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ScienceDirect
Phytomedicine 14 (2007) 289–300
Phytomedicine
www.elsevier.de/phymed

Acetylcholinesterase inhibitors from plants

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Abstract

Inhibition of acetylcholinesterase (AChE), the key enzyme in the breakdown of acetylcholine, is considered as a promising strategy for the treatment of neurological disorders such as Alzheimer's disease, senile dementia, ataxia and myasthenia gravis. A potential source of AChE inhibitors is certainly provided by the abundance of plants in nature. This article aims to provide a comprehensive literature survey of plants that have been tested for AChE inhibitory activity. Numerous phytoconstituents and promising plant species as AChE inhibitors are being reported in this communication.

Keywords: Plants; Botanicals; Acetylcholinesterase inhibition; Acetylcholine; Alzheimer's disease

Introduction

Principal role of acetylcholinesterase (AChE) is the termination of nerve impulse transmission at the cholinergic synapses by rapid hydrolysis of acetylcholine (ACh). Inhibition of AChE serves as a strategy for the treatment of Alzheimer's disease (AD), senile dementia, ataxia, myasthenia gravis and Parkinson's disease (Anonymous, 2000; Brenner, 2000; Rahman and Choudhary, 2001). There are a few synthetic medicines, e.g. tacrine, donepezil, and the natural product-based rivastigmine for treatment of cognitive dysfunction and memory loss associated with AD (Oh et al., 2004). These compounds have been reported to have their adverse effects including gastrointestinal disturbances and problems associated with bioavailability (Schulz, 2003; Melzer, 1998), which necessitates the interest in finding better AChE inhibitors from natural resources.

AD is one of the most common forms of dementia affecting so many elderly people. Besides the neuropathologic hallmarks of this disease, namely neurofibrillary tangles and neuritic plaques, it is characterized neurochemically by a consistent deficit in cholinergic neurotransmission, particularly affecting cholinergic neurons in the basal forebrain (Price, 1986; Kasa et al., 1997). The evidence stems from data of several authors that demonstrated the reduction in activity of enzymes involved in the synthesis of acetylcholine, i.e. choline acetyl transferase or excess degradation of ACh by AChE (Davies and Maloney, 1976; Sims et al., 1983; DeKosky et al., 1992). The first AChE inhibitors (AChEIs) specifically approved for the treatment of AD was introduced in 1993 as 1, 2, 3, 4-tetrahydro-9-aminoacridine (tacrine) (Whitehouse, 1993). Currently, several AChE inhibitors, such as donepezil (Kelly et al., 1997), galantamine (Scott and Goa, 2000) and rivastigmine (Gottwald and Rozanski, 1999) are available for

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doi:10.1016/j.phymed.2007.02.002

the symptomatic treatment of patients with mild-to-moderate AD. Cholinesterase inhibitory therapy may be considered, by its pharmacological nature, as a simple symptomatic short-term intervention. However, data emerging from long-term mostly open label trials is that the maintenance of the clinical effect can be prolonged to at least 1 year. In some clinical studies, the data indicate that beneficial effects can be maintained for up to 36 months; These effects of stabilization of the cognitive status of the patients suggest conceivably a structural effect of the treatment on pathological features of the disease; Giacobini (2002) suggested that the effects may arise from the interaction of these drugs with the amyloid cascade, influencing the expression and/or the metabolic processing of the amyloid precursor protein (APP) and slowing down one of the major pathological steps of the disease process. In traditional practices of numerous plants have been used to treat cognitive disorders, including neurodegenerative diseases and different neuropharmacological disorders. Ethnopharmacological approach and bioassay-guided isolation have provided a lead in identifying potential AChE inhibitors from plant sources, including those for memory disorders. This article highlights on the plants and/or their active constituents so far reported to have AChE inhibitory activity.

Several methods for screening of AChE inhibitory activity from natural resources has been reported based on Ellman's reactions (Ellman et al., 1961). Moreover, Spectrophotometric determination thin-layer chromatography method (Ingkaninan et al., 2000; Marston et al., 2002) and micro-plate assay (Ingkaninan et al., 2000, Brlihlmann et al., 2004) have been reported to be useful. HPLC method for detection of AChE inhibition on immobilized AChE column (Andrisano et al., 2001) and HPLC with on-line coupled UV–MS–biochemical detection for AChE inhibitory activity (Ingkaninan et al., 2000) have also been reported.

Plants as a source of acetylcholinesterase inhibitors

A variety of plants has been reported to show AChE inhibitory activity and so may be relevant to the treatment of neurodegenerative disorders such as AD. A list of plants reported to have significant AChE inhibitory activity is shown in Table 1.

Bacopa monniera and *Ginkgo biloba* are well-known cognitive enhancers in Indian and Chinese traditional medicine systems. Standardized extracts of *Bacopa monniera* and *G. biloba* both showed a dose-dependent inhibitory effect on AChE activity (Das et al., 2002). Eighty percent methanolic extract of *Myricaria elegans* Royle was found to have significant AChE inhibitory activity (Ahmad et al., 2003).

