**GINKGO BILOBA A SOURCE OF BIOACTIVE NATURAL PRODUCTS: A REVIEW**


1Natural Product Microbe Division, Indian Institute of Integrative Medicine (CSIR) Canal Road, Jammu. Pin: 180001 *
2Department of Pharmaceutical chemistry, Bhupal Nobles' College of Pharmacy Udaipur, Rajasthan-313001
3Department of medicinal chemistry, P.S.I.T, Kanpur

**ARTICLE INFO**

**Article history**
Received 06/08/2014
Available online
13/12/2014

**Keywords**
Arylidene, Ginkgo biloba, Bioactive natural products.

**ABSTRACT**
Ginkgo biloba, a prolific source of structurally diverse bioactive metabolites has provided some of the most important products to the pharmaceutical industry. It is known to produce a diverse array of compounds ranging from flavonoids, lactones, polyprenols, alkyl phenols, carotenoids, carbohydrates, hydrocarbons, organic acids, polyprenols, and steroids, glycolipids, have been identified as constituents of ginkgo leaves. The structural diversity of these complex compounds reasoned for its potent and interesting biological activities like anti-tumor activity, anti-microbial activity, antifungal activity, scavenging free radical, lowering platelets aggregation, improvement in blood flow and lowering neurological effects etc. Ginkgo is often used for memory disorders including Alzheimer’s disease. It is also used for conditions that seem to be due to reduced blood flow in the brain, especially in older people. These conditions include memory loss, headache, ringing in the ears, vertigo, difficulty concentrating, mood disturbances, and hearing disorders.

**Corresponding author**
Sandeep Kumar Kushwaha
Natural Product Microbe Division,
Indian Institute of Integrative Medicine (CSIR) Canal Road.
Jammu. Pin: 180001
Email-sandeep.kush44@gmail.com
Introduction
Nature has been always a plentiful source of very simple to extremely complex structured molecules which plays an important role in human dietary function as well as in prevention of human disease. The World Health Organization (WHO) estimated in 1985 that approximately 65% of the population of the world predominately relied on plant-derived traditional medicines for their primary health care, while plant products also play an important, though more indirect role in the health care systems of the remaining population who mainly reside in developed countries. There are written evidence where the herbs and potions are being used by ancient civilizations of the Chinese, Indians and North Africans for treatment of various diseases. Furthermore many commonly used herbal dietary supplements in the United States are prepared by using herbal plants. It is estimated that there are approximately 1500 herbal plants used as herbal dietary supplements or ethnic traditional medicines. The U.S. National Toxicology Program (NTP), which performs research focused on the most critical public health issues, has been conducting a series of long-term studies on the toxicity of herbal medicines and related dietary supplements products nominated by the public and Federal agencies. They have found that Echinacea, golden seal, ginseng, kava, ginkgo and aloe vera are among the most widely sold herbal dietary supplements in the United States, where Ginkgo biloba extract is listed as the fifth or sixth most frequently used herbal dietary supplement in the United States, and the third best-selling herbal product in health food stores in the United States in 1997.

History:
Ginkgo biloba (also known as maiden hair tree) is a unique tree sharing the history from last 150 million years. It has been flourished during the Jurassic and Cretaceous periods and is one of the best known examples of a living fossil. Ginkgo tree is not only a living fossil but also a great example of “survivor” and “bearer of hope.” This is because of its endurance power during the bombing of Hiroshima and Japan when nearly all the plants in the affected territory got died. The Ginkgo survivors were not only retained but also went on to produce buds without major distortions. This survival is owed to its remarkable adaptability, unusual resistance to pests such as insects, bacteria, viruses and fungi. The existence of Ginkgo tree was also got affected by advancement of civilization which nearly led them to extinction, but thanks to Chinese monk who planted it around their temples. This may be due to the reason that they considered it as a sacred tree. It is also found that Ginkgo tree when exposed to high temperature it secretes a sap having the fire retardant properties. This also serves as a reason of its high availability around the Buddhist temples throughout China and Japan. Now days Ginkgo biloba trees are cultivated in large plantations all over the world including China, France, USA, Asia, Europe, North America, New Zealand, and Argentina1 with an annual output of 8000 tonnes of leaves. As a source of natural products, a variety of compounds has been isolated from ginkgo leaves such as flavonoids, lactones, polypropenols, alkyl phenols, carotenoids, carbohydrates, hydrocarbons, organic acids, polypropenols, and steroids, glycolipids, have been identified as constituents of ginkgo leaves. These constituents are responsible for its diverse pharmacological activities like improvement in neurological disorder, 

Isolation of molecules:
The isolation of three compounds i.e. isophytol (1), nerolidol (2), and linalool (3) from the leaves of Ginkgo biloba. They further investigate their antibacterial and antifungal potential at animal and plant pathogenic strains (Salmonella enterica, Staphylococcus aureus and Aspergillus niger) on the basis of their inhibition halos. All the isolated compounds were found to inhibit the growth of tested strains, where nerolidol showed the highest activity in the range of 3.9–15.6 μg/mL (MIC), 31.3–62.5 μg/mL (MBC) and 62.5 μg/mL (MFC). Nerolidol was also found to have antifungal activity against T. mentagrophytes which may result in irreversible cellular disruption.5

