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Spectrum of Pharmacological Activities from *Bauhinia variegata*: A Review

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ABSTRACT

Bauhinia variegata Linn., commonly known as 'Camel's foot tree or Orchid tree', belonging to the most primitive subfamily (Caesalpinoideae) of the Leguminosae family is widely distributed in most tropical countries and have been used frequently in folk medicine for varied purposes; to treat different kinds of pathologies, particularly diabetes, infections, as well as pain and inflammation. The chemical composition and biological potential of the plant is widely studied. Results of preclinical studies have suggested its possible applications in clinical research.*B. variegata* appears to be a promising therapeutic agent and can certainly play an important role in the discovery of new and effective medicinal agents.

Key words: *Bauhinia variegata* (BV), Flavanoids, Lectin, Biological properties.

INTRODUCTION

There are about one hundred species under the genus Bauhinia (Caesalpiniaceae) and eight are native to India. *B.variegata* Linn., commonly known as 'Kachnar', is a small sized deciduous tree with dark brown and more or less smooth bark, up to 8m tall; propagated by seed. The roots and bark are astringent, acrid, cooling, constipating, depurative, anthelmintic, vulnerary, anti-inflammatory and styptic. They are useful in vitiated conditions of pitta and kapha, diarrhoea, dysentery, skin diseases, leprosy, intestinal worms, tumours, inflammations, scrofula, proctoplosis, haemorrhoids, haemoptysis, cough, menorrhagia and diabetes.

Root decoction is used in dyspepsia and flatulence and act as an antidote to snake poison.

Plant extract act as an effective measure for controlling insect pest like *Plutella xylostella. B.variegata var. candida* is a promising source of edible wild vegetable flowers with plenty of nutrients. This plant may serve as a potential source for low cost proteins.The tree is susceptible to 'Brown Root Rot' caused by *Phellinus noxius*.[15] The abundance of phytophagous mites is higher, being *Lorryia Formosa* Cooreman the dominant species.[83]

PHYTOCHEMISTRY

The stem bark is reported to contain 5,7 dihydroxy and 5,7 dimethoxy flavanone-4-O-a-L rhamnopyrosyl-ß-D-glycopyranosides, Kaempferol-3-glucoside, lupeol, and betasitosterol. Seeds contain protein, fatty oil-containing oleic acid, linoleic acid, palmitic acid, and stearic acid. Flowers contain cyanidin, malvidin, peonidin, and kaempferol. Root contains flavanol glycosides.

A new flavone glycoside, 5-hydroxy7,3',4',5'-tetra-methoxyflavone5 -0-ß-D-xylopyranosyl (1 → 2)-α-L-rhamnopyranoside, has been isolated from 95% ethanolic extract of seeds (1).[103] The flavonoid content includes Apigenin, Luteolin, Quercitrin and Rutin but not Quercitin.[11]

B. variegata lectin (BVL) has been purified from the seeds with a pattern similar to other lectins isolated from the same genus, *B. purpurea* agglutinin (BPA).[38] Seeds and seedlings have the same lectin.[37] A new phenanthraquinone 'Bauhinione' (2) has been isolated from the stems.[109]

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No alkaloids and unbound anthraquinones while glycosides; cardiac glycosides, and Flavanoids are present.[81] Phytochemical analysis of the root bark yielded a new flavanone, (2S)-5,7-demethoxy-3',4'-methylenedioxy flavanone (3) and a new dihydrodibenzoxepin, 5,6-dihydro-1,7-dihydroxy-3,4-dimethoxy-2-methyldibenz[b,f]oxepin (4) together with three known Flavanoids.[82] The structures of the new compounds were elucidated on the basis of spectral studies.

PHARMACOLOGICAL PROFILE:

