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Phytochemistry and Pharmacological properties of F*icus religiosa*: an overview

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ABSTRACT

Ficus religiosa Linn is a large evergreen tree found throughout India, wild as well as cultivated. It is popular indigenous system of medicine like Ayurveda, Siddha, Unani and Homeopathy. In traditional system of medicine, various parts such as stem bark, root bark aerial roots, vegetative buds, leaves, fruits and latex are used in diabetes, vomiting, burns, gynaecological problems, dysentery, diarrhea, nervous disorders, tonic and astringent. Phytochemical investigation of plant barks, showed the presence tannins, saponins, flavonoids, steroids, terpenoids and cardiac glycosides. According to Ayurvedic system of medicine, F. religiosa (Peepal tree) is well known to be useful in diabetes. The present work is an attempt to compile an up-to-date and comprehensive review of F. religiosa that covers its ethnobotanical, natural product chemistry, pharmacological data.

Keywords: *Ficus religiosa*, Pharmacognosy, Phytochemistry, Medicinal application.

INTRODUCTION

Plants have played a significant role in maintaining human health and improving the quality of human life for thousands of years and have served humans well as valuable components of medicines, seasonings, beverages, cosmetics and dyes. Herbal medicine is based on the premise that plants contain natural substances that can promote health and alleviate illness. In recent times, focus on plant research has increased all over the world and a large body of evidence has collected to show immense potential of medicinal plants used in various traditional systems. Today, we are witnessing a great deal of public interest in the use of herbal remedies [1]. This review emphasizes the traditional use and clinical potentials of *Ficus religiosa.* Through which the authors hope to attract the attention of natural product researchers throughout world on the explored potential of *F. religiosa.* This review has been compiled using references from major database such as Chemical Abstracts, Medicinal and Aromatic Plants Abstracts, Pubmed, Duke's Phytochemical and Ethnobotany database, and Image database. The available information on *F.*

religiosa has been divided into four sections, which are ethnopharmacology, morphology, phytochemistry and pharmacological studies.

Ficus religiosa Linn (Moraceae) commonly known as 'Peepal tree' is a large widely branched tree with leathery, heart-shaped, long-tipped leaves on long slender petioles and purple fruits growing in pairs. The tree is regarded as a sacred tree to both Hindus as well as Buddhists. It has got mythological, religious and medicinal importance in Indian culture since ancient times [2-4]. The tree grows throughout India and widely cultivated in south-east Asia especially in vicinity of temples. In Ayurveda, *F. religiosa* belongs to a class of drugs called rasayana. Rasayana are rejuvenators, antioxidants and relieve stress in the body [5,6]. In India it is known by several vernacular names, the most commonly used ones being Asvatthah (Sanskrit), Sacred fig (Bengali), Peepal (Hindi), Arayal (Malayalam), Ravi (Telgu) and Arasu (Tamil) [7].

Ethnopharmacology

Traditional Uses

F. religiosa is a well known ethnomedicinal tree used in Ayurveda. Its use in the Indian traditional folk medicine also well documented. The use of different parts of *F. religiosa* in traditional system of medicine (Table 1)**.**

Table 1. Ethnomedicinal uses of different parts of *F. religiosa*

Plant Parts	Traditional Uses (as/in)
Bark	Astringent, cooling, aphrodisiac, antibacterial against *Staphylococcus aureus* and *Escherichia coli*, gonorrhoea, diarrhoea, dysentery, haemorrhoids and gastrohelcosis, anti-inflammatory, burns [7].
Bark Decoction	Cooling, gonorrhea, skin diseases, scabies, hiccup, vomiting [8].
Leaves and tender shoots	Purgative, wounds, skin diseases [7].
Leaf juice	Asthma, cough, sexual disorders, diarrhea, haematuria, toothache, migraine, eye troubles, gastric problems, scabies [7-9].
Leaf decoction	Analgesic for toothache [9].
Dried fruit	Tuberculosis, fever, paralysis, hemorrhoids [10].
Fruit	Asthma, laxative, digestive [7].
Seeds	Refrigerant, laxative [7].
Latex	Neuralgia, inflammations, haemorrhages [7].