Methanolic extracts of seven herbs *Acorus calamus, Acorus gramineus, Bupleurm facaltum, Dioscorea batatas, Epimedium koreanum, Poria cocos* and *Zizyphi jujuba,* used in traditional Korean medicine for improvement of memory and cognition in old age have been tested for cholinesterase inhibitory properties and significant inhibition of the enzyme was shown by extracts from *Acorus calamus* and *E. koreanum* (Oh et al., 2004). Ingkaninan et al. (2000, 2003) screened the methanolic extracts of 32 plants used in Thai traditional rejuvenating and neurotonic remedies, for inhibitory activity on AChE and found that the extracts from roots of *Stephania suberosa* and *Tabernaemontana divaricata* showed significant inhibitory activity.

The chloroform:methanol (1:1) extracts of a number of the plant species namely *Corydalis solida* (L.) Swartz subsp. *solida* and *Glaucium corniculatum* (L.) J. H. Rudolph (Papaveraceae), *Rhododendron ponticum* L. subsp. *ponticum* and *Rhododendron luteum* Sweet. (Ericaceae), *Buxus sempervirens* L. (Buxaceae), *Vicia faba* L. (Fabaceae), *Robinia pseudoacacia* L. (Caeselpiniaceae), *Tribulus terrestris* L. and *Zygophyllum fabago* L. (Zygophyllaceae), *Lycopodium clavatum* L. (Lycopodiaceae), *Fumaria vaillantii* Lois., *Fumaria capreolata* L., *Fumaria kralikii* Jordan, *Fumaria asepala* Boiss., *Fumaria densiflora* DC., *Fumaria flabellata* L., *Fumaria petteri* Reichb. subsp. *thuretii* (Boiss.) Pugsley, *Fumaria macrocarpa* Boiss. ex Hausskn., *Fumaria cilicica* Hauskkn., *Fumaria parviflora* Lam. and *Fumaria judaica* Boiss. (Fumariaceae) were screened for their anti-cholinesterase activity (Orhan et al., 2004). The extracts of *Rhododendron ponticum*, *Rhododendron luteum*, *Corydalis solida*, *Glaucium corniculatum*, and *Buxus sempervirens* showed remarkable inhibitory activity above 50% inhibition rate at 1 mg/ml.

Amongst plants that have been investigated for dementia therapy, *Salvia* is one of the most numerous genera within the family Lamiaceae and grows in many parts of the world. It causes inhibition of AChE as well as nicotinic activity (Perry et al., 2000, 2001).

Phytoconstituents having acetylcholinesterase inhibitory activity

Work on new bioactive compounds from medicinal plants has led to the isolation and structure elucidation of a number of exciting new pharmacophores. A list of phytoconstituents having significant AChE inhibitory activity is provided in Table 2 and structures of these compounds are shown in Fig. 1. *Physostigma venenosum* was used traditionally in Africa as a ritual poison, claimed to determine the guilt or innocence of person accused of a crime. Treatment with the indole alkaloid physostigmine [1], an AChE inhibitor isolated from

P. venenosum, has improved cognitive function in several in vivo studies. Physostigmine, a short-acting reversible AChE inhibitor, is also reported to have shown significant cognitive benefits in both normal and AD patients, but clinical use may be limited by its short half-life, which would require multiple daily dosing (Da-Yuan et al, 1996; Mukherjee, 2001).

Chemical structure of physostigmine has provided a template for the development of rivastigmine [2], an AChE inhibitor that is licensed for use in the UK for the symptomatic treatment of mild-to-moderately severe AD (Foye et al., 1995). Rivastigmine is reported to inhibit AChE in the cortex and hippocampus, brain areas involved in cognition. Thus, it is apparent that plant-derived alkaloid AChE inhibitors may be important for the development of more appropriate drug candidates for the treatment of AD (Foye et al., 1995).

Galanthus nivalis was used traditionally in Bulgaria and Turkey for neurological conditions. Galantamine [3] is an Amaryllidaceae alkaloid obtained from *Galanthus nivalis* L. Galantamine is reported to be more selective for AChE than butyrylcholinesterase, and provides complete oral bioavailability. It is licensed in Europe for AD treatment, was well tolerated and significantly improved cognitive function when administered to AD patients, in multi-center randomized-controlled trials (Lopez et al., 2002). Initially derived from extracts of snowdrop and daffodil bulbs, this phenanthrene alkaloid is now synthetically produced. It is a reversible competitive AChE inhibitor that also allosterically modulates nicotinic receptors (this effect is probably independent of its cholinesterase inhibition). It has an elimination half-life of about 6 h. Metabolism produces four compounds, one of which is more active as a cholinesterase inhibitor than galantamine itself. Over 2000 patients have been involved in double-blind placebo-controlled trials of galantamine where positive effects on cognitive symptoms have been associated with significant benefits in activities of daily living (Da-Yuan et al, 1996). Other Amaryllidaceae alkaloids such as assoanine [4], epinorgalantamine [5], oxoassoanine [6], sanguinine [7], 11-hydroxygalantamine [8] have also been reported to possess AChE activity (Lopez et al., 2002).