The isolation of two new fatty alcohols (7S,8R,11S)-nonacosanetriol (4) and (10R,12R,15S)-nonacosanetriol (5) from the petroleum ether extract of Ginkgo biloba sarcotesta. Since fatty alcohols, fatty acids, and their esters are already well known for their many biological and pharmacological functions including inhibiting cholesterol synthesis, increasing low-density lipoprotein processing, anti-thrombosis and anti-platelet aggregation activity.6 Compounds 4 and 5 were also found to have the potential to prolong thrombin time and it is well known that Platelet activation could be inhibited by inhibiting thromboxane A₂ pathway, ADP pathway, thrombin
and phosphodiesterase. In the present article author studied that Platelet aggregation induced by ADP in vitro, can be moderately reversed by using compounds 4 and 5. This may also be due to the direct activation of thrombin by compounds 4 and 5 to attain the antithrombin effect which is another way to exhibit the antiplatelet activity. Notably this was the first report regarding the anticoagulative activities of biflavones in Ginkgo biloba.

The isolation of two new diterpenoid compounds, ginkgolide P (6) and ginkgolide Q (7) from the leaves of Ginkgo biloba. The compounds were tested against platelet aggregation induced by platelet activating factor (PAF). Compounds 6 and 7 showed inhibitory activities with IC$_{50}$ values of 15.65 ± 2.94, 11.01 ± 2.73, 3.33 ± 0.92, and 0.57 ± 0.15 µM, respectively.

Two polymeric proanthocyanidins epigallocatechin-(4β→8)-epigallocatechin-(4β→2)-phloroglucinol (8), epigallocatechin-(4β→6)-epigallocatechin-(4β→2)-phloroglucinol (9) has been isolated by Qadan et al. from the leaves of Ginkgo biloba by in 2011. Both compounds possess a potent antioxidant activity having 37.7% of the total of this leaf extract.

The ginkgolide N [(1,7,10-trihydroxy-3,14-dehydroginkgolide), a novel ginkgolide (10) from the leaves of Ginkgo biloba in 2009. Ginkgolide N is well known for its various biological activities. Ma et al. have described that ginkgolide N can protect PC12 cells injury induced by glutamate. Ginkgolide N was also found to reduce the LDH supernatant fluid of serum which resulted in improved survival rates. In the next year same group$^{viii}$ showed that when GN was given to PC12 cells of 6 models, it shows the protective effect which may be related to NO toxic injury and calcium overload during later stage of injury. Also Shian and Coworkers$^{viii}$ had that GN reduces the content of brain water, infarction area percentage, content of MDA and Ca$^{2+}$ level which may led to the protection of rats from cerebral focal ischemia and reperfusion injury.
The isolation of a novel polyhydroxylated C29-sterol, 25-ξ-methyl-22-homo-5α-cholest-7,22-diene-3β,6β,9α-triol, (11) from the cultures of an endophytic fungus, *Chaetomium globosum* ZY-22 originated from the *Ginkgo biloba*. Notably the molecule is mimic of naturally occurring steroidal metabolites where it contains structurally unique 25-methyl C10-side chain. In addition, they report a wide range of polyhydroxy ergostane-type sterols from the extract.

The isolated 5-(3-hydroxypropyl)-6-methoxy-2-(3’-methoxy-4-hydroxy)-3-benzo[b]furancarbaldehyde (12) from the bark of *Ginkgo biloba* having anti platelet activity.

Ginkbilobin-2 (13) had been isolated by Takuya and co-workers5 from the seeds of *Ginkgo biloba*. It consists of 108 amino acids with three disulfide bonds. The tertiary structure of Gnk2 was composed of two α-helices and a five-stranded β-sheet, having a compact single-domain architecture was over expressed in the periplasm of E. coli BL21 (DE3) by a co-expression system using pET-26b vector (Novagen) and pDsbABCD1 vector encoding the four Dsb proteins that catalyze the formation and isomerization of disulfide bonds. It also possesses strong antifungal activity5 toward *F. oxysporum*. Many phytopathogenic fungi including *F. oxysporum* produces extracellular aspartic proteases and found to be active at acidic pH. Accordingly, this protein seems to have a great significance in reducing risk of attack by proteases which make it quite evident that the inhibition of fungal growth is due to inhibition of a single hydrolytic enzyme. It was also found to be a inhibitor of aspartic protease pepsin.

In the same year Peng et al10 described the isolated of three new flavonoids kaempferol-3-O-rhamnoside-7-O-(6-feruloylgluco-(1–3)-rhamnoside), quercetin-3-O-rhamnoside-7-O-(6-feruloylgluco-(1--3)-rhamnoside), and isorhamnetin-3-O-β-D-glucosyl-(1–2)-α-L-rhamnoside (14-16) from leaves of *Ginkgo biloba*.
The isolation of two new biflavone glucosides, ginkgetin 7''-O-β-D-glucopyranoside (17) and isoginkgetin 7-O-β-D-glucopyranoside (18) from the leaves of *Ginkgo biloba* after refluxing them in MeOH.