Alternative and Complementary Medicinal Uses: The barks of *F. religiosa* is an important ingredient in many Ayurvedic formulations, such as *Nalpamaradi tailam, Chandanasavam, Nyagrodhadi churna* and *Saribadyasavam* [11,12].

Morphology and microscopy

Morphology: *F. religiosa* is a large deciduous tree with few or no aerial roots. It is often epiphytic with the drooping branches bearing long petioled, ovate, cordate shiny leaves. Leaves are bright green, the apex produced into a linear-lanceolate tail about half as long as the main portion of the blade. The receptacles occurring in pairs and are axillary, depressed globose, smooth and purplish when ripe. The bark is flat or slightly curved, varying from 5 to 8 mm in thickness, outer surface is grey or ash with thin or membranous flakes and is often covered with crustose lichen brown or ash coloured, surface has shallow irregular vertical fissures and uneven due to exfoliation of cork, inner surface smooth, yellowish to orange brown and fibrous [7,13]. (Figure 1)

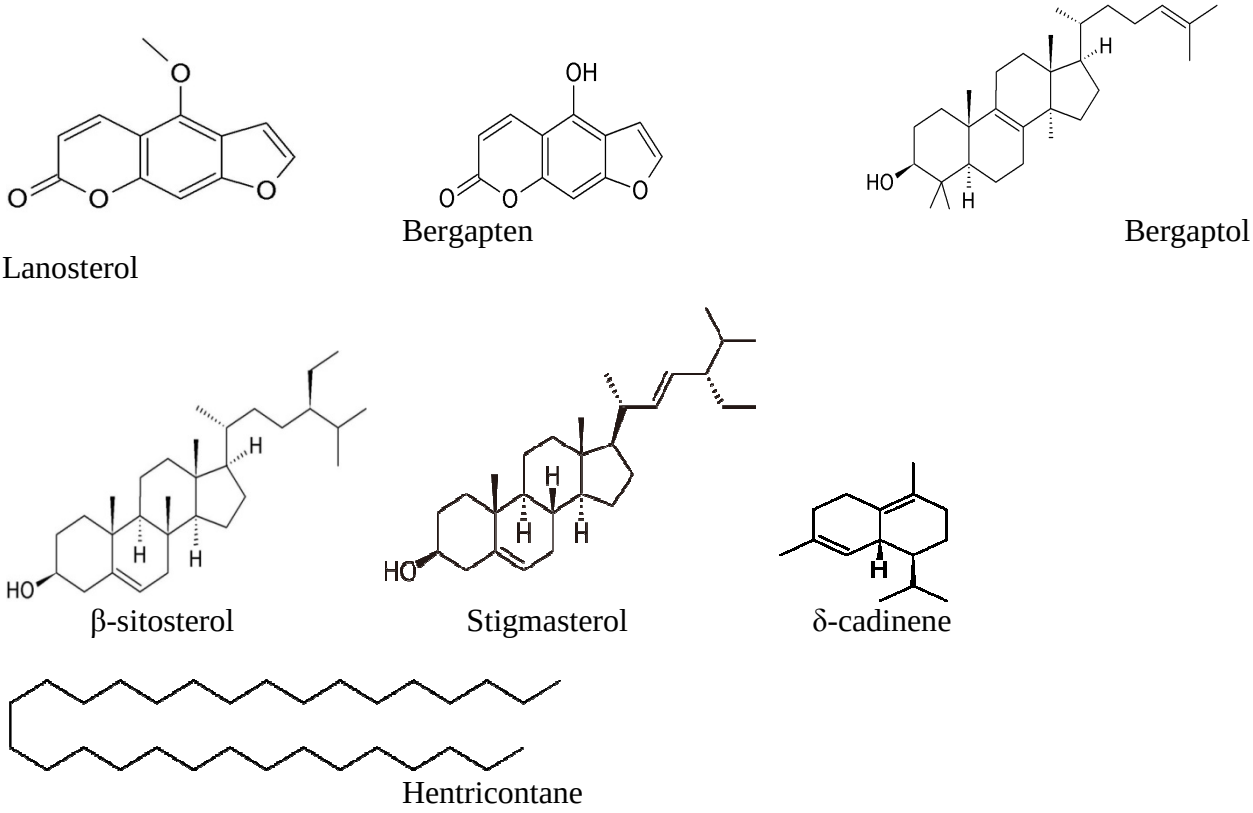
Figure 1. Morphology of *Ficus religiosa*

