaloids (berberine), terpenes, etc., which have shown BACE1 inhibition. These compounds show significant potential to act as therapeutic drugs. Further efficient strategies for inhibiting BACE1 activity is required in order to reduce the side effects caused by biological functions due to its long-term use [24].

The C99 cleavage can be impaired without interfering with multifunctional neuregulin 1 processing, which is a potential substrate of BACE1. This, combined with the possibility of compartmentalisation of the target, specifically targeting BACE1 inhibitors to endosomal compartments, preventing action on non-amyloid substrates [25], has paved the future for the development of a promising BACE1-based anti-AD therapeutic approach. The identification of further β APP-selective natural BACE1 inhibitors could be advantageous, as this would prevent the secondary adverse effects due to cleavage of other substrates supporting some important physiological functions. The comprehension of safety profiles of many natural compounds showing potential BACE1 inhibition is already well understood due to its long-term use in traditional Chinese medicine (TCM) and Ayurvedic medicine. Some of the natural compounds that are widely used in traditional medicinal care with an emphasis on flavonoids, phenolic compounds, tannins, alkaloids, chalcones, and terpenes and exhibit effective action by the inhibition of BACE1 activity at low concentrations are discussed below.

Search Strategy

A systemic search was carried out for literature in electronic databases, including PubMed, Scopus, Embase, Web of Science, Science Direct, and Google Scholar and were screened for natural compounds that exhibited potential BACE1 activity inhibition. *In vivo*, *in vitro*, and clinical evidence that assessed the therapeutic and preventive potential of natural compounds against the BACE1 enzyme involved in the production of $A\beta$, which is a major component of amyloid plaques and neurofibrillary tangles, were col-



lected. Relevant articles were searched to obtain natural BACE1 activity inhibitors for the mitigation of AD and its symptoms. The following key words were used to obtain significant data about the topic: Alzheimer's disease, BACE1, natural compounds, BACE1 activity, and BACE1 inhibitors. The articles focusing on plant extracts or modified compound derivatives showing BACE1 inhibition were excluded in the study. Only full length articles available in the English language were reviewed.

Flavonoids Having Potential Anti-beta-Site Amyloid Precursor Protein-Cleaving Enzyme 1 Activity

Descamps et al. [26] identified two bioflavonoids, rutin (**1**) (found in *Fagopyrum esculentum* Moench) and galangin (**2**) (found in *Alpinia officinarum* Hance), which have the ability to impair BACE1 cleavage by acting as β APP-selective BACE1 inhibitors. Galangin in cell culture studies and AD transgenic mice studies (J20 mice) conducted (at dosages of 50 μ M and 40 mg/kg, respectively) showed inhibition of BACE-dependent β APP nuclear signalling, without affecting neuregulin. Hence, these commonly used nutritional supplement showed a novel mechanism to modulate β APP processing even at lower concentrations, avoiding potential toxicity caused by direct inhibition of BACE1.

Baicalein (**3**) (5,6,7-trihydroxy-2-phenyl-chromen-4-one) is a flavone isolated from the roots of *Scutellaria baicalensis* Georgi. It is used in TCM and known to have potent antioxidant and free radical scavenging properties [27]. Recently, it gained attention due to its ability to act against neurodegenerative diseases [28, 29]. Baicalein (**3**) has been shown to have more potent anti-BACE1 activity when compared with other flavonoid compounds like luteolin and quercetin, with an IC_{50} value of about 10 μ M [30]. Baicalein (**3**) has been shown to inhibit BACE1 activity as well as $A\beta$ oligomerisation and fibrillation, and prevents $A\beta$ -induced toxicity in PC12 cells

along with the disaggregation of preformed $A\beta$ amyloid fibrils [31]. The ability of the compound to cross the BBB is found to be negatively correlated with dose [31]. Gu et al. [32] suggest that the long-term oral administration of baicalein (**3**) leads to the reduction in BACE1 protein levels. Durairajan et al. [33] reported that prolonged treatment of baicalein led to enhanced $A\beta$ deposition in both N2a-Swedish APP cells and TgCRND8 APP transgenic mice [33]. However, no significant changes in BACE1 protein levels were obtained in TgCRND8 APP transgenic mice when treated with baicalein. The $A\beta$ increasing effect of baicalein might be due to its off-target action, probably via impairing ubiquitin proteasomal clearance function. These are significantly visible only while the administration of the compound is done in increased dosages of 25 mg/kg/day. Zhang et al. [28] reported that Chinese hamster ovary cells expressing wild-type APP and Tg2576 mice, when treated with baicalein, showed reduced $A\beta$ through promotion of the non-amyloidogenic pathway. This contrast in data might be due to the difference in incubation time. The data showing a decrease in the sAPP β level is not shown in the study of Zhang et al. Hence, baicalein may modulate BACE1 in an extremely dose-dependent manner.

Camellikaempferoside B (**4**) is a natural acylglycoside flavone compound that is isolated from Fuzhuan brick tea [fermented *Camellia sinensis* (L.) Kuntze]. The structure of the compound contains groups of *p*-coumaric acid and rhamnopyranosyl along a kaempferol backbone. Yang et al. [34] showed that camellikaempferoside B (**4**) does not interfere with BACE1 expression, but it reduces BACE1 activity at a concentration of 25 μ M in both a cell-free system and in APP-expressing cells. This compound acts on several of the active sites in BACE1 via hydrogen bonds, leading to a reduction in BACE1 activity and $A\beta$ production. Preformed fibril disaggregation is shown by camellikaempferoside B (**4**), and this compound has also been shown to form structurally abnormal $A\beta$ oligomers, which are not involved in pathogenesis [34].

Quercetin (**5**) is a flavonoid compound that is abundantly found in plants like *Allium cepa* L., *Malus pumila* Mill., etc. [35].

Shimmyo et al. [36] has shown that quercetin (5) reduces BACE1 activity in a cell-free system (IC_{50} of $5.4 \pm 0.5 \mu\text{M}$). In a neuronal cell system, the compound has also showed BACE1 reducing activity (IC_{50} of $50 \mu\text{M}$). The compound appears to maintain its stability for about 24 h *in vivo*. Lu et al. [37] reported that quercetin-treated male C57BL/6 strain mice exhibited reduced BACE1 expression. Quercetin may reduce BACE1 expression and, thus, negatively regulate the amyloidogenic processing of βAPP . But the data provided does not show much significant reduction in BACE1 protein levels as claimed. Nevertheless, the abundance, stability, and wide availability of the compound and its action on the neuronal cell system provides a need for further studies.