3-Nonadec-8-enyl-benzene-1,2-diol (19) isolated from leaves of *Ginkgo biloba*. It is found to be a inhibitor of CYP2C9 having *in vitro* IC$_{50}$ values of 2.1±0.7 against human cancer cell line.

The isolated three new compounds 7E-2β,3α-dihydroxy-megastigm-7-en-9-one, 3-[5,7-dihydroxy-2-(4-methoxyphenyl)-4-oxo-4H-chromen-8-yl]-4-methoxy benzoic acid and 4'-O-methyl myricetin 3-O-(6-O-α-L-rhamnopyranosyl)-β-D-glucopyranoside acid (20-22) from leaves of *Ginkgo biloba* after extracting with acetone at room temperature. They possessed potent antioxidant activity, when evaluated on intracellular reactive oxygen species in HL-60 cells. 21 and 22 compound showed IC$_{50}$ value of 18µg/mL and 12µg/mL respectively.

Two coumaroyl flavonol glycosides, isorhamnetin 3-O-α-L-[6'''-p-coumaroyl-(β-D)-glucopyranosyl-(1,2)-rhamnopyranoside], and kaempferol 3-O-α-L-[6'''-p-coumaroyl-(β-D)-glucopyranosyl-(1,2)-rhamnopyranoside]-7-O-β-D-glucopyranoside, (23, 24) were isolated from the n-BuOH extract of *Ginkgo biloba* leaves by Yuping et al. Both compounds showed profound antioxidant activities in DPPH and cytochrome-c reduction assays using the HL-60 cell culture system. For compound 23, IC$_{50}$ value for DPPH assay and cytochrome-c reduction assays are 17.5 and 17.5. for compound 24, theses values are 18.4 and 17.9.
In the same year Wang et al. reported the isolation of isolation of doubly bonded ginkgolide 1, 10-dihydroxy-3, 14-didehydroginkgolide and 10-hydroxy-3, 14-didehydroginkgolide, named as ginkgolide K and ginkgolide L, (25, 26) respectively. It was found that it helps in increasing cells viability of H₂O₂ exosed cells on treatment with with concentrations (10, 50, 100 µM) of ginkgolide K (GK). This led to the LDH release under the suppressed conditions, attenuated ROS level, prevented cytochrome-c release from mitochondria and boosted MMP expression. In addition, ginkgolide K was found to be selective in caspase-3 and caspase-9 inhibition where caspase 8 remain unaffected in exogenous H₂O₂-treated PC12 cells. These results demonstrated that ginkgolide K protected PC12 cells from H₂O₂-induced apoptosis by restoring MMP expression, ameliorating oxidative stress and subsequently leading to inhibit the activity of caspase-3 protein. These results make it as a promising candidate for neuroprotective cerebral ischemia treatment.

The isolation of Ginkbilobin, (27), the protein from Ginkgo biloba seeds. The characterisation of N-terminal amino acid sequence of ginkbilobin was analyzed by means of automated Edman degradation using a Hewlett-Packard 1000A protein sequencer equipped with an HPLC system. It was found to possess strong antifungal activity against a number of fungal species including Botrytis cinerea, Mycosphaerella arachidicola, Fusarium oxysporum, Rhizoctonia solani, and Coprinus comatus. Ginkbilobin showed moderate antibacterial activity against E. coli. Antifungal protein usually exhibited potent inhibitory activity toward some fungal species. Authors had also gone for in vivo studies where they found that Ginkbilobin inhibited translation in the cell-free rabbit reticulocyte lysate system with a low potency. This low activity comes in contrast to the high translation-inhibiting activity of ribosome inactivating proteins.

**Benefits of Ginkgo Biloba**

Ginkgo biloba’s natural health benefits have been recognized by practitioners of traditional medicine. They use the health benefits to treat circulatory disorders and enhance memory. The top five health benefits of ginkgo biloba are:

1) **Dementia and Alzheimer’s disease:** Ginkgo biloba was used originally to regulate blood flow to the brain. Further studies revealed that it may protect nerve cells damaged by Alzheimer’s disease. Many studies reveal that ginkgo biloba can improve memory and thinking in people affected by Alzheimer's or vascular dementia.

2) **Glaucoma:** According to one study, people with glaucoma who took 120 mg of ginkgo biloba daily for eight weeks realized that their vision had improved.

3) **Memory Enhancement:** Ginkgo biloba is popular as a "brain herb." Studies have proven that it can improve memory in people with dementia. Ginkgo biloba is commonly added to nutrition bars, soft drinks, and fruit smoothies to boost memory and enhance cognitive performance.

www.iajpr.com
4) **Macular Degeneration:** It is a progressive, degenerative eye disease that impairs the retina. It is one of the most common causes of blindness in the United States. The flavonoids found in ginkgo biloba may help cure or alleviate some retinal problems.

**Conclusion**

Ginkgo is marketed in dietary supplement form with claims it can enhance cognitive function in people without known cognitive problems, but such claims are unfounded as it has no effect on memory or attention in healthy people. Ginkgo has been studied as a possible treatment for dementia and Alzheimer's disease, with mixed results.

**References**