1.Antioxidant

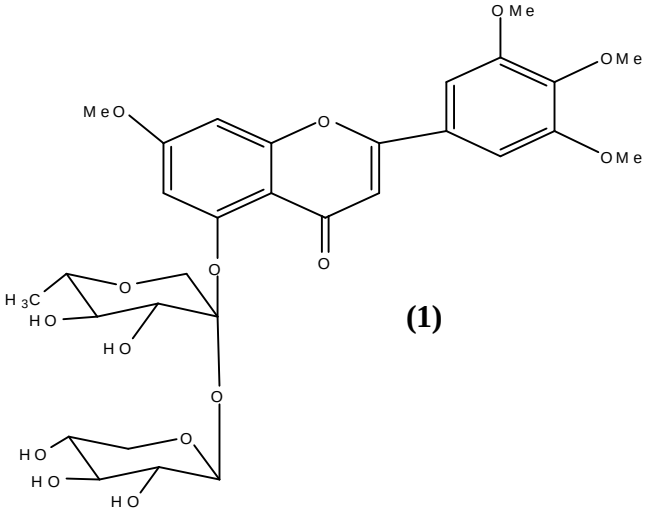
The aqueous and ethanolic extracts of *B. variegata* L. have shown significant antioxidant activity.[54] The % free radical scavenging activity gradually increases with increasing concentrations of *B. variegata* extracts in DPPH radical scavenging assay. Dose dependent antioxidant activity pattern was also observed in phosphomolybdate assay. Antioxidant activity was directly correlated with the amount of total phenolic contents in the extracts. *B. variegata* in L-dopa extract has shown the highest FRAP values.[30]

2.Anti-inflammatory

A new triterpene saponin, named as 23-hydroxy-3alpha-[O-alpha-L-1C4-rhamnopyranosyl-(1"→ 4')-O-alpha-L-4C1-arabinopyranosyl-oxy]olean-12-en-28-oic acid O-alpha-L-1C4-rhamnopyranosyl-(1""→ 4"")-O-beta-D-4C1-glucopyranosyl-(1""→6"')-O-beta-D-4C1-glucopyranosyl ester, isolated from the leaves, was found nontoxic (LD50) and to have significant anti-inflammatory activity.[55] It also showed antinociceptive effects that are more potent than the reference drugs. The mechanism responsible for the antinociceptive action of the extract is partly related to the modulated release or action of proinflammatory mediators involved in the models of pain used. It also showed a slight antischistosomal activity.

A novel flavonol glycoside, 5,7,3',4'-tetrahydroxy-3-methoxy-7-O-α-L-rhamnopyranosyl(1→ 3)-O-ß- D-galactopyranoside (5), molecular formula $C_{28}H_{32}O_{16}$, isolated from ethyl acetate soluble fraction of the 90% ethanolic extract of the roots of *B. variegata* showed marked anti-inflammatory activity as tested by carrageenan induced hind paw oedema method.[104]

In the continuing search for novel anti-inflammatory agents, six Flavanoids namely, kaempferol (6), ombuin (7), kaempferol 7,4'-dimethylether 3-0-ß-D-glucopyranoside (8), kaempferol 3-0-ß-D-glucopyranoside (9), isorhamnetin 3-0-ß-D-glucopyranoside (10) and hesperidin (11), together with one triterpene caffeate, 3ß-trans-(3,4-dihydroxycinnamoyloxy) olean-12-en-28-oic acid (12) were isolated from the non-woody aerial parts of B. variegata. Compounds 6-12 were evaluated as inhibitors of some macrophage functions involved in the inflammatory process. These seven compounds significantly and dose dependently inhibited lipopolysaccharide



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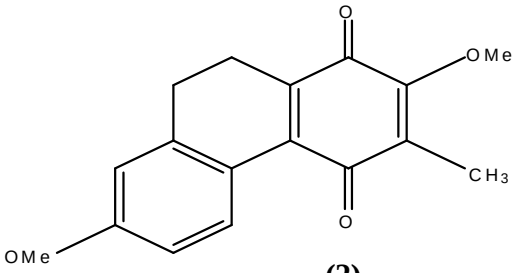

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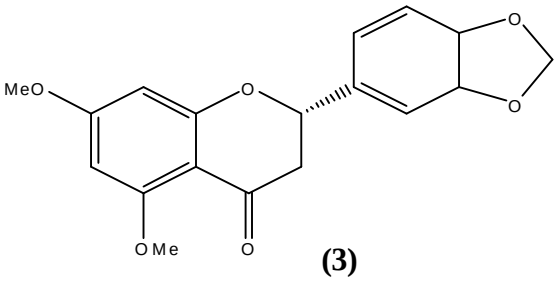

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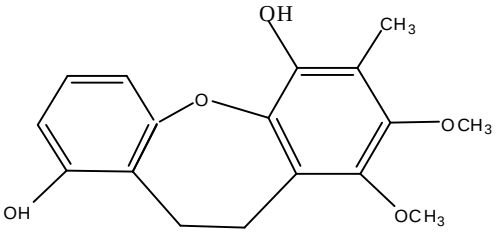