Myricetin (6) is a plant-derived flavonoid compound that belongs to the class of polyphenols [38]. Due to its structural similarity to quercetin (5), myricetin (6) is sometimes referred to as hydroxyquercetin [39]. Since myricetin (6) has antioxidant properties, it has also been reported to show a neuroprotective effect against neuronal cell injury induced by $A\beta$ [40]. The small molecular weight and hydrophobic characteristics may help it to cross the BBB, thus giving it a therapeutic advantage [41]. Shimmyo et al. [40] have reported that myricetin (6) has dual activity, as it can directly inhibit BACE1 activity without affecting protein expression and showed activation of α secretase (ADAM10) in a cell-free enzyme activity assay. The IC_{50} of myricetin (6) was calculated to be $2.8 \mu\text{M}$ in inhibiting BACE1 activity. Three hydrogen bonds formed by myricetin (6) with BACE1 (one each with Gln 73 and Trp 198, and one with Asp32) stabilise the binding. The effect of myricetin (6) on neuronal cells is less than expected. Myricetin (6) has been shown to be unstable. This might be because, after 24 h of treatment, the compound is metabolised, losing the hydroxyl groups essential for BACE1 inhibition [40]. Myricetin (6) exhibits β -sheet structure disruption activity and also inhibits $A\beta$ fibril generation [42].

Genistein (7) is an isoflavone compound isolated from *Glycine max* (L.) Merr., which inhibited BACE1 activity in a dose-dependent manner with an IC_{50} value of $6.3 \times 10^{-5} \text{M}$. It inhibits BACE1 activity in a noncompetitive reversible manner. In *in vivo* and cell-based studies, genistein (7) has also been shown to inhibit $A\beta$ -induced inflammation and cell death [43–45]. Even at a higher concentration of 500 mg/kg/day in rats, it was found to be pharmacologically safe [46]. It was noted that even at a lower concentration, the compound was able to cross the BBB without causing any neurotoxicity [46]. The significance of the compound, to be considered as a potential candidate for AD treatment, requires further studies. The structure of flavonoids exhibiting potent anti-BACE1 activity is depicted in ► Fig. 2.

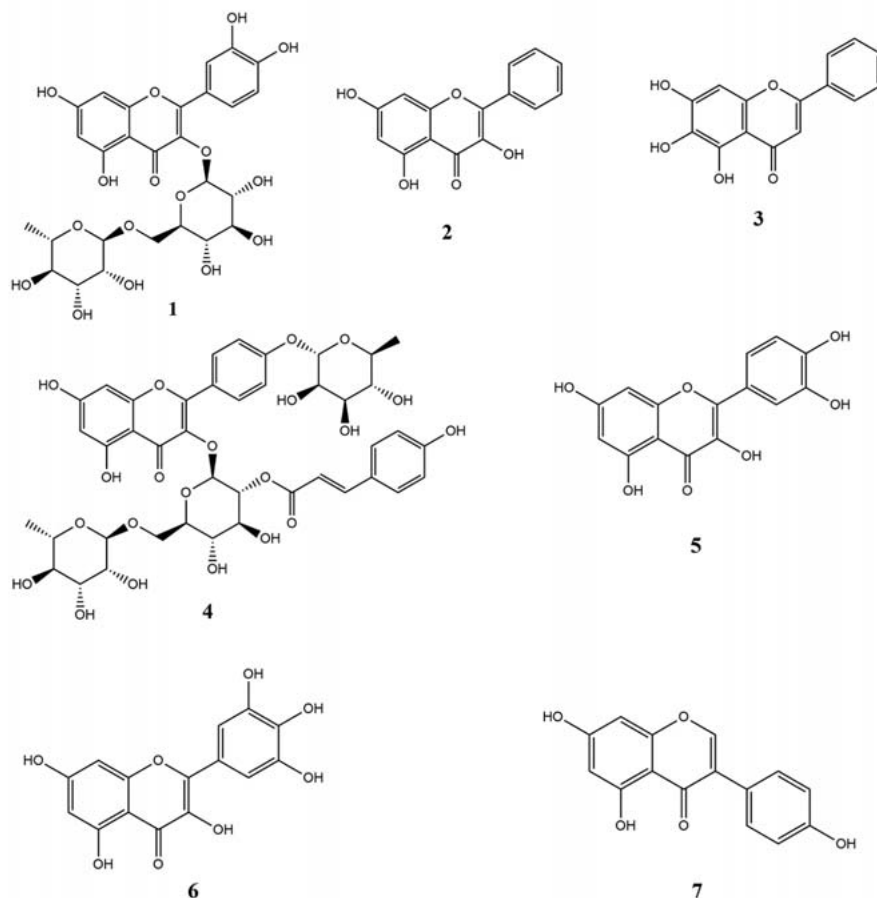
Phenolics and Tannins Having Potential Anti-beta-Site Amyloid Precursor Protein-Cleaving Enzyme 1 Activity

Salvianolic acid B (8) (Sal B) was isolated from the root of *Salvia miltiorrhiza* Bunge (Lamiaceae family) [47]. This plant is widely used to treat cardiovascular and cerebrovascular diseases [48, 49]. Sal B (8) is a water-soluble polyphenolic caffeic acid derivative [50]. Lin et al. [51] showed that Sal B (8) can protect neuronal PC-

12 cells from $A\beta$ -induced toxicity. Further studies conducted by Durairajan et al. [52] reported that Sal B (8) can disaggregate pre-formed fibrils and inhibit $A\beta$ fibril formation. Tang et al. and Durairajan et al. independently reported that Sal B modulated BACE1 activity in SH-SY5Y-APPsw cells and it decreased $A\beta$ generation in H4-SwedAPP, N2a-SwedAPP, and HEK-BACE1 cells. Many, possibly important, variations can be noticed in these studies: i) Tang et al. suggested that BACE1 expression was reduced at $50 \mu\text{M}$ [53], but Durairajan et al. [54] showed that Sal B (8) does not affect BACE1 expression. It only reduces BACE1 activity; ii) Tang et al. used a narrow range of concentrations ($25\text{--}50 \mu\text{M}$) of Sal B (8), whereas the other study used concentrations varying from $1\text{--}50 \mu\text{M}$; iii) The higher concentrations of Sal B (8) may affect cellular viability, possibly leading to toxicity. Durairajan et al., in their studies, reported cellular toxicity and viability by the LDH and MTT analyses and found the range of concentrations to be safe. Tang et al. did not provide cellular viability results; iv) The decrease in the level of sAPP β coincides with a decrease in CTF β fragments. No data on CTF β fragments was provided in the other study; and v) Durairajan et al. provided data of molecular docking and Sal B shows slight binding to the catalytic domain, whereas Tang et al. did not provide docking studies. Yu et al. [55] reported that Sal B shows negligible binding to the catalytic sites using molecular docking methods. Tang et al. suggest that Sal B suppresses BACE1 expression. But due to discrepancies between these results, further studies are required to clearly understand the mechanism of action of Sal B on BACE1 (► Fig. 3).