Microscopy: An external features of bark of *F. religiosa* showed that bark differentiated into outer thick periderm and inner secondary phloem. Periderm is differentiated into phellem and phelloderm. Phellem zone is 360 mm thick and it is wavy and uneven in transection. Phellem cells are organized into thin tangential membranous layers and the older layers exfoliate in the form of thin membranes. The phelloderm zone is broad and distinct. Phelloderm cells are turned into lignified sclereids. Secondary phloem differentiated into inner narrow non-collapsed zone and outer broad collapsed zone. Non-collapsed zone consists of radial files of sieve tube members, axial parenchyma, and gelatinous fibres. Outer collapsed phloem has dilated rays, crushed obliterated sieve tube members, thick walled and lignified fibres, and abundant tannin filled parenchyma cells. Laticifers are fairly abundant in the outer secondary phloem zone. Phloem rays are both uniseriate and multiseriate. Multiseriate rays are homocellular and uniseriate rays are either homocellular or heterocellular [13].
Physical constants: Total ash 7.86 % w/w, acid insoluble ash 0.41 % w/w, alcohol soluble extract 7.21 % w/w and water soluble extractive 15.76 % w/w [13].

Phytochemistry
Preliminary phytochemical screening of *F. religiosa* barks, showed the presence tannins, saponins, flavonoids, steroids, terpenoids and cardiac glycosides [13,14]. The barks of *F. religiosa* showed the presence of bergapten, bergaptol, lanosterol, β-sitosterol, stigmasterol, lupen-3-one, β-sitosterol-d-glucoside (phytosterolin), vitamin k_1[15-18]. The bark also contains tannin, wax, saponin, β-sitosterol, leucocyanidin-3-0-β-D-glucopyrancoside, leucopelargonidin-3-0-β-D-glucopyranoside, leucopelargonidin-3-0-α-L- rhamnopyranoside, lupeol, ceryl behenate, lupeol acetate, α-amyrin acetate, leucoanthocyanidin and leucoanthocyanin [19]. Leaves yield campestrol, stigmasterol, isofucosterol, α-amyrin, lupeol, tannic acid, arginine, serine, aspartic acid, glycine, threonine, alanine, proline, tryptophan, tryosine, methionine, valine, isoleucine, leucine, n-nonacosane, n-hentricontanen, hexa-cosanol and n-octacosan [20-22]. The fruit of *F. religiosa* contains asgaragine, tyrosine, undecane, tridecane, tetradecane, (e)-β-ocimene, α-thujene, α-pinene, β-pinene, α-terpinene, limonene, dendrolasine, dendrolasine α-ylangene, α-copaene, β-bourbonene, β-caryophyllene, α-trans bergamotene, aromadendrene, α-humulene, alloaromadendrene, germacrene, bicyclogermacrene, γ-cadinene and δ-cadinene [23]. Alanine, threonine, tyrosine have been reported in seeds of *F. religiosa* [24]. The crude latex of *F. religiosa* shows the presence of a serine protease, named religiosin. Religiosin is an acidic

protein acts optimally at pH 8.0-8.5 and temperature 50°C. The extinction coefficient (ε1%280) of religiosin is 29.47 $m^{-1}cm^{-1}$ with 16 tryptophan, 26 tyrosine, and 11 cysteine residues per molecule. The enzyme exhibits milk-clotting as well as detergent activity [25]. Reverse Phase High Performance Liquid Chromatographic analysis of flavonoids in *F. religiosa* using kaempferol, rhamnetin, myricetin, isorhamnetin and quercetin as a standards. The findings showed that quercetin was most abundant flavonol present in *F. religiosa* [26]. The structures of active constituents reported in *F. religiosa* are given in (Figure 2).