The lycopodium alkaloid huperzine A [9] related to the quinolizidines, is a potent, yet reversible, inhibitor of AChE and is used in China for treating patients with myasthenia gravis and AD. The source of huperzine A is *Huperzia serrata,* a moss that has been used for treating contusions, strains, hematuria and swelling in Chinese folk medicine (Wang and Tang, 1998). It improved memory retention processes in cognitively impaired aged and adult rats (Raves et al., 1997). In a multi-center, double blind trial, huperzine A significantly improved memory and behavior in AD patients, and was reported to be more selective for AChE than butyrylcholinesterase and less toxic than the synthetic AChE inhibitors donepezil and tacrine. It may also have potential in the attenuation of memory deficits and neuronal damage that occurs after ischemia, so may therefore is beneficial in the treatment of cerebrovascular-type dementia (Raves et al., 1997).

Numerous essential oils and their monoterpene constituents have been investigated for their effects on AChE, and have shown weak inhibitory activity. For example, the essential oils from *Melissa officinalis* and *Rosmarinus officinalis* have been reported to inhibit erythrocyte AChE in vitro (Howes et al., 2003a, b). Other monoterpenes that are reported to inhibit AChE include geraniol, 3-carene, α-caryophyllene and limonene. The structural diversity of the active anti-cholinesterase terpenoids complicates the prediction of potential structure–activity relationships. One feature associated with AChE inhibition is a hydrophobic ligand. The hydrophobic active site of AChE is reported to be susceptible to hydrophobic interactions. Monoterpenes consist of a hydrocarbon skeleton, which may contribute to their anti-cholinesterase activity. Monoterpenes may be cyclic (e.g. 1,8-cineole and α-pinene) or acyclic (e.g. geraniol and linalool), a feature that may also influence anti-cholinesterase activity. Further investigations may determine if a cyclic component or particular functional group favors AChE inhibition. Considering the relatively weak anti-cholinesterase activity of terpenoids reported to date, it is unlikely that they may be used therapeutically for cognitive disorders. However, analogues of active terpenoid compounds may be developed to enhance efficacy.

More recently, the stilbene oligomer viniferin [10] from *Caragana chamlague*, has also been identified as reversible and non-competitive inhibitor of AChE (Da-Yuan et al, 1996). Structure–activity relationship suggested that the nitrogen substituents at C-3 and/or C-20 of steroidal skeleton and the hydrophobic properties of the pregnane skeleton are the key structural features contributed to the inhibitory potency of pregnane-type steroidal alkaloids against AChE (Khalid et al., 2004).

Bioassay-guided fractionation of the methanolic extract resulted in the isolation of three furanocoumarins, isoimperatorin, imperatorin and oxypeucedanin as active principles from the methanolic extract of the roots of *Angelica dahurica*, which inhibited AChE activity in a dose-dependent manner (Kim et al., 2002). In a bioassay-guided search for AChE inhibitors four isoquinoline alkaloids, corynoxidine, protopine, palmatine and berberine have been isolated from the methanolic extract of the aerial parts of *Corydalis speciosa* (Kim et al., 2004). Bioassay-directed phytochemical investigations on a number of medicinal plants of Pakistan and Iran have led to the isolation of AChE inhibitors such as buxamine B [11], *N*, *N*-dimethyl

