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RESEARCH ARTICLE

ETHNOBOTANY, PHYTOCHEMISTRY AND PHARMACOLOGICAL EVALUATION OF *VERNONIA ANTHELMINTICA* (L.) WILLD.: AN OVERVIEW

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ABSTRACT

Medicinal plants are the nature's gift to human being to have disease free healthy life. It plays a vital role to preserve our health. *Vernonia anthelmintica* (L.) Willd. is a potential medicinal plant and the seeds were used for many human ailments. In the Ayurveda, an Indian system of medicine, the seeds were used to cure helminthes parasites, skin diseases, leucoderma and fevers, while in Unani system seeds were used to controlling asthma, renal disorders, itching of eyes and inflammations. Seeds decoction used in dysentery, gonorrhoea, gastric troubles and seed paste applied externally to scorpion sting and decoction given orally to control helminthes parasites.

INTRODUCTION

The *Vernonia* genus has about one thousand species and the members of the genus are widely used as food and medicine. A total of 109 *Vernonia* species were reported in the literature to have medicinal properties. The genus *Vernonia* is named after Willian Veron, an English Botanist who collected and identified this genus. *Vernonia anthelmintica* Willd. (Syn. *Centratherum anthelminticum* Kuntze or *Ascaradia indica* or *Conyza ascaradia* or *Serratula anthelmintica*) belongs to family Asteraceae, is an annual herb distributed throughout India. (Nandkarni, 1982; Mashelkar, 2008)

Taxonomic Classification

- Kingdom : Plantae
- Division : Angiosperms
- Class : Eudicots
- Subclass : Asterids
- Order : Asterales
- Family : Asteraceae

- Subfamily : Cichorioideae
- Tribe : Vernonieae
- Genus : *Vernonia*
- Species : *Vernonia anthelmintica*

Vernacular Names (Kirtikar and Basu, 2003)

- Sanskrit : Somaraja, Vakuchi, Agnibija, Aranyajiraka, Putiphali, Krishnaphala, Kananajiraka, Tiktajiraka
- Hindi : Bakshi, Kalijhiri, Somraj, Vapchi, Buckshi
- Gujarati : Kalijiri, Kadvojiri
- English: Purple Fleabane
- Marathi : Kalajira, Kalenjiri, Kalijiri, Ranachajire
- Bengali : Babchi, Bukshie, Hakuch, Kaliziri, Somraj
- Punjabi : Bukoki, kakshma, Kaliziri
- Malayalam : Kattujirakam, Puvankuruntala
- Telugu : Nelavavili, Garitikamma, Vishakantakamula
- Urdu : Janglijiri
- French: Herbe aux mouches
- Arabic: Atarilal, Itrilal, Kamunebari

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Figure 1. *Vernonia anthelmintica*



Figure 2. *Vernonia anthelmintica* - Seeds

Botanical Description

Vernonia anthelmintica is an annual, erect and leafy plant about 1.5 m in height. The stems are branched and pubescent. Leaves are 5-9 cm long, 2.5-3.2cm broad, lanceolate or elliptic-lanceolate, acute-apex, coarsely serrate, pubescent on both surfaces, petiolate with tapering base. Heads are 1.3-2cm diameter, subcorymbose with about 40 flowers and linear bract near the top of the peduncle. Outer involucre bracts are linear, hairy, shorter than those of the inner rows; intermediate bracts with herbaceous hairy tips, linear, acute, equaling or shorter than the innermost; innermost bracts usually the longest, linear, subacute, often tipped with purple. Pappus reddish, the exterior row very short, subpaleaceous, persistent; the inner hairs somewhat flattened, deciduous, much shorter than the glabrous corolla. Achenes are 4.5-6 mm long, oblong-cylindrical, 10 - ribbed, pubescent. (Mashelkar, 2008; Kirtikar and Basu, 2003)