Figure 2. List the few important structure of active constituents found in *F. religiosa.*



Pharmacological activity of *f. Religiosa*

Antidiabetic activity: Aqueous extract of *F. religiosa* in a doses of 50 and 100 mg/kg shows pronounced reduction in blood glucose levels in normal, glucose-loaded hyperglycemic and streptozotocin (STZ) induced diabetic rats and effect was compared with glybenclamide, a well known hypoglycemic drug. Aqueous extract of *F. religiosa* showed significant increase in serum insulin, body weight, glycogen content in liver and skeletal muscle of STZ-induced diabetic rats, also reduced the serum triglyceride and total cholesterol level. The results suggested potential traditional use of *F. religiosa* [27]. Ambike et al. investigated that a phytosterolin isolated from *F. religiosa* root bark when given at a dose of 25 mg/kg orally to fasting rabbits produced a maximum fall of the blood sugar level, equivalent to 81% of the tolbutamide standard, after 4 hrs, while with *i.v.* injections of 5-7.5 mg/kg a maximum effect was achieved after 2 h [18].

Anti-inflammatory activity: A study was investigated for the effect of a methanol extract of *F. religiosa* leaf (MFL) on lipopolysaccharide (LPS)-induced production of NO and proinflammatory cytokines, such as tumor necrosis factor-alpha, interleukin-beta (IL) and IL-6 in BV-2 microglial cells, a mouse microglial line. MFL inhibited LPS-induced production of NO and proinflammatory cytokines in a dose-dependent manner. MFL also attenuated the expression

of mRNA and proteins of inducible nitric oxide synthase and proinflammatory cytokines, suggesting the blockage of transcription levels, respectively. The molecular mechanism of MFL-mediated attenuation underlies the down-regulation of the extracellular signal-regulated kinase, c-Jun *N*-terminal kinase and p38 mitogen-activated protein kinase signaling pathway, and suppresses the nuclear factor kappaB activation. The results suggest that MFL exhibits anti-inflammatory properties in LPS-induced activation of BV-2 microglial cells, and that might have a therapeutic potential for various neurodegenerative diseases [28].

Methanolic extract of *F. religiosa* stem bark at a doses of 125, 125 and 500 mg/kg, 30 min prior to 0.1 ml carrageenan injection (1% in 0.9% saline) into the sub plantar region of the left hind paw. Where compared using an aqueous solution of indomethacin as standard reference group. The paw volume was measured plethysmographically just before and 3h after carrageenan administration. The group treated with 3 doses of crude extract showed inhibition of oedema formation of 52.99, 55.41 and 56.29%, respectively by the 3rd h [29]. Aqueous extract of dried powdered bark of *F. religiosa* used in paw oedema, were induced by an injection of carrageenan (0.1 ml in a 1% solution) into the plantar surface of the right hind limb. Control group received phenylbutazone (100 mg/kg). For testing the effect in chronic inflammation sterile cotton pellets (10 mg) were implanted and the animals were treated with three different doses (50, 100 and 200 mg/kg) of the extract for seven days. A significant anti-inflammatory effect was observed in both acute and chronic models of inflammation; the extract also protected mast cells from degranulation induced by various degranulators [30].

Analgesic activity: Sreelekshmi *et al*. investigated the analgesic activity of the *F. religiosa* stem bark methanolic extract using the acetic acid-induced writhing (extension of hind paw) model in mice. Aspirin were used as standards drugs. It exhibited reduction in the number of writhing of 71.56 and 65.93% respectively at a dose of 250 mg/kg and 500 mg/kg. The abdominal constriction is related to the sensitization of nociceptive receptors to prostaglandins. Thus suggest that extract showed the analgesic effect probably by inhibiting synthesis or action of prostaglandins [29].