Table 1. Plants with acetylcholinesterase inhibitory activity

Plant	Family	Parts used	Type of extract	Activity (% inhibition) (concentration)	References
Abutilon indicum Linn.	Malvaceae	Whole	Methanolic	30.66 ± 1.06 (0.1 mg/ml)	Ingkaninan et al. (2003)
Acanthus ebracteatus Vahl.	Acanthaceae	Aerial part	Methanolic	36.19 ± 8.00 (0.1 mg/ml)	Ingkaninan et al. (2003)
Aegle marmelos (Linn.) Correa ex Roxb.	Rutaceae	Fruit pulp	Methanolic	44.65 ± 3.04 (0.1 mg/ml)	Ingkaninan et al. (2003)
Albizia procera (Roxb.) Benth.	Leguminosae	Bark	Methanolic	40.71 ± 0.46 (0.1 mg/ml)	Ingkaninan et al. (2003)
Bacopa monniera Linn.	Scrophulariaceae	Whole	Ethanolic	42.9 ± 1.2 (0.1 mg/ml)	Das et al. (2002)
Butea superba Roxb.	Leguminosae	Root barks	Methanolic	55.87 ± 5.83 (0.1 mg/ml)	Ingkaninan et al. (2003)
Buxus sempervirens Linn.	Buxaceae	Whole	Chloroform: methanol (1:1)	61.76 ± 0.76 (1 mg/ml)	Orhan et al. (2004)
Carthamus tinctorius Linn.	Compositae	Flower	Methanolic	30.33 ± 9.22 (0.1 mg/ml)	Ingkaninan et al. (2003)
Cassia fistula Linn.	Leguminosae	Roots	Methanolic	54.13 ± 3.90 (0.1 mg/ml)	Ingkaninan et al. (2003)
Corydalis solida Linn.	Papaveraceae	Whole	Chloroform:methanol (1:1)	87.56 ± 1.24 (1 mg/ml)	Orhan et al. (2004)
Cyperus rotundus Linn.	Cyperaceae	Whole	Methanolic	44.19 ± 2.27 (0.1 mg/ml)	Ingkaninan et al. (2003)
Euphorbia antiquorum Linn.	Euphorbiaceae	Stem	Methanolic	42.31 ± 9.10 (0.1 mg/ml)	Ingkaninan et al. (2003)
Fumaria vaillantii Lois.	Fumariaceae	Whole	Chloroform: methanol (1:1)	94.23 ± 0.47 (1 mg/ml)	Orhan et al. (2004)
Fumaria capreolata Linn.	Fumariaceae	Whole	Chloroform: methanol (1:1)	96.89 ± 0.17 (1 mg/ml)	Orhan et al. (2004)
Fumaria kralikii Jordan	Fumariaceae	Whole	Chloroform:methanol (1:1)	84.98 ± 1.07 (1 mg/ml)	Orhan et al. (2004)
Fumaria asepala Boiss.	Fumariaceae	Whole	Chloroform: methanol (1:1)	91.99 ± 0.70 (1 mg/ml)	Orhan et al. (2004)
Fumaria densiflora DC.	Fumariaceae	Whole	Chloroform: methanol (1:1)	93.42 ± 0.92 (1 mg/ml)	Orhan et al. (2004)
Fumaria flabellate Linn.	Fumariaceae	Whole	Chloroform: methanol (1:1)	92.14 ± 1.01 (1 mg/ml)	Orhan et al. (2004)
Fumaria petteri Reichb subsp. *thuretii* (Boiss.)	Fumariaceae	Whole	Chloroform: methanol (1:1)	89.45 ± 0.86 (1 mg/ml)	Orhan et al. (2004)
Fumaria macrocarpa Boiss. ex Hausskn.	Fumariaceae	Whole	Chloroform: methanol (1:1)	93.43 ± 0.64 (1 mg/ml)	Orhan et al. (2004)
Fumaria cilicica Hausskn.	Fumariaceae	Whole	Chloroform: methanol (1:1)	88.03 ± 0.65 (1 mg/ml)	Orhan et al. (2004)
Fumaria parviflora Lam.	Fumariaceae	Whole	Chloroform: methanol (1:1)	87.02 ± 0.31 (1 mg/ml)	Orhan et al. (2004)
Fumaria judaica Boiss.	Fumariaceae	Whole	Chloroform: methanol (1:1)	96.47 ± 0.63 (1 mg/ml)	Orhan et al. (2004)