Ethnobotany

Vernonia anthelmintica (L.) Willd. is a potential medicinal plant, known as jungle kalajeera (Hindi) by the local people. According to Ayurveda, seeds are hot, acrid, astringent, anthelmintic; cure ulcers, vata and kapha, mostly used in skin disease, leucoderma and fever. According to Unani system of medicine, the seeds are anthelmintic, purgative; used for asthma, kidney troubles, hiccup, inflammatory swellings, to remove blood from the liver, sores and itching of the eyes. The seeds are anthelmintic, stomachic, tonic, diuretic and

antiperiodic in action. The powdered seeds are applied externally in paralysis of the legs at Mundas of Chota Nagpur. The juice of the leaf is given to cure phlegmatic discharges from the nostrils. In Ceylon, the plant is used for fever convulsions. ((Nandkarni, 1982; Mashelkar, 2008; http://species.wikimedia.org/wiki/ernonia_anthelmintica)

Phytochemistry

Nine new highly oxygenated stigmastane-type steroids, vernoanthelein A-I and two new stigmastane-type steroidal glycosides, vernoantheleside A and B were isolated from the aerial parts of *Vernonia anthelmintica* Willd. The structures of compounds 1-11 were determined on the basis of IR, MS, 1D-NMR, and 2D-NMR, and their absolute configurations were deduced using single-crystal X-ray diffraction and the CD exciton chirality method. Compounds 1, 5, 7, 9 and 10 were tested for their effects on estrogen biosynthesis in human ovarian granulosa-like cells (KGN cells). (Lei *et al.*, 2012) A new steroid, vernoanthelesterone A (1), and five known steroids were isolated from the aerial parts of *Vernonia anthelmintica* Willd. Compound 1 possesses a $\Delta 8(14)$ -15-one moiety, few steroids with this moiety have been reported. Compounds 1-6 were tested for their antibacterial activities and their effects on estrogen biosynthesis in human ovarian granulosa-like cells (KGN cells). Compound 2 showed the ability to promote estrogen biosynthesis with EC₅₀ of 56.95 μ g/ mL and also exhibited the antibacterial activities against *Bacillus cereus*, *Staphylococcus aureus*, *Bacillus subtilis* and *Escherichia coli* with MICs ranging from 3.15 to 15.5 μ g/ mL. The structures of 1-6 were determined on the basis of IR, MS, 1D and 2D NMR. (Lei *et al.*, 2012) Chemical synthesis of two trisaccharides related to the saponin isolated from *Centartherum anthelminticum* is reported. Stereoselective, high yielding glycosylation strategies were developed using H₂SO₄ immobilized on silica for activation of trichloroacetimidate donors, or in conjunction with N-iodosuccinimide for activation of a thioglycoside.

A late stage TEMPO-mediated oxidation was performed for the formation of the required uronic acid moiety. (Mandai and Mukhopadhyay, 2007) Two novel elemanolide dimers, Vernodalidimers A (1) and B (2) were isolated possessing a rare tri-cyclic ortho ester moiety, from the seeds of *Vernonia anthelmintica*. Their structures were elucidated by 1D and 2D NMR data and CD spectra. Vernodalidimers A (1) and B (2) exhibited potent cell growth inhibitory activity against HL-60 cells (IC₅₀ 0.072 and 0.47 OI. M. respectively). (Yongqiang *et al.*, 2010) Several flavonoids including 2',3,4,4'-tetrahydroxychalcone, 5,6,7,4'-tetrahydroxyflavone and butin, were collected from the seeds of *Vernonia anthelmintica* Willd by high-speed counter-current chromatography using a two-step operation. Two different types of solvent systems were used: chloroform -dichloromethane-methanol water (2:2:3:2, v/v) and 1, 2 di-chloro-ethane-methanol-acetonitrile-water (4:1.1:0.25:2, v/v). From 1 kg of seeds of *Vernonia anthelmintica* Willd the method yielded about 45 mg of 2',3,4,4'-tetrahydroxychalcone, 40 mg of 5,6,7,4'-tetrahydroxyflavone, and 55 mg of butin. Each isolated component showed 95 -97% purity as determined by high-performance liquid chromatography analysis. These purified compounds were characterized by MS and NMR (Guilian *et al.*, 2004). A new glycosylated triterpene has been isolated from the seeds of *Centratherrum anthelminticum*, a medicinally