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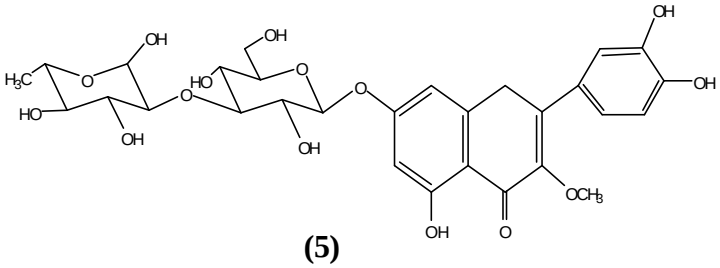

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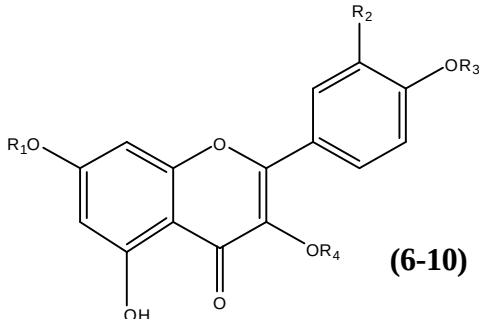
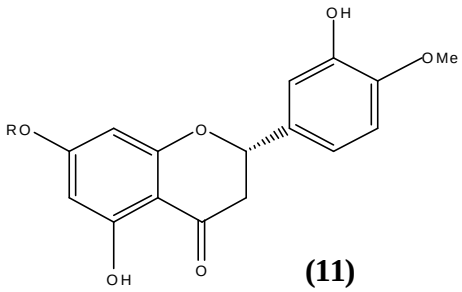


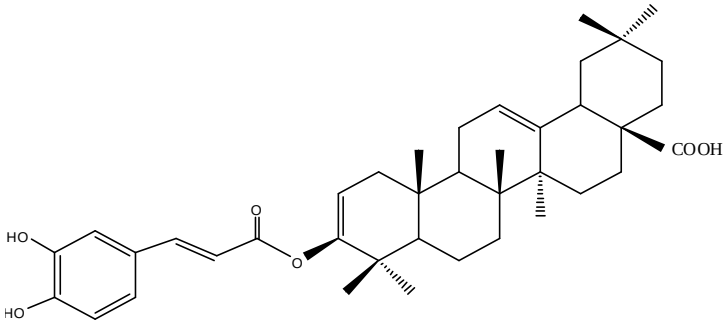
(6-10)

Compound No.	R_1	R_2	R_3	R_4
6	H	H	H	H
7	Me	OH	Me	H
8	Me	H	Me	Glc
9	H	H	H	Glc
10	H	OMe	H	Glc



(11)

R = Rha-Glc



(12)

(LPS) and interferon (IFN)-γ induced nitric oxide (NO), and cytokines (tumour necrosis factor (TNF)-α- and interleukin (IL)-12.[80]

3.Anticarcinogenic & Antimutagenic

Anticarcinogenic and antimutagenic potential of *B. variegata* extract was evaluated in Swiss albino mice using a skin carcinogenesis and melanoma tumour model, along with micronucleus and chromosomal aberration tests. In the skin papilloma model, significant prevention, with delayed appearance and reduction in the cumulative no. of papillomas was observed in the DMBA + Kachanar + croton oil treated group as compared to the DMBA + Croton Oil group. C57 Bl mice which received a 50 % methanolic extract of Kachanar extract at the doses of 500 and 1,000 mg/ kg body weight for 30 days showed increase in life span and tumour size was significantly reduced as compared to controls. In antimutagenicity studies, a single application of Kachanar extract at doses of 300, 600 and 900 mg/kg dry weight, 24 hours prior the i.p. administration of cyclophosphamide (at 50 mg/kg) significantly prevented micronucleus formation and chromosomal aberrations in bone marrow cells of mice, in a dose dependent manner.[2]

In another study, ethanol extract of *B. variegata* (EBV) was evaluated against Dalton's ascitic lymphoma (DAL) in Swiss albino mice. EBV treatment was found to enhance peritoneal cell counts, probably mediated through enhancement and activation through macrophage or through some cytokine production inside the peritoneal cavity produced by EBV treatment. EBV-treated groups were able to reverse the changes in the haematological parameters consequent to tumour inoculation. Flavanoids which have been shown to possess antimutagenic and anticarcinogenic activity and lectins reported to produce structural variation of the cell envelope may account for this effect.[76]