Ferulic acid (9) is a phenolic compound that is included in the human diet, as it is found in cereals like *Oryza sativa* L. and *Triticum aestivum* L., in fruits like *Solanum lycopersicum* L., *Ananas comosus* (L.) Merr., and *Citrus sinensis* (L.) Osbeck, and in vegetables [56]. The compound is known to possess anti-inflammatory, anti-carcinogenic and antioxidant properties [57–59]. Mori et al. [56] reported that ferulic acid (9) acts by targeting BACE1, both in *in vitro* and *in vivo* studies conducted. At the concentration of $1.57 \mu\text{M}$, this compound significantly reduced $A\beta$ variants [57]. In a cell-free BACE1 activity assay, it was demonstrated that ferulic acid (9) acts on BACE1 by both directly attenuating BACE1 enzymatic activity and targeting BACE1 stability without affecting mRNA expression levels. Ferulic acid (9) has a low molecular weight (194.18g/mol) [60], high bioavailability in rat models [57], and remains stable in the body, which serves as an advantage in therapeutics; however, due to its nature as a charged molecule with a hydroxyl group, its ability to cross the BBB remains uncertain [56]. Some reports have suggested the presence of the molecule in rodent brains following peripheral administration [60]. Hence, ferulic acid (9) and its derivatives have the potential to combat AD.

Tannic acid (10) is present in plants like *Quercus velutina* Lam., *Camellia sinensis* (L.) Kuntze, etc. Oral administration of tannic acid (10) improved behavioural impairment, reduced cerebral amyloidosis, and increased the anti-amyloidogenic βAPP processing in *in vivo* studies conducted in transgenic PSAPP mice (30 mg/kg/day dosage) without exhibiting any side effects [61]. Moreover, tannic acid has shown to dose dependently downregulate the generation of $A\beta_{40}$ and $A\beta_{42}$ and inhibit the level of CTF β cleavage products [61]. It was also noted that tannic acid inhibits BACE1 expression and β secretase activity without altering BACE1 mRNA, promoting



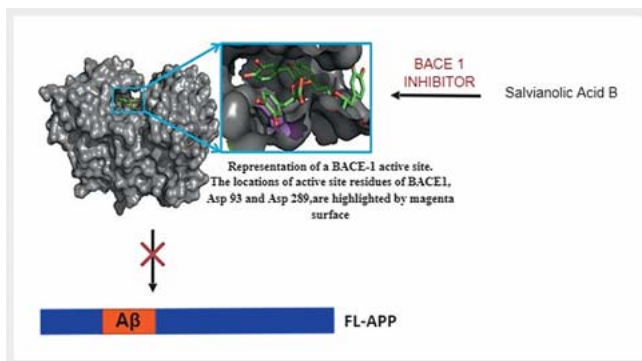
► **Fig. 2** Structures of flavonoids rutin (1), galangin (2), baicalein (3), camellikaempferoside (4), quercetin (5), myricetin (6), and genistein (7) having BACE1 inhibitory activity.

non-amyloidogenic β APP processing [61]. These properties increase the potential of tannic acid to progress into clinical studies. The structures of phenolics and tannins showing anti-BACE1 activity are depicted in ► **Fig. 4**.

Alkaloids Showing Inhibition of beta-Site Amyloid Precursor Protein-Cleaving Enzyme 1 Activity

Berberine (11) is a multifunctional isoquinoline alkaloid with neuropharmacological properties, which can be isolated from plants like *Coptis chinensis* Franch., *Berberis vulgaris* L., and many more [62]. Asai et al. [63] showed that berberine modulated β APP processing, resulting in a reduction of the $A\beta$ protein. In our previous study, the chronic administration of berberine (11) in transgenic AD mice for approximately 4 months showed significant mitigation of $A\beta$ pathology without influencing BACE1 protein levels at an oral dosage of 25 or 100 mg/kg per day [62]. *In vivo* studies have suggested that berberine (11) is able to cross the BBB and reach the brain in a dose- and time-dependent manner [64].

It was reported that the berberine reduced BACE1 activity and prevented the neurodegeneration of the hippocampus in a rabbit model of AD [65]. Cai et al. [66] established that berberine inhibits β/γ -secretases (main components PS1, Aph-1 α , and Pen-2) activity and enhances α -secretase, thereby alleviating $A\beta$ pathology in the brains of AD transgenic mice. However, the BACE1 inhibitory activity of berberine (11) was found to be less (IC_{50} value > 100 μ M) [67]. In another study, through surface plasmon resonance (SPR) binding analysis and docking studies, the direct binding of berberine and BACE-1 was illustrated [68]. Reports of berberine acting on the BACE1 expression levels are also available, hence, further studies are required for the identification of the potential mechanism of action of the compound [69]. No potential toxicity was shown by the compound in both the *in vivo* and *in vitro* studies conducted, but protoberberine alkaloids such as the epiberberine (12) groenlandicine (13) exhibit promising dose-dependent BACE1 noncompetitive inhibition with IC_{50} values of 8.55 and 19.68 μ M, respectively [67]. Further evaluation of the protoberberine compounds can provide valuable insight on its mechanism of action. The structures of alkaloids having anti-BACE1 activity are depicted in ► **Fig. 4**.



► **Fig. 3** Salvianolic B binds to the active site (Asp 32, Asp 228) of BACE1 thus preventing its activity. Similarly, the other BACE1 inhibitors bind to the active site of the enzyme and disturb its enzyme activity.

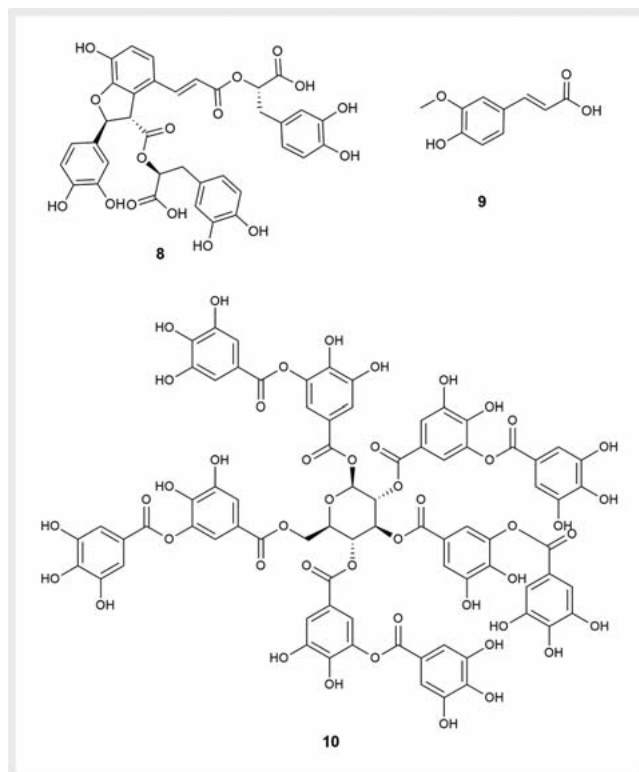
Chalcones

2, 2',4'-Trihydroxychalcone acid (TDC) (**14**) from *Glycyrrhiza glabra* L. (licorice) (chalcones-flavanoids) noncompetitively inhibits BACE1 activity (K_i value of 3.08 μM). With an IC₅₀ of 2.5 μM, TDC (**14**) showed a dose-dependent decrease in the generation of Aβ₄₀ and Aβ₄₂ levels in HEK293-APPsw cells by effectively suppressing BACE1 activity on the βAPP [70]. In the above cell-based assay and *in vivo* (B6C3-Tg mice) study, TDC (**14**) effectively decreased Aβ in cells by suppressing BACE1 activity without exhibiting any off-target effect on α and γ secretases and showed no effect on BACE1 protein levels. It ameliorated the neurobehavioural activities and memory impairment in an AD mouse model at a dosage of 9 mg/kg per day with no obvious animal toxicity [70].