Antioxidant activity: The antioxidant activity of the aqueous extract of *F. religiosa* was investigated in streptozotocin-induced diabetic rats. Since the oxidative stress is the major cause and consequence of type 2 diabetes. Free radicals generated during oxidative stress damage the insulin receptors and thereby decrease the number of sites available for insulin function. The aqueous extract drug reported to contain tannins, flavonoids and polyphenols. At doses 100 and 200 mg/kg of aqueous extracts of *F. religiosa* shows significantly decrease in fasting blood glucose and increase in body weight of diabetic rats as compared to untreated rats. *F. religiosa* at 100 mg/kg dose decreased significantly ($p<0.05$) superoxide dismutase (SOD) and at dose of 200 mg/kg significantly enhanced catalase (CAT) ($p<0.05$) and glutathione peroxidase (GSH-Px) ($p<0.01$) activity in type 2 diabetic rats. It modulated the SOD activity in dose dependent manner. Decrease in CAT activity due to less availability of NADPH or gradual decrease in erythrocyte CAT concentration by excessive generation of oxygen radical that inactivates the enzyme. Aqueous extract restored the erythrocyte GSH. Decrease in MDA marker by *F. religiosa* showed the erythrocyte CAT concentration by excessive generation of oxygen radical that inactivates the enzyme. Aqueous extract restored the erythrocyte GSH. Decrease in MDA marker by *F. religiosa* showed the ability of rasayana drug (rejuvenators, antioxidant, and relieve stress). The results suggesting that the *F. religiosa*, a rasayana group of plant drug having antidiabetic along with antioxidant potential was beneficial in treatment of type 2 diabetes [31].

The methanolic extract of *F. religiosa* showed significant antiradical activity by bleaching 1, 1-diphenyl-2picrylhydrazyl (DPPH) radical, EC_{50} from 11.75 µg was comparable to pyrogallol. It showed good superoxide scavenging potential, EC_{50} from 50.65 µg comparable to that of ascorbic acid and maximum reductive potential at a concentration of 400 µg, which was comparable to that of gallic acid and tannic acid. These finding suggest the rich phytochemical content of *F. religiosa* have good antioxidant activity [14].

The ethanolic extract was subjected to screen for antioxidant activity using DPPH radical scavenging method. The percentage peroxide value for *F. religiosa* extract was found in the range of 6.34% to 13.35% for the extract with 200 µg/ml to 1000 µg/ml strength extract [32]. A study was carried to localize the oxidative stress enzymes, peroxidase and catalase; and to quantify the main reactive oxygen species, hydrogen peroxide in *F. religiosa*. The results explained that plants grown in adverse habitat showed 55% higher H_2O_2 production with about 30% increase in peroxidase activity. The three substrates tested for peroxidase activity (guaiacol, ascorbate and *o*-dianisidine), *o*-dianisidine was most preferred substrate of *F. religiosa*. Cytosolic peroxidase activity showed eleven fold increase over cell wall bound peroxidase. Similarly, catalase activity in specimens from adverse habitat showed about two fold increase during day time [33].

Anticonvulsant activity: An exhaustive study was performed on figs (fruit) of *F. religiosa* showed promising anticonvulsant activity in experimental model were seizure induced by maximum electroshock (MES), picrotoxin and pentylenetetrazol (PTZ). Along with cyproheptadine, a nonselective serotonin antagonist (4 mg/kg, i.p.) was used to study the reversal of protective effect of extract in the above mentioned models. Acute toxicity, neurotoxicity and potentiation of phenobarbitone induced sleep by extract were also studied. The highest amount of serotonin (5-HT) in figs of this plant as compare to figs of other species reported in Ayurveda. Furthermore serotonergic neurotransmission is known to modulate a wide variety of experimentally induced seizure and is involved in seizure protection in various animal models of epilepsy by altering various GABAnergic and glutamatergic functions. *F. religiosa* was measured by using high-performance liquid chromatography (HPLC). The analysis was carried out at 277 nm using 5 µm particles Hypersil GOLD C-18 RP column as a stationary phase. While 25mM phosphate buffer (pH 2.5) and acetonitrile used as mobile phase at flow rate of 1ml min^{-1}. It showed an absorption peak with a retention time of 12.563 min same retention time as that of standard serotonin solution. Hence indicate the presence of serotonin in the extract. The study showed significant decrease in the duration of tonic hind limb extension in a dose dependent manner in MES model. In picrotoxin model study extract caused a delay of the latency to clonic convulsions and activity was found to be euipotent as that of diazepam treated group at a dose of 100 mg/kg. The extract showed no protection against PTZ induced convulsion at any dose. Pretreatment with cyproheptadine showed inhibition of anticonvulsant effect of the extract in both models (MES and picrotoxin). Further the extract showed no mortality and behavioural changes in acute toxicity model. This study used rotarod test to determine neurotoxic effect like ataxia, abnormal gait, reduced or inhibited righting reflexes and muscle relaxation. The extract showed no neurotoxicity and all animals able to maintain equilibrium on rotaing rod for more than 3 min. The results confirm and justify the use of *F. religiosa* in ethnomedical treatment of epilepsy and presence of high serotonin content in its figs led to hypothesize that figs of *F. religiosa* may possess anticonvulsant properties via modulating brain serotonin levels, which will be of clinical usefulness [3].