The chemopreventive and cytotoxic effect of EBV was evaluated in N-nitrosodiethylamine (DEN, 200mg/kg) induced experimental liver tumour in rats and human cancer cell lines. EBV was found to be cytotoxic against human epithelial larynx cancer (Hep2) and human breast cancer (HBL-100) cells.[77]

4.Antimicrobial

B. variegata collected from Nepal was found to have antimicrobial activity against *Bacillus subtilis, Pseudomonas aeruginosa, Salmonella typhi, Shigella dysenteriae, S. aureus* and *Vibrio cholerae*. The same plant, collected in India, showed remarkable activity against gram +ve and gram –ve bacteria, being more effective against gram +ve bacteria.[27] The largest zone of inhibition was found to be exhibited against *B. subtilis*. In disc diffusion assays several solvent extracts of *B. variegata* inhibited the growth of *Staphylococcus aureus.*[70]

5.Antihyperlipidemic

Lipids are one of the most susceptible targets of free radicals. This oxidative destruction is known as lipid peroxidation and may induce many pathological events. In the preliminary studies, it was found out that the aqueous and ethanolic extracts of *B. variegata* Linn. have shown promising antihyperlipidemic activity.[73] It may partly owe its antihyperlipidemic activity to its antioxidant activity. A study on antihyperlipidemic activity of butanolic fraction of total methanol extract of leaves against Triton WR-1339 induced hyperlipidemia in rats showed not only significant reduction in cholesterol, triglyceride, LDL,VDL level, but also an increase in HDL level.[43]

6.Hemagglutinator

BVL, mol. wt. 81,000; purified from seeds has shown hemagglutinating activity. N-acetyl-D-galactosamine is the most potent inhibitor of BVL. Lactose and galactose can also inhibit the agglutination of rabbit erythocytes by BVL.[38,69]The hemagglutinating activity of dimeric 64-kDa Melibiose-binding lectin was stable after 30 min. exposure to temperatures up to 70^0C and found to inhibit proliferation in hepatoma HepG2 cells, breast cancer MCF7 cells and HIV-1 reverse transcriptase activity.[49]

7.Hepatoprotective

B. variegata alcoholic stem bark extract (SBE) exhibits significant hepatoprotective activity in CCl_4 intoxicated Sprague-Dawley rats.[12] Hepatotoxins increase the levels of total lipids in liver. Total lipid content in serum and liver registered a significant hike, which was retrieved to near normalcy in SBE treated rats. This is the clear indication of the improvement of the functional integrity of the liver cells. CCl_4 impairs the capacity of the liver to synthesize albumin. So the protein content of serum decreases in such cases. Retrieval of protein concentration to normalcy further confirms *B. variegata* stem bark extract's hepatoprotective effect and its use as liver tonic.

8.Insulin Release Enhancer

The presence of insulin-like molecule was recently demonstrated in the leaves, where a 'chloroplast protein' was found that has a partial amino acid sequence identical to that of Bovine insulin. This protein may be responsible for the lowering of blood glucose concentration when it is injected in alloxan induced diabetic mice.[9] A major metabolite of the ethanolic extract of leaves; roseoside, demonstrates insulinotropic activity toward pancreatic ß-cells of the INS-1 cell line and may act in conjunction with the chloroplast protein to contribute to the overall antidiabetic properties.[28]

9.Nephroprotective

The nephroprotective activity of the ethanolic extract of *Bauhinia variegata* (Linn.) whole stem against cisplatin-induced nephropathy was investigated by an *in vivo* method in rats. Treatment with the ethanol extract of BV at the dose level of 400 mg/kg b.w. for 14 days (EEBV400 group) significantly lowered the serum level of creatinine and urea, decreased urine creatinine and albumin with a significant weight gain, and increased urine output when compared with the toxic group. The histological damages in the BV extract-treated group were minimal in contrast to the toxic rats.[65]

10.Proteinase Inhibitor

Inhibitors isolated from different species of Bauhinia seeds inhibit blood clotting enzymes, as well as other serine and cysteine proteinases. Two varieties *B. variegata* seeds, shown to possess Plant Kunitz type inhibitors – B. variegata trypsin inhibitors, viz. *Bauhinia variegata Candida* trypsin inhibitor (Bvc TI) and *B. variegata lilac* trypsin inhibitor (BvlTI) are proteins, Mr about 20,000, with four cysteine residues forming two disulphide bridges in one polypeptide chain.[63] The complete sequences have been determined by automated Edman degradation of the reduced and carboxymethylated proteins of the peptides resulting from *Staphylococcus aureus* protease and trypsin digestion.