important plant. The structural analysis of its acetylated derivative was performed by ^1H , ^{13}C NMR, ^1H - ^1H COSY, HMQC, HMBC and DEPT spectroscopy. The saponin was shown to contain hederagenin and six sugar residues forming two glycosyl chains. The complete structure of the saponin was established as 3-O-[β -D-glucopyranosyl-(13)- α -L-rhamnopyranosyl-(12)- α -L-arabinopyranosyl]-28-O-[β -D-glucuronopyranosyl-(1-4)- α -L-rhamnopyranosyl-(1-3)- β -D-glucopyranosyl]-hederagenin. (Mehta *et al.*, 2004) In the present investigation the *In vivo* radiotracer experiments using [^{14}C] acetate as the precursor were conducted to investigate the biosynthesis of vernolic acid (12, 13-epoxy-cis-9-octadecenoic acid) in the seeds of *Vernonia galamensis*. The acetate precursor radioactively labeled vernolate in phosphatidylcholine (PC) diacylglycerol, and triacylglycerol. Time-course kinetics of the incorporation of the radioactive tracer indicated that vernolate is synthesized while the acyl moiety is esterified to PC. Pulsechase experiments provided additional supporting evidence that vernolate is synthesized while esterified to PC. These results are consistent with the hypothesis that linoleoyl PC is the precursor of vernoleoyl-PC. Subsequently, vernolate is quickly moved from the PC pool to the triacylglycerol pool, where it accumulates (Linsen *et al.*, 1998).

The seed oil of *Vernonia anthelmintica* on reaction with diols (mono-, di-, tri-, or tetraethylene glycols) in the presence of boron trifluoride etherate, followed by saponification and esterification (methanol/ H^+), gives the oligoethylene glycol ethers: methyl 12(13)-hydroxy-13(12) [2-hydroxy-ethyl-1-oxy]-octadec-9-enoate; methyl 12(13)-hydroxy-13(12)-[5-hydroxy-3-oxapentyl-1-oxy]-octadec-9-enoate; methyl 12(13)-hydroxy-13(12)-[8-hydroxy-3,6-dioxaoctyl-1-oxy]-octadec-9-enoate; and methyl 12(13)-hydroxy-13(12)-[11-hydroxy-3,6,9-trioxaundecyl-1-oxy]-octadec-9-enoate. Methyl 12, 13-dihydroxyoctadec-9-enoate is a co-product in all reactions. (Singh, 1997) *Vernonia anthelmintica* (L.) Willd. seed was found to contain enzymes that were active only after the seed was ground. By deactivation of the enzymes, an oil rich in trivernolin (the triglyceride of epoxyoleic acid) was produced, and pure trivernolin was isolated. Acceleration of the enzyme activity altered the composition of the oil as evidenced by changes in free fatty acid content, iodine values, and oxirane oxygen (epoxy) content. Investigation showed that these changes were due, at least partially, to the conversion of epoxyoleic acid to (+)-threo-12,13 dihydroxyoleic acid, which was isolated in pure form. Pure (-)-threo-12, 13-dihydroxyoleic acid was prepared by acetolysis of *V. anthelmintica* seed oil. Neither of these isomers had been obtained previously from mature *anthelmintica* seed. (Scott *et al.*, 1963)

Pharmacological Evaluation

Anti-inflammatory Activity

Anti-inflammatory Activity with Petroleum ether and alcoholic extracts of *V. anthelmintica* seeds (100 mg and 200 mg/kg p.o.) showed anti-inflammatory activity in acute and subacute models of inflammation. Both petroleum ether and alcoholic extracts showed significant reduction in paw oedema in carrageenan-induced model. In subchronic inflammatory phase both extracts provoked a significant reduction of transudation phase and too little extent proliferative phase

when tested in cotton pellet-induced granuloma model. Both the extracts also reduced alkaline phosphatase activity in serum. The histopathology of granuloma tissue showed significant inhibition of lymphocytes, neutrophils, exudates, necrosis and giant cell when compared with control without ulcerogenic effect. The results suggested that petroleum ether and alcoholic extracts may exert anti-inflammatory activity through prostaglandin inhibition, reduced myeloperoxidase and antitransudation. (Purnima *et al.*, 2010)

Antiarthritic Activity

The ethanolic extract of seeds of *V. anthelmintica* EVA (250 and 750 mg/kg) and EVA (500 mg/kg) showed significant ($p < 0.05$ and $p < 0.01$ respectively) inhibition of paw edema from 1 to 6 has compared to carrageenan control, but the effect was not dose-dependent. EVA 250 mg/kg showed significant ($p < 0.01$) prevention of the paw edema on 28th day, whereas the treatment with EVA 500 mg/kg showed significant ($p < 0.01$) prevention in the paw edema during 21st and 28th day as compared to the arthritis control. In xylene induced edema treatment with EVA (250, 500 and 750 mg/kg) showed significant ($p < 0.01$) decrease in ear thickness as compared to control during the 30 to 60 min. (Otari *et al.*, 2010)