Ginkgo biloba Linn.	Coniferae	Whole	Ethanolic	50% (268.33 μg)	Das et al. (2002) Perry et al. (1998)
Glaucium corniculatum (Linn.) J.H. Rudolph.	Papaveraceae	Whole	Chloroform:methanol (1:1)	86.55 ± 0.67 (1 mg/ml)	Orhan et al. (2004)
Lycopodium clavatum Linn.	Lycopodiaceae	Whole	Chloroform:methanol (1:1)	49.85 ± 1.33 (1 mg/ml)	Orhan et al. (2004)
Mammea harmandii Kosterm.	Guttiferae	Flower	Methanolic	33.63 ± 8.00 (0.1 mg/ml)	Ingkaninan et al. (2003)
Melissa officinalis Linn.	Lamiaceae	Aerial part	Volatile oil	—	Perry et al. (1998)
Michelia champaca Linn.	Magnoliaceae	Leaf	Methanolic	34.88 ± 4.56 (0.1 mg/ml)	Ingkaninan et al. (2003)
Mimosa pudica Linn.	Leguminosae	Whole	Methanolic	21.40 ± 6.68 (0.1 mg/ml)	Ingkaninan et al. (2003)
Mimusops elengi Linn.	Sapotaceae	Flower	Methanolic	32.81 ± 5.36 (0.1 mg/ml)	Ingkaninan et al. (2003)
Musa sapientum Linn.	Musaceae	Fruit	Methanolic	29.14 ± 4.73 (0.1 mg/ml)	Ingkaninan et al. (2003)
Myricaria elegans Royle	Tamaricaceae	Aerial	Methanolic	74.8% (0.2 μg/ml)	Ahmad et al. (2003)
Nelumbo nucifera Gaertn.	Nelumbonaceae	Stamen	Methanolic	23.77 ± 2.83 (0.1 mg/ml)	Ingkaninan et al. (2003)
Paederia linearis Hook. f.	Rubiaceae	Whole	Methanolic	29.31 ± 6.39 (0.1 mg/ml)	Ingkaninan et al. (2003)
Piper interruptum Opiz	Piperaceae	Stems	Methanolic	65.16 ± 8.13 (0.1 mg/ml)	Ingkaninan et al. (2003)
Piper nigrum Linn.	Piperaceae	Seeds	Methanolic	58.02 ± 3.83% (0.1 mg/ml)	Ingkaninan et al. (2003)
Plumbago indica Linn.	Plumbaginaceae	Root	Methanolic	30.14 ± 3.28 (0.1 mg/ml)	Ingkaninan et al. (2003)
Ptychopetalum olacoides Benth.	Olacaceae	Root	Ethanol	Dose dependent activity at doses of 50 and 100 mg/kg, i.p.	Siqueira et al. (2003)
Rhododendron luteum Sweet.	Ericaceae	Whole	Chloroform:methanol (1:1)	76.32 ± 0.58 (1 mg/ml)	Orhan et al. (2004)
Rhododendron ponticum Linn. subsp. *Ponticum*	Ericaceae	Whole	Chloroform:methanol (1:1)	93.03 ± 1.12 (1 mg/ml)	Orhan et al. (2004)
Rhodiola rosea Linn.	Crassulaceae	Root	Methanol	42.00 ± 3.20 (10 g/l)	Hillhouse et al. (2004)
Salvia lavandulaefolia Vahl.	Lamiaceae	Whole	Steam distilled oil	63.0 ± 3.7 (0.1 μg/ml)	Perry et al. (1996, 2000, 2001)
Salvia officinalis Linn.	Lamiaceae	Whole	Ethanol 95% Steam distilled oil	68.2 ± 15.6 (2.5 mg/ml) 52.4 ± 0.8 (0.1 μg/ml)	Perry et al. (1996, 2000, 2001)
Stephania suberosa Forman.	Menispermaceae	Roots	Methanolic	91.93 ± 10.80 (0.1 mg/ml)	Ingkaninan et al. (2003)
Streblus asper Lour.	Moraceae	Seed	Methanolic	30.51 ± 4.21 (0.1 μg/ml)	Ingkaninan et al. (2003)
Tabernaemontana divaricata (Linn.) R. Br. Ex	Apocynaceae	Roots	Methanolic	93.50 ± 0.37 (0.1 mg/ml)	Ingkaninan et al. (2003)
Terminalia bellirica (Gaertn.) Roxb.	Combretaceae	Fruit	Methanolic	39.68 ± 8.15 (0.1 mg/ml)	Ingkaninan et al. (2003)
Tiliacora triandra (Colebr.) Diel	Menispermaceae	Root	Methanolic	42.29 ± 2.89 (0.1 mg/ml)	Ingkaninan et al. (2003)
Vicia faba Linn.	Fabaceae	Whole	Chloroform:methanol (1:1)	45.23 ± 1.03 (1 mg/ml)	Orhan et al. (2004)

Table 2. Phytoconstituents having acetylcholinesterase inhibitory activity

Alkaloids					
Name of alkaloid	Class	Sources	Plant family	Activity	References
Assoanine	Steroidal alkaloid	*Narcissus assoanus*	Amaryllidaceae	50% inhibition at $3.87 \pm 0.24\,\mu M$	Lopez et al. (2002)
Buxamine B	Steroidal alkaloid	*Bucus hyrcana Bucus papillosa*	Buxaceae	50% inhibition at $7.56 \pm 0.008\,\mu M$	Rahman and Choudhary (2001)
Coronaridine	Indole alkaloid	*Tabernaemontana australis*	Apocynaceae	Minimum concentration of $25\,\mu M$ to produce detectable spot in TLC	Andrade et al. (2005)
Corynoline	Isoquinoline alkaloid	*Corydalis incisa*	Papaveraceae	50% inhibition at $30.6\,\mu M$	Kim (2002)
N, *N*-dimethyl buxapapine	Steroidal alkaloid	*Bucus papillosa*	Buxaceae	50% inhibition at $7.28 \pm 0.06\,\mu M$	Rahman and Choudhary (2001)
Epinorgalantamine	Steroidal alkaloid	Narcissus confuses N. perezchiscanoi *Narcissus leonensis N. legionensis Narcissus poeticus*	Amaryllidaceae	50% inhibition at $9.60 \pm 0.65\,\mu M$	Lopez et al. (2002)
Galantamine	Steroidal alkaloid	Galanthus nivalis *Narcissus confuses Lycorus radiate*	Amaryllidaceae	50% inhibition at $1.07 \pm 0.18\,\mu M$	Rhee et al. (2001); Rizzi et al. (1999); Ingkaninan et al. (2003); Lopez et al. (2002)
(-)-Huperzine A	Quinolizidine alkaloid	*Huperzia serrata Huperzia dalhousieana*	Lycopodiaceae	50% inhibition at $10^{-4}\,\mu M$	Tang et al. (1994) Orhan et al. (2004); Ashani et al. (1994)
11-Hydroxygalantamine	Steroidal alkaloid	*Narcissus poeticus*	Amaryllidaceae	50% inhibition at $1.61 \pm 0.21\,\mu M$	Lopez et al. (2002)
Oxoassoanine	Steroidal alkaloid	*Narcissus assoanus*	Amaryllidaceae	50% inhibition at $47.21 \pm 1.13\,\mu M$	Lopez et al. (2002)
Palmatine	Isoquinoline alkaloid	*Corydalis speciosa*	Papaveraceae	50% inhibition at $5.8\,\mu M$	Kim et al. (2004)
Physostigmine	Indole alkaloid	*Physostigma venenosum*	Leguminosae	50% inhibition at $6 \times 10^{-4}\,\mu M$	Karczmar (1998)
Protopine	Isoquinoline alkaloid	*Corydalis speciosa*	Papaveraceae	50% inhibition at $16.1\,\mu M$	Kim et al. (2004)
Rupicoline	Indole alkaloid	*Tabernaemontana australis*	Apocynaceae	Minimum concentration of $25\,\mu M$ to produce detectable spot in TLC	Andrade et al. (2005)
Sanguinine	Steroidal alkaloid	*Eucharis grandiflora*	Amaryllidaceae	50% inhibition at $0.10 \pm 0.01\,\mu M$	Lopez et al. (2002)