Antimicrobial activity: The antimicrobial activity of ethanolic extracts of *F. religiosa* (leaves) was examined using the agar well diffusion method. The test was performed against four

bacteria: *Bacillus subtilis* (ATCC 6633), *Staphylococcus aureus* (ATCC 6538), *Escherichia coli* (ATCC 11229), *Pseudomonas aeruginosa (*ATCC 9027) and against two fungi: *Candida albicans* (IMI 349010) and *Aspergillus niger* (IMI 076837). The results showed that 25mg/ml of the extract was active against all bacterial strains and effect against the two fungi was comparatively much less [34]. The antibacterial activity of different extracts from the bark of *F. religiosa* was tested against diarrhoeal enterotoxigenic *Escherichia coli* using disc diffusion method. The antibacterial activities of extract were compared with standard antibiotics. The sensitivity of the organisms measured in terms of zone of inhibition ranged from 8.00 to 14.00 mm at 4mg/ml of different extract. The results revealed that methanol extract exhibits good activity compared to chloroform and aqueous extract. Petroleum ether and hexane extract did not show any activity [35].

Broad-spectrum antibacterial and antifungal properties of Ethanolic extracts of 22 traditionally used Indian medicinal plants were studied for their antimicrobial activity against seven bacteria (*Staphylococcus aureus, Salmonella typhimurium, S. paratyphi, S. typhi, E. coli, Shigella dysenteriae and Pseudomonas aeruginosa*) and five filamentous fungi (*Aspergillus niger, Alternaria alternata, Fusarium chlamydosporum, Rhizoctonia bataticola* and *Trichoderma viride*). Of these, 16 plant extracts showed varied level of antibacterial activity against one or more test bacteria. Similarly antifungal activity was detected among 17 plant extracts respectively. *F. religiosa* (leaves) demonstrated more antibacterial activity with less antifungal activity [36]. Zaidi et al. reported the potential *in vitro* anti-*Helicobacter pylori* activity of medicinal plants from Pakistan that is used to cure GI disorders. *Helicobacter pylori* was isolated from the antral biopsy specimens and confirmed through the standard microbiology procedures. The 70% aqueous-ethanol extracts of *Ficus religiosa* completely inhibited the growth of *Helicobacter pylori* at 500µg/ml in all strains and demonstrate anti-*Helicobacter pylori* activity with MBC value ranged from 125 to 250µg/ml [37].

Iqbal et al. investigated that *F. religiosa* bark methanolic extract was 100% lethal for *Haemonchus. contortus* worms during *in vitro* testing [38]. The acetone extracts of seven plant species *Tamarindus indica, F. indica, F. religiosa, Tabernaemontana livaricate, Murraya koenigii, Chenopodium album* and *Syzygium cuminii* were evaluated for their ovicidal activity. *Murraya, Tabernaemontana* and *Chenopodium* showed 70%, 75% and 66.6% ovicidal action at 100% dose level whereas at the same dose level *T. Indica, F. indica, F. religiosa and S. cuminii* showed 48.3%, 41.6%, 13.3%, 53.3% ovicidal action respectively [39]. The preliminary screening of antibacterial activity of *F. religiosa* by agar-well diffusion assay was investigated. The chloroform extracts of *F. religiosa* showed a strong inhibitory activity against growth infectious *Salmonella typhi, Salmonella typhimurium* and *Proteus vulgaris* at a MIC of 39, 5 and 20 µg/ml respectively [40].