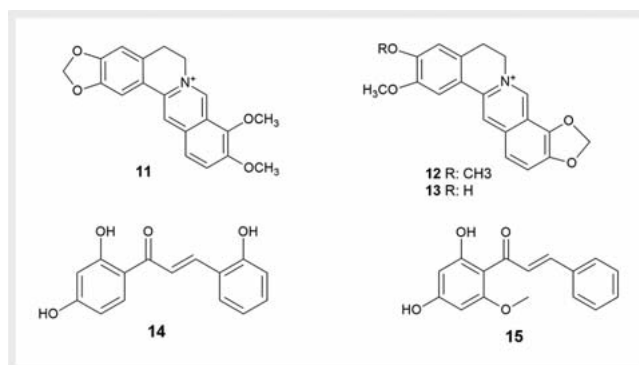
Cardamonin (**15**), a chalconoid, isolated from *Boesenbergia rotunda* (L.) Mansf. has a strong inhibition value with an IC₅₀ value of 4.35 ± 0.38 μM. The compound does not affect the TACE (α secretase) to cause any detrimental effects. The docking studies, with -9.5 kcal/mol results, suggest its affinity to tightly bind to the enzyme and it has been proved to easily pass the BBB [71]. The oral administration of cardamonin (**15**) for 30 weeks at the dose of 10 mg/kg did not exhibit any apparent toxicity, thus suggesting its safe consumption, but further tests such as *in vitro*, *in vivo* and cell viability should be done in order to justify the effectiveness of the compound [72]. Further confirmation of these results warrant the use of chalcones as therapeutic agents for AD. The structures of alkaloids and chalcones having anti-BACE1 activity are depicted in ► **Fig. 5**.

Terpenes

Gracilins are secondary metabolites that are derived from the marine sponge *Spongionella Bowerbank*. Leirós et al. [73] isolated several natural compounds from the marine sponge and conducted several *in vivo* and *in vitro* studies and found that gracilins can effectively reduce tau hyperphosphorylation and Aβ accumulation. Its successful action of Aβ reduction may be caused by its effective inhibition of the BACE1 enzyme. In SH-SY5Y tau441 human cell lines and 3xTg-AD mice studies, it was noted that gracilin L (**16**) at a mere concentration of 1 μM exhibited a significant BACE1 reduction, decreasing its activity by 24.6 ± 4.2% [74]. Even though

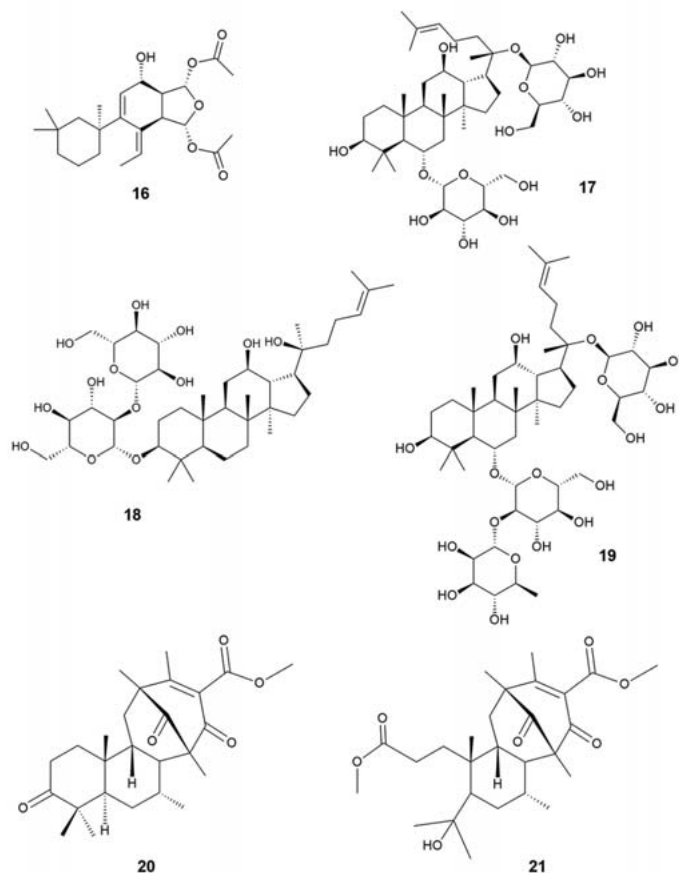


► **Fig. 4** Structures of salvianolic acid B (**8**), ferulic acid (**9**), and tannic acid (**10**) having BACE1 inhibitory activity.



► **Fig. 5** Structures of alkaloids [berberine (**11**), epiberberine (**12**), groenlandicine (**13**)] and chalcones [2,2',4'-trihydroxychalcone acid (**14**), cardamonin (**15**)] with anti-BACE1 activity.

the other gracilins obtained showed BACE1 inhibition activity, the % of decrease is comparatively not significant. Gracilin L (**16**) reduced the expression of tau levels by 48.2 ± 8.5% at a concentration of 1 μM. The levels of Aβ accumulation found *in vivo* after treatment with 0.04 mg/kg of gracilin L (**16**) showed an 86.2% decrease. The multitarget mechanism of action of the gracilin compound on various targets led to ERK inhibition, and BACE1 inhibition correlated with the action of reduced tau hyperphosphorylation. A decrease of Aβ highlights the value of the natural com-



► **Fig. 6** Structures of terpenoid compounds gracilin L (16), ginsenosides Re (17), Rg1 (18), Rg3 (19), asperterpene A (20), and asperterpene B (21) with anti-BACE1 activity.

found to be used for targeting AD. The neuroprotective action reported earlier by Leirós et al. [74] was further cemented by the current data. Further studies might be required in order to lament the priority and viability of usage of this compound for AD treatment.