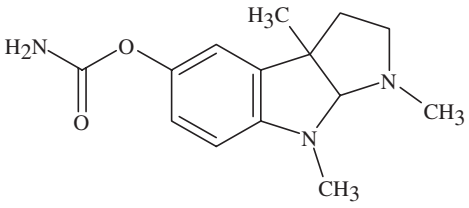
Sarsalignone	Steroidal alkaloid	*Sarcococca saligna*	Buxaceae	50% inhibition at $7.028 \pm 0.007\,\mu M$	Rahman and Choudhary (2001)
α-Solanine	Glycoalkaloid	*Solanum tuberosum*	Solanaceae	44.3% inhibition at 10 μM	Roddick (1989) Mc Gehee et al. (2000)
Vaganine	Steroidal alkaloid	*Sarcococca saligna*	Buxaceae	50% inhibition at $8.59 \pm 0.155\,\mu M$	Rahman and Choudhary (2001)
Voacangine	Indole alkaloid	*Tabernaemontana australis*	Apocynaceae	Minimum concentration of 25 μM to produce detectable spot in TLC	Andrade et al. (2005)
Voacangine hydroxyindolenine	Indole alkaloid	*Tabernaemontana australis*	Apocynaceae	Minimum concentration of 25 μM to produce detectable spot in TLC	Andrade et al. (2005)

Glycosides

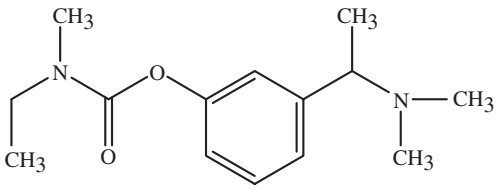
Name of glycoside	Class	Sources	Plant family	Activity	References
Cynatroside A	Pregnane glycoside	*Cynanchum atratum*	Asclepiadaceae	50% inhibition at 6.4 μM	Lee et al (2003)
Cynatroside B	Pregnane glycoside	*Cynanchum atratum*	Asclepiadaceae	50% inhibition at 3.6 μM	Lee et al (2003)
Norswertianolin	Bellidin 8-*O*-*β*-glucopyranoside	*Gentiana cambpestris*	Coniferae	Minimum concentration of 1.20 nM to produce detectable spot in TLC	Urbain et al. (2004)
Swertianolin	Bellidifolin 8-*O*-*β*-glucopyranoside	*Gentiana cambpestris*	Coniferae	Minimum concentration of 0.18 nM to produce detectable spot in TLC	Urbain et al. (2004)

Flavonoids, Xanthones, Stilbene oligomers and others

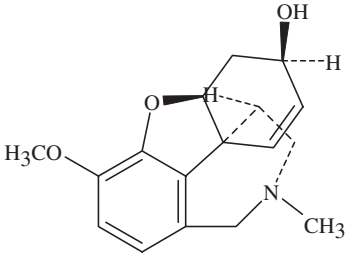
Name of compound	Class	Sources	Plant family	Activity	References
(+)-α-Viniferin	Stilbene oligomer	*Caragana chamlague*	Leguminosae	50% inhibition at 2.0 μM	Sung et al. (2002)
Bellidin	Xanthone	*Gentiana cambpestris*	Coniferae	Minimum concentration of 0.03 nM to produce detectable spot in TLC bioassay	Urbain et al. (2004)
Bellidifolin	Xanthone	*Gentiana cambpestris*	Coniferae	Minimum concentration of 0.15 nM to produce detectable spot in TLC	Urbain et al. (2004)
Ursolic acid	Hydroxy-heptamethyl-icosahydropicene carboxylic acid	*Origanum majorana*	Lamiaceae	50% inhibition at 7.5 nM	Chung et al. (2001)



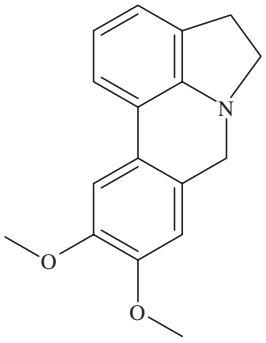
Physostigmine [1]



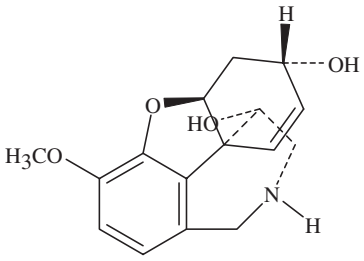
Rivastigmine [2]