11.Wound Healing Activity

Recently, through cloning and expression in *E. coli*, a recombinant isoform of the lectin of *B. variegata* (rBVL-1) was obtained, with DNA sequence and amino acids similar to other well-known Caesalpinoideae lectins.[56] A study to investigate the healing potential of topical administration of the lectin of *B. variegata* (nBVL) and its recombinant isoform (rBVL-1) on surgically induced skin wounds in a murine model was performed. The results indicated that lectin extracted from the seeds of *Bauhinia variegata* (nBVL) can stimulate the healing process of skin wounds in mice, possibly by acting on cells of the immune system, enhancing proinflammatory response, collagen synthesis by fibroblasts and angiogenesis by modulating the release of inflammatory cytokines and growth factors. The recombinant isoform of the lectin (rBVL-1) displayed pro-healing effects as well. The secretion of growth factors and consequent phenotypic change of fibroblasts into myofibroblasts may explain the observed acceleration in wound closure (faster wound contraction and increased collagen deposition by fibroblasts, even in the inflammatory phase), with effective healing accomplished by POD 7.

***B. variegata* – RECENT ACTIVITIES:**

Medicinally important *B. variegata* plant has shown capability to synthesize various shape and sized 'Gold Nanoparticles' (GNP) in very less time. Fast and eco-friendly green synthesis method developed using *B.variegata* leaf extract led to the synthesis of polyshaped GNP. Importantly, most of the GNP synthesis was over in 60 minutes. Attachment/adsorption of medicinally important molecules present in extract like saponins and carbohydrates will enhance the quality of GNP.[46]

B. blakeana, or the Hong Kong Orchid tree, regarded as a horticultural cultivar, is completely sterile and has shown to be the result of hybridization between the largely sympatric species, *B. purpurea* and *B. variegata*. The homology of internal transcribed spacer 1 sequences among *B. blakeana*, *B. purpurea* and *B. variegata* provide definitive molar evidence for de Wit's theory (de Wit 1956) that *B. blakeana* is an interspecific diploid hybrid of *B. purpurea* and *B. variegata*. The perfect match of atpB-rbcl intergenic spacer sequences in *B. purpurea* and *B. blakeana* indicated that the former is the female parent of the hybrid. However the current results could not disclose which variety of *B. variegata* is the paternal parent.[48,51]

A recent study on mixture design optimization of extraction and mobile phase media for fingerprint analysis of *B. variegata* L. has shown that the optimum solvent compositions to extract chemical substances of the three mobile phases for chromatographic analysis of *B. variegata* L. are acetone/ethanol (25:77v/v), dichloromethane/ethanol (70:30v/v) and 100% dichloromethane respectively.[24]

A rapid RP-HPTLC densitometry method for simultaneous determination of major Flavanoids viz. Apigenin, Luteolin, Quercitrin, Rutin, Quercitin has been developed.[11] The technique is precise, specific, accurate and reproducible. Statistical analysis proves that the method is repeatable and selective for analysis.

Formulation and preparations include Usirasava; Candanasava; Vidangarista; (Asava and Arista), Kanchanara guggulu (Guggulu), Kanchanara drava (Bhasma), Gandamala Kandana Rasa (Rasayoga) (Anonymous, 1978; 2000).

Two pharmaceutical or medicinal preparations containing *B. variegata* or active ingredients that have been extracted or chemically synthesized, viz. 'Herbal Composition for treatment of immunocompromised conditions' (PAT - US7344738) and 'Speckle removing plaster of white pharbitis hil' (PAT - CN101129792) are under the process of obtaining patent.

CONCLUSION

Though *B. variegata* L. appears to be a promising therapeutic agent, more elaborate work is required to characterize the exact active principles responsible for its dynamics, to elucidate its mechanisms of action using differenr cell lines and to obtain new and more potent synthetic derivatives. Additional investigations are needed to confirm the antidiabetic potential of this plant in humans.

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