Ginsenosides, the major pharmacologically active compounds isolated from various species of ginseng like *Panax ginseng* C.A. Mey., have also been observed to reduce BACE1, albeit with a different mode of action. Ginsenosides are steroidal triterpenoid saponins with a four-ring steroid backbone. Chen et al. [75] reported that three ginsenosides [Re (17), Rg1 (18) or Rg3 (19)] at a dose of 25 mg/kg significantly reduced the amount of A β 40/A β 42 conducted in a cellular-based assay in Tg2576 mice. Ji et al. [76], in the same year, reported that ginsenoside Re (17) protects PC12 cells from cellular injury induced by amyloid A β . Ginsenosides Re (17) was observed to reduce BACE1 activity along with BACE1 expression, having no effect on the total APP levels and sAPP α levels in *in vitro* studies [77]. Ginsenoside Rg1 (18) showed improved memory and learning capacity in *in vitro* studies conducted. Wang et al. [78] reported that ginsenoside Rg1 (18) downregulated BACE1 activity and protects against A β -induced cytotoxicity in *in vitro* studies conducted in PC12 cells. The IC₅₀ value of Rg1 was 6.18 \pm 0.96 μ M.

Asperterpenes A and B (20 and 21), meroterpenoids obtained from the soil-derived mold *Aspergillus terreus* Thom., have shown potent BACE1 inhibitory activities in a cell-based assay using HEK-BACE1 cells. The IC₅₀ values of the asperterpenes A and B (19 and 20), obtained were 78 and 59 nM, respectively. When HEK-293 and N2a-APP cell lines were treated with asperterpene A (20) at a concentration of 70 nM, it significantly reduced A β 42 formation and inhibited BACE1 activity. In animal studies conducted on triple transgenic mice (3XTgAD mice), asperterpene A (21) treatment ameliorated learning and memory deficit along with BACE1 activity (concentration 2 μ g/ μ L). The exposure of the cells to this compound did not affect cell viability or cause toxic effects in the *in vivo* and *in vitro* studies conducted [79]. Qi et al. [80] isolated new meroterpenoids, asperterpenes from *Aspergillus terreus*, cultured on *Oryza sativa* L. Of the 10 isolated compounds, asperterpenes E, F, and J exhibited better BACE1 inhibitory activities compared to others, with IC₅₀ values of 3.32, 5.85, and 31.68 μ M, respectively, in a BACE1 FRET (fluorescence resonance energy transfer) inhibition experiment. Terresterpenes A and B, which are compounds isolated from extracts of *A. terreus*, displayed potential BACE1 inhibitory activity in *in vitro* studies (IC₅₀ values of 5.98 and 11.42 μ M) [81]. Compounds isolated from *A. terreus*, up till now, have displayed one of the strongest inhib-

itions against BACE1 activity and reduction in the formation of A β . These findings thus warrant the use of asperterpenes as an anti-AD therapeutic agent. The structures of terpenes and terpenoids showing anti-BACE1 activity are depicted in ► Fig. 6.

Conclusion and Future Perspectives

The potential of BACE1 to act as a therapeutic target in the treatment of AD has only been investigated for the past decade. Evidence suggests that the timing of administration of BACE1 inhibitors may play a critical role in the successful treatment of AD. The advancement in future diagnostic technologies will lead to the identification of high-risk individuals easier and, hence, provide potential for early treatment [82]. The side effects related to BACE1 inhibition are of some concern, but the novel method of β APP-selective BACE inhibition reduces the risks. The identification and characterisation of natural BACE1 inhibitors have potential in anti-AD therapeutics, as the long-term use of these compounds in different ancient treatments also provides an advantage of its safety profile. All natural products discussed in this review article have the ability to effectively inhibit BACE1 activity and lower neurotoxic A β formation. Focussing on the effects of the natural compounds on their action as anti-AD therapeutic agents brings focus to their mechanistic view of action by specifically targeting BACE1 substrate without any off-target action. In the future, we may discover more natural compounds showing severe efficiency, specificity, bioavailability, and safety with an effective grip on the various novel mechanisms of action to efficiently treat AD.

Acknowledgements

This work was supported by grants from Hong Kong's Health and Medical Research Fund (HMRF 15163481) and the Department of Science and Technology's Core Research grant (SERB/CRG/2018/001596), Government of India. The authors also thank Dr. Martha Dahlen for her English editing and critical review.

Conflict of Interest

The authors declare that they have no conflict of interest.