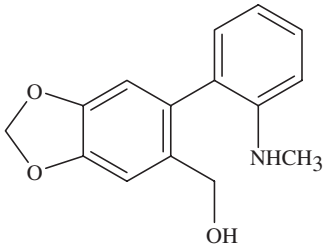
Galantamine [3]



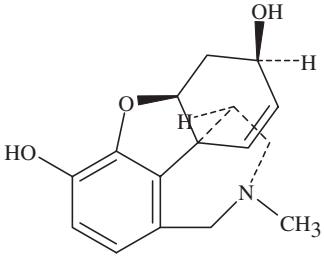
Assoanine [4]



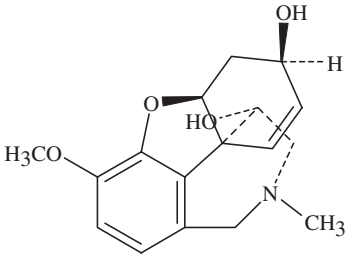
Epinorgalantamine [5]



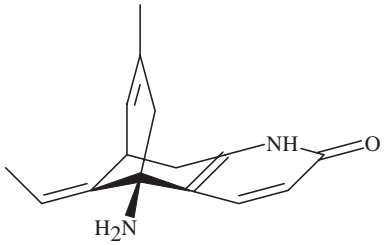
Oxoassoanine [6]



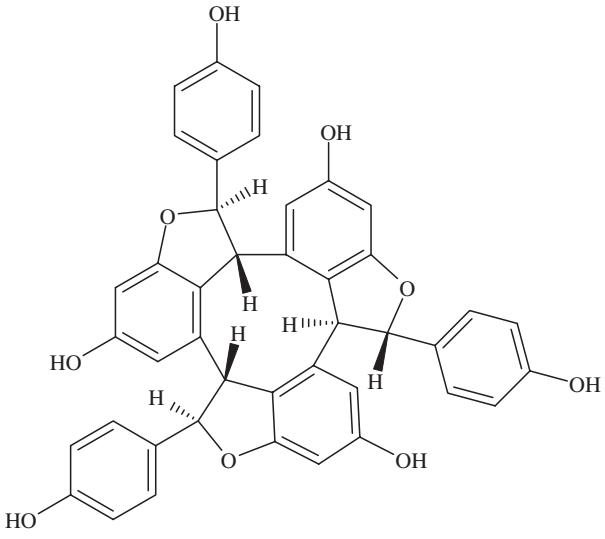
Sanguinine [7]



11-Hydroxygalantamine [8]



(-)-Huperzine A [9]



(+)-α-Viniferin [10]

Fig. 1. Structures of some acetylcholinesterase inhibitors obtained from plants.