Wound healing activity: The wound healing activity was investigate by excision and incision wound models using *F. religiosa* leaf extracts, prepared as ointment (5 and 10%) were applied on Wistar albino strain rats. Povidine iodine 5% was used as Standard drug. High rate of wound contraction, decrease in the period for epithelialisation, high skin breaking strength were observed in animals treated with 10% leaf extract ointment when compared to the control group of animals. It has been reported that tannins possess ability to increase the collagen content, which is one of the factor for promotion of wound healing [41,42].

Anti-amnesic activity: The anti-amnesic activity was investigated using *F. religiosa* methanol extract of figs of *F. religiosa* on scopolamine-induced anterograde and retrograde amnesia in mice. Figs were known to contain a high serotonergic content, and modulation of serotonergic

neurotransmission plays a crucial role in the pathogenesis of amnesia. During study, transfer latency (TL) to the preferred niche in the elevated plus-maze (EPM) and learning avoidance of passive behavior to avoid punishment in the modified passive avoidance paradigm (MPA) served as behavioral models for the assessment of memory. Scopolamine (1 mg/kg, i.p.) was administered before training for induction of anterograde amnesia and before retrieval for induction of retrograde amnesia in both models. TL in the EPM, step down latency (SDL), number of trials, and number of mistakes in the MPA were determined in vehicle control, *F. religiosa* figs treated (10, 50, and 100 mg/kg, i.p.), and standard groups (piracetam 200 mg/kg, i.p.). Cyproheptadine, a non-selective 5-$HT_{1/2}$ blocker (4 mg/kg, i.p.), was administered along with the *F. religiosa* figs to investigate the involvement of serotonergic pathways in the anti-amnesic effect of *F. religiosa* figs. The resulted had anti-amnesic activity against scopolamine-induced amnesia, in a dose-dependent manner. Inhibition of the anti-amnesic effect of *F. religiosa* figs by cyproheptadine substantiates the involvement of serotonergic pathways for its activity [43].

Anti-acetylcholinestrase activity: Methanolic extract of the stem bark of *F. religiosa* found to inhibit the acetylcholinestrase enzyme, thereby prolonging the half-life of acetylcholine. It was reported that most accepted strategies in alzheimer's diseases treatment is the use of cholinesterase inhibitors. The calculated 50% inhibitory dose (ID50) value was 73.69 µg/ml respectively. The results confirm and justify the popular traditional use of this plant for the treatment of alzheimer's diseases [44].

Proteolytic Activity: A comparison of the proteolytic activity of the latex of 46 species of *Ficus* was done by electrophoretic and chromatographic properties of the protein components. *F. religiosa* showed significant proteolytic activity [45].

CONCLUSION

Ficus religiosa is a widely branched tree with leathery, heart-shaped, long-tipped leaves, used in the Indian system of medicine, besides which folklore medicine also claims its use in diarrhoea, diabetes, urinary disorder, burns, haemorrhoids, gastrohelcosis, skin diseases, convulsion, tuberculosis, fever, paralysis, oxidative stress, bacterial infection etc. Research carried out using different in-vitro and in-vivo techniques of biological evaluation support most of these claims.

Presently there is an increasing interest worldwide in herbal medicines accompanied by increased laboratory investigation into the pharmacological properties of the bioactive ingredients and their ability to treat various diseases. Numerous drugs have entered the international through exploration of ethnopharmacology and traditional medicine. Although scientific studies have been carried out on a large number of Indian botanicals, a considerably smaller number of marketable drugs or phytochemical entities have entered the evidence-based therapeutics. Efforts are therefore needed to establish and validate evidence regarding safety and practices of Ayurvedic medicines [46-49].

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