References

- [1] Golde TE, DeKosky ST, Galasko D. Alzheimer's disease: The right drug, the right time. *Science* 2018; 362: 1250–1251
- [2] World Health Organization. Dementia. Available at <https://www.who.int/news-room/fact-sheets/detail/dementia>. Accessed September 21, 2019
- [3] Scheltens P, Blennow K, Breteler MM, de Strooper B, Frisoni GB, Salloway S, Van der Flier WM. Alzheimer's disease. *Lancet* 2016; 388: 505–517
- [4] Koss DJ, Jones G, Cranston A, Gardner H, Kanaan NM, Platt B. Soluble pre-fibrillar tau and β -amyloid species emerge in early human Alzheimer's disease and track disease progression and cognitive decline. *Acta Neuropathol* 2016; 132(6): 875–895
- [5] Selkoe DJ. Physiological production of the beta-amyloid protein and the mechanism of Alzheimer's disease. *Trends Neurosci* 1993; 16: 403–409
- [6] Edbauer D, Winkler E, Regula JT, Pesold B, Steiner H, Haass C. Reconstitution of gamma-secretase activity. *Nat Cell Biol* 2003; 5: 486–488
- [7] Dislich B, Lichtenthaler SF. The membrane-bound aspartyl protease BACE1: molecular and functional properties in Alzheimer's disease and beyond. *Front Physiol* 2012; 17: 3–8
- [8] Hussain I, Powell D, Howlett DR, Tew DG, Meek TD, Chapman C, Gloger IS, Murphy KE, Southan CD, Ryan DM, Smith TS, Simmons DL, Walsh FS, Dingwall C, Christie G. Identification of a novel aspartic protease (Asp 2) as beta-secretase. *Mol Cell Neurosci* 1999; 14: 419–427
- [9] Lin X, Koelsch G, Wu S, Downs D, Dashti A, Tang J. Human aspartic protease memapsin 2 cleaves the beta-secretase site of beta-amyloid precursor protein. *Proc Natl Acad Sci U S A* 2000; 97: 1456–1460
- [10] Fukumoto H, Rosene DL, Moss MB, Raju S, Hyman BT, Irizarry MC. Beta-secretase activity increases with aging in human, monkey, and mouse brain. *Am J Pathol* 2004; 164: 719–725
- [11] Bennett BD, Babu-Khan S, Loeffler R, Louis JC, Curran E, Citron M, Vassar R. Expression analysis of BACE2 in brain and peripheral tissues. *J Biol Chem* 2000; 275: 20647–20651
- [12] Farzan M, Schnitzler CE, Vasilieva N, Leung D, Choe H. BACE2, a beta-secretase homolog, cleaves at the beta-site and within the amyloid-beta region of the amyloid-beta precursor protein. *Proc Natl Acad Sci U S A* 2000; 97: 9712–9717
- [13] Voytyuk I, Mueller SA, Herber J, Snellinx A, Moechars D, van Loo G, Lichtenthaler SF, De Strooper B. BACE2 distribution in major brain cell types and identification of novel substrates. *Life Sci Alliance* 2018; 1: e201800026
- [14] Roberds SL, Anderson J, Basi G, Bienkowski MJ, Branstetter DG, Chen KS, Freedman SB, Frigon NL, Games D, Hu K, Johnson-Wood K, Kappenman KE, Kawabe TT, Kola I, Kuehn R, Lee M, Liu W, Motter R, Nichols NF, Power M, Robertson DW, Schenk D, Schoor M, Shopp GM, Shuck ME, Sinha S, Svensson KA, Tatsuno G, Tintrup H, Wijsman J, Wright S, McConlogue L. BACE knockout mice are healthy despite lacking the primary beta-secretase activity in brain: implications for Alzheimer's disease therapeutics. *Hum Mol Genet* 2001; 12: 1317–1324
- [15] Luo Y, Bolon B, Kahn S, Bennett BD, Babu-Khan S, Denis P, Fan W, Kha H, Zhang J, Gong Y, Martin L, Louis JC, Yan Q, Richards WG, Citron M, Vassar R. Mice deficient in BACE1, the Alzheimer's β -secretase, have normal phenotype and abolished β -amyloid generation. *Nat Neurosci* 2001; 4: 231–232
- [16] Jonsson T, Atwal JK, Steinberg S, Snaedal J, Jonsson PV, Bjornsson S, Stefansson H, Sulem P, Gudbjartsson D, Maloney J, Hoyte K, Gustafson A, Liu Y, Lu Y, Bhargale T, Graham RR, Huttenlocher J, Bjornsdottir G, Andreassen OA, Jonsson EG, Palotie A, Behrens TW, Magnusson OT, Kong A, Thorsteinsdottir U, Watts RJ, Stefansson K. A mutation in APP protects against Alzheimer's disease and age-related cognitive decline. *Nature* 2012; 488: 96–99
- [17] Fan LY, Chiu MJ. Combination and current concepts as well as future strategies for the treatment of Alzheimer's disease. *Neuropsychiatr Dis Treat* 2014; 10: 439–451
- [18] Anand R, Gill KD, Mahdi A. Therapeutics of Alzheimer's disease: past, present and future. *Neuropharmacol* 2013; 76: 27–50
- [19] Cummings J, Lee G, Ritter A, Zhong K. Alzheimer's disease drug development pipeline. *Alzheimers Dement (N Y)* 2018; 4: 195–214
- [20] Piton M, Hirtz C, Desmetz C, Milhau J, Lajoie AD, Bennis K, Lehmann S, Gabelle A. Alzheimer's disease: advances in drug development. *J Alzheimers Dis* 2018; 65: 3–13
- [21] Mullard A. BACE inhibitor bust in Alzheimer trial. *Nat Rev Drug Discov* 2017; 16: 155
- [22] Buggia-Prévo V, Fernandez CG, Riordan S, Vetrivel KS, Roseman J, Waters J, Bindokas VP, Vassar R, Thinakaran G. Axonal BACE1 dynamics and targeting in hippocampal neurons: a role for Rab11 GTPase. *Mol Neurodegener* 2014; 9: 1
- [23] Gautam V, D'Avanzo C, Hebisch M, Kovacs DM, Kim DY. BACE1 activity regulates cell surface contactin-2 levels. *Mol Neurodegener* 2014; 9: 4