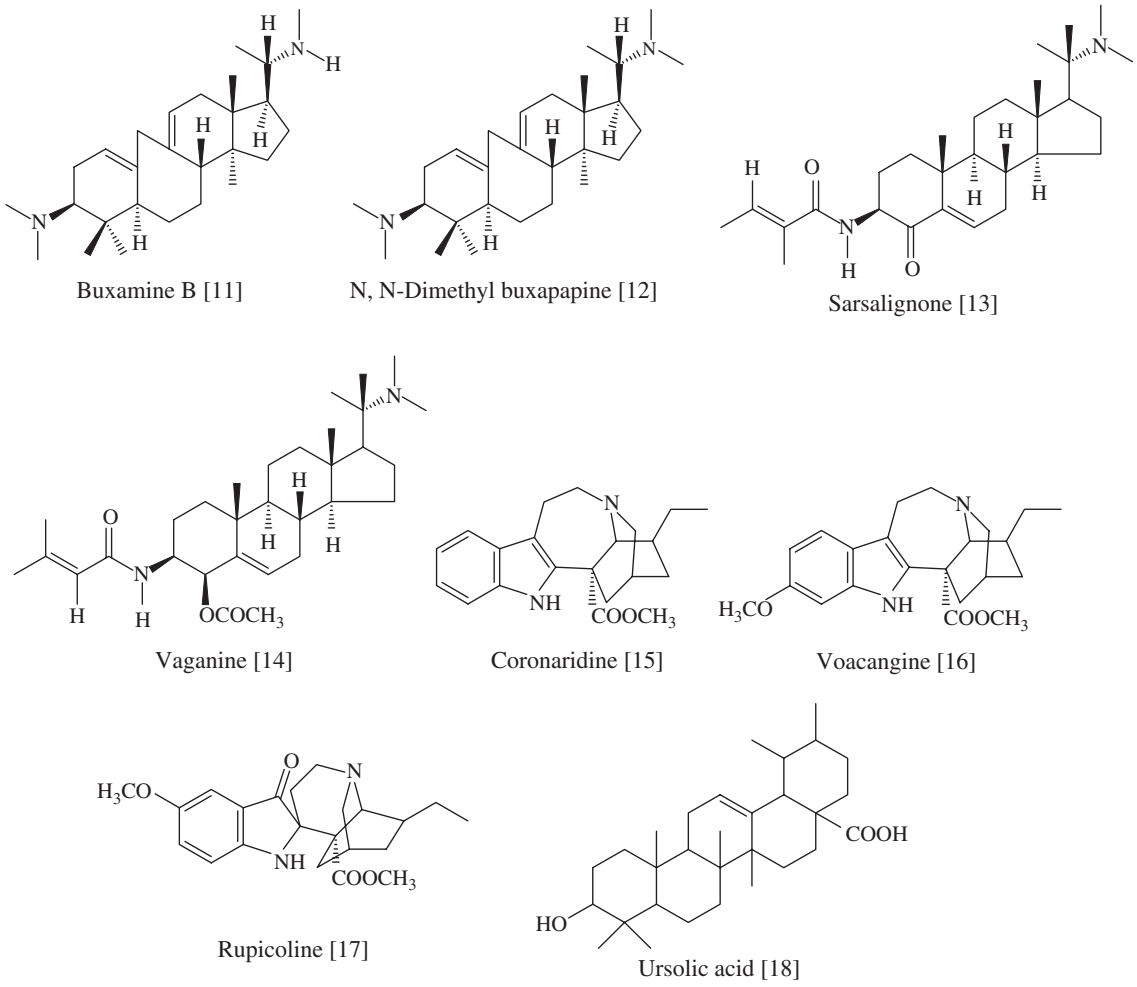


Fig. 1. (*Continued*)

buxapapine [12], sarsalignone [13] and vaganine [14] (Rahman and Choudhary, 2001). Indole alkaloids coronaridine [15], voacangine [16], voacangine hydroxyindolenine and rupicoline [17] isolated from the chloroform extract of stalk of *Tabernaemontana australis* showed anti-cholinesterasic activity at the same concentration as the reference compounds physostigmine and galantamine, by thin-layer chromatography assay using the modified Ellman's method (Andrade et al., 2005). Ursolic acid [18] obtained from *Origanum majorana* has also been reported to possess AChE inhibitory activity (Chung et al., 2001).

Conclusion

Acetylcholinesterase (AchE) inhibitors have therapeutic applications in Alzheimer's disease (AD), senile dementia, ataxia, myasthenia gravis and Parkinson's disease. Central cholinergic system is considered as the most important neurotransmitter system involved in the regulation of cognitive functions. Cholinergic neuronal loss in hippocampal area is the major feature of AD and enhancement of central cholinergic activity by use of anti-cholinesterase is presently the mainstay of the pharmacotherapy of senile dementia of Alzheimer type (Enz et al., 1993; Siddiqui and Levey, 1999). The search for plant derived inhibitors of AChE has accelerated in view of the benefits of these drugs not only in the treatment of AD but in other forms of dementia, such as dementia with Lewy bodies (Perry et al., 1994), vascular dementia (Erkinjuntti et al., 2002) and Down's syndrome (Kishnani et al., 1999). Along with the prototype inhibitor of AChE physostigmine, obtained from the plant *Physostigma venenosum*, other molecules with highly significant anti-cholinesterase activity are huperzine-A, galantamine, α-viniferin and ursolic acid obtained from *Huperzia serrata*, *Galanthus nivalis* and *Narcissus* sp., *Caragana chamlague* and *Origanum majorana*, respectively.

Majority of studies have focused on the anti-cholinesterase alkaloids, such as physostigmine and galantamine. So far, more than 35 alkaloids have been reported to have AChE inhibitory activity. The other

major classes of compound reported to have such activity are the terpenoids, glycosides and coumarins. Plants belonging to families Acanthaceae, Apocynaceae, Amaryllidaceae, Angelicae, Araceae, Asclepiadaceae, Berberidaceae, Buxaceae, Combretaceae, Compositae, Coniferae, Cyperaceae, Ebenaceae, Ericaceae, Euphorbiaceae, Fumariaceae, Gentianaceae, Guttiferae, Lamiaceae, Leguminosae, Lilliaceae, Lycopodiaceae, Malvaceae, Magnoliaceae, Menispermaceae, Molluginaceae, Moraceae, Musaceae, Nelumbonaceae, Papaveraceae, Piperaceae, Rubiaceae, Rutaceae, Sapotaceae, Solanaceae and Tamaricaceae have been reported to have AChE inhibitory potential. For many of the plants and compounds that have demonstrated activities anticholinesterase activity relevant to AD therapy, the clinical data are very limited. Clinical efficacy and potential toxicity of active plants and compounds in larger trials requires further assessment, before recommendations concerning their routine use can be identified.

Acknowledgements

The authors wish to express their gratitude to the Commonwealth Scholarship Commission, Association of Commonwealth Universities, UK, for the Commonwealth Academic Staff Fellowship Award to Dr. Pulok K. Mukherjee through the selection made from the University Grants Commission (UGC), India. Thanks are also due to the Department of Biotechnology (DBT), Government of India for providing financial assistance through research project to the School of Natural Product Studies, Jadavpur University.

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