- [24] Yan R. Physiological functions of the β -site amyloid precursor protein cleaving enzyme 1 and 2. *Front Mol Neurosci* 2017; 10: 97
- [25] Ben Halima S, Mishra S, Raja KMP, Willem M, Baici A, Simons K, Brüstle O, Koch P, Haass C, Calhiser A, Rajendran L. Specific inhibition of β -secretase processing of the Alzheimer disease amyloid precursor protein. *Cell Rep* 2016; 14: 2127–2141
- [26] Descamps O, Spilman P, Zhang Q, Libeu CP, Poksay K, Gorostiza O, Campagna J, Jagodzinska B, Bredesen DE, John V. A β PP-selective BACE inhibitors (ASBI): novel class of therapeutic agents for Alzheimer's disease. *J Alzheimers Dis* 2013; 37: 343–355
- [27] Gao D, Sakurai K, Chen J, Ogiso T. Protection by baicalein against ascorbic acid-induced lipid peroxidation of rat liver microsomes. *Res Commun Mol Pathol Pharmacol* 1995; 90: 103–114
- [28] Zhang SQ, Obregon D, Ehrhart J, Deng J, Tian J, Hou H, Giunta B, Sawmiller D, Tan J. Baicalein reduces β -amyloid and promotes non amyloidogenic amyloid precursor protein processing in an Alzheimer's disease transgenic mouse model. *J Neurosci Res* 2013; 91: 1239–1246
- [29] Xue X, Liu H, Qi L, Li X, Guo C, Gong D, Qu H. Baicalein ameliorated the upregulation of striatal glutamatergic transmission in the mice model of Parkinson's disease. *Brain Res Bull* 2014; 103: 54–59
- [30] Paris D, Mathura V, Ait-Ghezala G, Beaulieu-Abdelahad D, Patel N, Bachmeier C, Mullan M. Flavonoids lower Alzheimer's A β production via an NF κ B dependent mechanism. *Bioinformation* 2011; 6: 229–236
- [31] Lu JH, Ardah MT, Durairajan SSK, Liu LF, Xie LX, Fong WF, Hasan MY, Huang JD, El-Agnaf OM, Li M. Baicalein inhibits formation of α -synuclein oligomers within living cells and prevents A β peptide fibrillation and oligomerisation. *Chembiochem* 2011; 12: 615–624
- [32] Gu XH, Xu LJ, Liu ZQ, Wei B, Yang YJ, Xu GG, Yin XP, Wang W. The flavonoid baicalein rescues synaptic plasticity and memory deficits in a mouse model of Alzheimer's disease. *Behav Brain Res* 2016; 311: 309–321
- [33] Durairajan SSK, Huang YY, Yuen PY, Chen LL, Kwok KY, Liu LF, Song JX, Han QB, Xue L, Chung SK, Huang JD, Baum L, Senapati S, Li M. Effects of Huanglian-jie-du-tang and its modified formula on the modulation of amyloid- β precursor protein processing in Alzheimer's disease models. *PLoS One* 2014; 9: e92954
- [34] Yang S, Liu W, Lu S, Tian YZ, Wang WY, Ling TJ, Liu RT. A novel multifunctional compound camellikaempferoside b decreases A β production, interferes with A β aggregation, and prohibits A β -mediated neurotoxicity and neuroinflammation. *ACS Chem Neurosci* 2016; 7: 505–518
- [35] Priprem A, Watanatorn J, Suttthiparinyanont S, Phachonpai W, Muchimapura S. Anxiety and cognitive effects of quercetin liposomes in rats. *Nanomedicine* 2008; 4: 70–78
- [36] Shimmyo Y, Kihara T, Akaike A, Niidome T, Sugimoto H. Flavonols and flavones as BACE1 inhibitors: structure-activity relationship in cell-free, cell-based and *in silico* studies reveal novel pharmacophore features. *Biochim Biophys Acta* 2008; 1780: 819–825
- [37] Lu J, Wu DM, Zheng YL, Hu B, Zhang ZF, Shan Q, Zheng ZH, Liu CM, Wang YJ. Quercetin activates AMP-activated protein kinase by reducing PP2C expression protecting old mouse brain against high cholesterol-induced neurotoxicity. *Pathol* 2010; 222: 199–212
- [38] Semwal DK, Semwal RB, Combrinck S, Viljoen A. Myricetin: A dietary molecule with diverse biological activities. *Nutrients* 2016; 8: 90
- [39] Umadevi I, Daniel M, Sabnis SD. Chemotaxonomic studies on some members of Anacardiaceae. *Proc Plant Sci* 1988; 98: 205–208
- [40] Shimmyo Y, Kihara T, Akaike A, Niidome T, Sugimoto H. Multifunction of myricetin on A β : neuroprotection via a conformational change of A β and reduction of A β via the interference of secretases. *J Neurosci Res* 2007; 24: 368–377
- [41] Ramezani M, Darbandi N, Khodaghohi F, Hashemi A. Myricetin protects hippocampal CA3 pyramidal neurons and improves learning and memory impairments in rats with Alzheimer's disease. *Neural Regen Res* 2016; 11: 1976–1980
- [42] Ono K, Yoshiike Y, Takashima A, Hasegawa K, Naiki H, Yamada M. Potent anti-amyloidogenic and fibril-destabilizing effects of polyphenols *in vitro*: implications for the prevention and therapeutics of Alzheimer's disease. *J Neurochem* 2003; 87: 172–181
- [43] Vallés SL, Borrás C, Gambini J, Friull J, Ortega A, Sastre J, Pallardó FV, Viña J. Oestradiol or genistein rescues neurons from amyloid beta-induced cell death by inhibiting activation of p38. *Aging Cell* 2008; 7: 112–118
- [44] Valles SL, Dolz-Gaiton P, Gambini J, Borrás C, Lloret A, Pallardo FV, Viña J. Estradiol or genistein prevent Alzheimer's disease-associated inflammation correlating with an increase PPAR gamma expression in cultured astrocytes. *Brain Res* 2010; 1312: 138–144
- [45] Ma W, Ding B, Yu H, Yuan L, Xi Y, Xiao R. Genistein alleviates b-amyloid induced inflammatory damage through regulating toll-like receptor 4/nuclear factor κ B. *J Med Food* 2015; 18: 273–279
- [46] Tsai TH. Concurrent measurement of unbound genistein in the blood, brain and bile of anesthetized rats using microdialysis and its pharmacokinetic application. *J Chromatogr A* 2005; 1073: 317–322
- [47] Chen W, Chen G. Danshen (*Salvia miltiorrhiza* Bunge): A prospective healing sage for cardiovascular diseases. *Curr Pharm Des* 2017; 23: 5125–5135
- [48] Gao Y, Zhang K, Zhu F, Wu Z, Chu X, Zhang X, Zhang Y, Zhang J, Chu L. *Salvia miltiorrhiza* (Danshen) inhibits L-type calcium current and attenuates calcium transient and contractility in rat ventricular myocytes. *J Ethnopharmacol* 2014; 158: 397–403
- [49] Zhou X, Chan SW, Tseng HL, Deng Y, Hoi PM, Choi PS, Or PM, Yang JM, Lam FF, Lee SM, Leung GP, Kong SK, Ho HP, Kwan YW, Yeung JH. Danshensu is the major marker for the antioxidant and vasorelaxation effects of Danshen (*Salvia miltiorrhiza*) water-extracts produced by different heat water-extractions. *Phytomedicine* 2012; 19: 1263–1269
- [50] Jiang RW, Lau KM, Hon PM, Mak TC, Woo KS, Fung KP. Chemistry and biological activities of caffeic acid derivatives from *Salvia miltiorrhiza*. *Curr Med Chem* 2005; 12: 237–246
- [51] Lin YH, Liu AH, Wu HL, Westenbroek C, Song QL, Yu HM, Ter Horst GJ, Li XJ. Salvianolic acid B, an antioxidant from *Salvia miltiorrhiza*, prevents A β (25–35)-induced reduction in BPRP in PC12 cells. *Biochem Biophys Res Commun* 2006; 348: 593–599
- [52] Durairajan SSK, Yuan Q, Xie L, Chan WS, Kum WF, Koo I, Liu C, Song Y, Huang JD, Klein WL. Salvianolic acid B inhibits Ab fibril formation and disaggregates preformed fibrils and protects against Ab-induced cytotoxicity. *Neurochem Int* 2008; 52: 741–750
- [53] Tang Y, Huang D, Zhang MH, Zhang WS, Tang YX, Shi ZX, Deng L, Zhou DH, Lu XY. Salvianolic Acid B inhibits A β generation by modulating BACE1 activity in SH-SY5Y-APPsw cells. *Nutrients* 2016; 8: 333
- [54] Durairajan SSK, Chirasani VR, Shetty SG, Iyaswamy A, Malampati S, Song J, Liu L, Huang J, Senapati S, Li M. Decrease in the generation of amyloid- β due to salvianolic acid B by modulating BACE1 activity. *Curr Alzheimer Res* 2017; 14: 1–9
- [55] Yu T, Paudel P, Seong SH, Kim JA, Jung HA, Choi JS. Computational insights into β -site amyloid precursor protein enzyme 1 (BACE1) inhibition by tanshinones and salvianolic acids from *Salvia miltiorrhiza* via molecular docking simulations. *Comput Biol Chem* 2018; 74: 273–285
- [56] Mori T, Koyama N, Guillot-Sestier MV, Tan J, Town T. Ferulic acid is a nutraceutical β -secretase modulator that improves behavioural impairment and Alzheimer-like pathology in transgenic mice. *PLoS One* 2013; 8: e55774
- [57] Srinivasan M, Sudheer AR, Menon VP. Ferulic acid: therapeutic potential through its antioxidant property. *J Clin Biochem Nutr* 2007; 40: 92–100
- [58] Kawabata K, Yamamoto T, Hara A, Shimizu M, Yamada Y, Matsunaga K, Tanaka T, Mori H. Modifying effects of ferulic acid on azoxymethane-induced colon carcinogenesis in F344 rats. *Cancer Lett* 2000; 157: 15–21
- [59] Sultana R, Ravagna A, Mohammad-Abdul H, Calabrese V, Butterfield DA. Ferulic acid ethyl ester protects neurons against amyloid β -peptide (1–

- 42)-induced oxidative stress and neurotoxicity: relationship to antioxidant activity. *J Neurochem* 2005; 92: 749–758
- [60] Qin J, Chen D, Lu W, Xu H, Yan C, Hu H, Chen B, Qiao M, Zhao X. Preparation, characterization, and evaluation of liposomal ferulic acid *in vitro* and *in vivo*. *Drug Dev Ind Pharm* 2008; 34: 602–608
- [61] Mori T, Rezai-Zadeh K, Koyama N, Arendash GW, Yamaguchi H, Kakuda N, Horikoshi-Sakuraba Y, Tan J, Town T. Tannic acid is a natural β -secretase inhibitor that prevents cognitive impairment and mitigates Alzheimer-like pathology in transgenic mice. *J Biol Chem* 2012; 287: 6912–6927
- [62] Durairajan SSK, Liu LF, Lu JH, Chen LL, Yuan Q, Chung SK, Huang L, Li XS, Huang JD, Li M. Berberine ameliorates β -amyloid pathology, gliosis, and cognitive impairment in an Alzheimer's disease transgenic mouse model. *Neurobiol Aging* 2012; 33: 2903–2919
- [63] Asai M, Iwata N, Yoshikawa A, Aizaki Y, Ishiura S, Saido TC, Maruyama K: Berberine alters the processing of Alzheimer's amyloid precursor protein to decrease Abeta secretion. *Biochem Biophys Res Commun* 2007; 352: 498–502
- [64] Tan XS, Ma JY, Feng R, Ma C, Chen WJ, Sun YP, Fu J, Huang M, He CY, Shou JW, He WY, Wang Y, Jiang JD. Tissue distribution of berberine and its metabolites after oral administration in rats. *PLoS One* 2013; 8: e77969
- [65] Panahi N, Mahmoudian M, Mortazavi P, Hashjin GS. Effects of berberine on beta-secretase activity in a rabbit model of Alzheimer's disease. *Arch Med Sci* 2013; 9: 146–150
- [66] Cai Z, Wang C, He W, Chen Y. Berberine alleviates amyloid-beta pathology in the brain of app/ps1 transgenic mice via inhibiting β/γ -secretases activity and enhancing α -secretases. *Curr Alzheimer Res* 2018; 15: 1045–1052
- [67] Jung HA, Min BS, Yokozawa T, Lee JH, Kim YS, Choi JS. Anti-Alzheimer and antioxidant activities of *Coptidis rhizoma* alkaloids. *Biol Pharm Bull* 2009; 32: 1433–1438
- [68] Chu M, Chen X, Wang J, Guo L, Wang Q, Gao Z, Kang J, Zhang M, Feng J, Guo Q, Li B, Zhang C, Guo X, Chu Z, Wang Y. Polypharmacology of berberine based on multi-target binding motifs. *Front Pharmacol* 2018; 9: 801
- [69] Zhang H, Zhao C, Cao G, Guo L, Zhang S, Liang Y, Qin C, Su P, Li H, Zhang W. Berberine modulates amyloid- β peptide generation by activating AMP-activated protein kinase. *Neuropharmacol* 2017; 125: 408–417
- [70] Zhu Z, Li C, Wang X, Yang Z, Chen J, Hu L, Jiang H, Shen X. 2,2',4'-trihydroxychalcone from *Glycyrrhiza glabra* as a new specific BACE1 inhibitor efficiently ameliorates memory impairment in mice. *J Neurochem* 2010; 114: 374–385
- [71] Youn K, Jun M. Biological evaluation and docking analysis of potent BACE1 inhibitors from *Boesenbergia rotunda*. *Nutrients* 2019; 11: 662
- [72] James S, Aparna JS, Paul AM, Lankadasari MB, Mohammed S, Binu VS, Santhoshkumar TR, Reshmi G, Harikumar KB. Cardamomin inhibits colonic neoplasia through modulation of microRNA expression. *Sci Rep* 2017; 7: 13945
- [73] Leirós M, Alonso E, Rateb ME, Houssen WE, Ebel R, Jaspars M, Alfonso A, Botana L. Gracilins: *Spongionella*-derived promising compounds for Alzheimer disease. *Neuropharmacol* 2015; 93: 285–293
- [74] Leirós M, Sánchez JA, Alonso E, Rateb ME, Houssen WE, Ebel R, Jaspars M, Alfonso A, Botana LM. Spongionella secondary metabolites protect mitochondrial function in cortical neurons against oxidative stress. *Mar Drugs* 2014; 12: 700–718
- [75] Chen F, Eckman EA, Eckman CB. Reductions in levels of the Alzheimer's amyloid beta peptide after oral administration of ginsenosides. *FASEB J* 2006; 20: 1269–1271
- [76] Ji ZN, Dong TT, Ye WC, Choi RC, Lo CK, Tsim KW. Ginsenoside Re attenuate beta-amyloid and serum-free induced neurotoxicity in PC12 cells. *J Ethnopharmacol* 2006; 107: 48–52
- [77] Cao G, Su P, Zhang S, Guo L, Zhang H, Liang Y, Qin C, Zhang W. Ginsenoside Re reduces A β production by activating PPAR γ to inhibit BACE1 in N2a/APP695 cells. *Eur J Pharmacol* 2016; 793: 101–108
- [78] Wang YH, Du GH. Ginsenoside Rg1 inhibits beta-secretase activity *in vitro* and protects against Abeta-induced cytotoxicity in PC12 cells. *J Asian Nat Prod Res* 2009; 11: 604–612
- [79] Qi C, Bao J, Wang J, Zhu H, Xue Y, Wang X, Li H, Sun W, Gao W, Lai Y, Chen JG, Zhang Y. Asperterpenes A and B, two unprecedented meroterpenoids from *Aspergillus terreus* with BACE1 inhibitory activities. *Chem Sci* 2016; 7: 6563–6572
- [80] Qi C, Liu M, Zhou Q, Gao W, Chen C, Lai Y, Hu Z, Xue Y, Zhang J, Li D, Li XN, Zhang Q, Wang J, Zhu H, Zhang Y. BACE1 inhibitory meroterpenoids from *Aspergillus terreus*. *J Nat Prod* 2018; 81: 1937–1945
- [81] Qi C, Qiao Y, Gao W, Liu M, Zhou Q, Chen C, Lai Y, Xue Y, Zhang J, Li D, Wang J, Zhu H, Hu Z, Zhou Y, Zhang Y. New 3,5-dimethylorsellinic acid-based meroterpenoids with BACE1 and AChE inhibitory activities from *Aspergillus terreus*. *Org Biomol Chem* 2018; 16: 9046–9052
- [82] Voytyuk I, De Strooper B, Chavez-Gutierrez L. Modulation of γ - and β -secretases as early prevention against Alzheimer's disease. *Biol Psychiatry* 2018; 83: 320–327