Antidiabetic and Antihyperlipidemic Activity

Ethanolic extract of *Vernonia anthelmintica* seeds showed significant hypoglycemic effect in STZ (Streptozotocin) induced diabetic rats. At a dose of 100 mg/kg body weight of extract showed a significant antihyperglycemic activity in the diabetic treated rats with a maximum fall of 82.3% in the blood glucose level after the 6th hour of treatment when compared with other fractions. Administration of ethanolic extract (100 mg/kg body weight) for 45 days showed significant reduction in plasma glucose, HbA1C, cholesterol, triglycerides, LDL, VLDL, free fatty acids, phospholipids and HMG-CoA reductase in STZ diabetic rats. Also significant decrease in plasma insulin, protein, HDL and hepatic glycoen observed in STZ diabetic rats and that was normalized after 45 days of treatment with the active fraction of *V. anthelmintica* seeds. Thus the seeds of *V. anthelmintica* showed significant antidiabetic and antihyperlipidemic property without toxic effects. (Shaik *et al.*, 2010)

Anthelmintic Activity

The seeds of *V. anthelmintica* showed anthelmintic activity. *In vitro* studies revealed higher anthelmintic effects ($p \geq 0.05$) of crude methanol extract (CME) as compared with crude aqueous extract (CAE) of *V. anthelmintica* seeds on live *Haemonchus contortus* as evident from their mortality. *In vivo* studies, seeds of *V. anthelmintica* were administered as crude powder (CP), CAE, and CME to sheep naturally infected with mixed species of gastrointestinal nematodes. *In vivo* maximum reduction (73.9%) in fecal egg counts per gram (EPG) was recorded in sheep treated with *V. anthelmintica*. CAE at 3g kg^{-1} body weight on day 5 post-treatment (PT) followed by CP at 3g kg^{-1} (55.6%) on day 3 PT. CME did not exhibit anthelmintic activity *in vivo*. (Zafar *et al.*, 2006)

Anticancer Activity

The chloroform fraction of *Centratherum anthelminticum* seeds (CACF) showed cytotoxic effect and TNF- α inhibition activity with NF- κ B activation response. The cytotoxicity of CACF was tested using the MTT (methyl thiazolyltetrazolium) assay; CACF effective inhibitory concentrations (IC₅₀) for A-549, PC-3, MCF-7, and WRL-68 cells were 31.42 ± 1.54 , 22.61 ± 1.17 , $8.1:1 \pm 0.9$, and 54.93 ± 8.3 μ g/mL, respectively. CACF effectively and dose-dependently inhibited TNF- α release, in-vitro and in-vivo. CACF inhibited TNF- α secretion in stimulated RAW264.7 macrophage supernatants with an IC₅₀ of 0.012 μ g/mL, without affecting their viability; the highest dose tested reduced serum TNF- α by 61 %. Acute toxicity testing in rats revealed that CACF was nontoxic at all doses tested. Matching the cytotoxic activity towards a mechanistic approach, CACF dose-dependently exhibited in-vitro inhibitory effects against the activation of NF- κ B translocation in MCF-7 cells. Thus these studies revealed the potential of CACF in the treatment of breast cancer and in oxidative stress conditions with associated inflammatory responses. (Aditya *et al.*, 2012)

Antimicrobial Activity

Methanolic extracts of *V. anthelmintica* showed antibacterial and antifungal activities. Antibacterial study performed against six bacteria viz., *Escherichia coli*, *Citrobacter*, *Shigella flexenari*, *Yersinia aldovae*, *Staphylococcus aureus* and *Pseudomonas aeruginosa* while the antifungal activity of these extracts performed against six fungi, viz., *Saccharomyces cerevisiae*, *Aspergillus parasiticus*, *Trichophyton rubrum*, *Macrophomina*, *Fusarium solani* and *Candida albicans*. Methanolic extract of the seeds showed significant antibacterial activity against all tested bacterials except *Yersinia aldovae* while showed antifungal activity against only *Trichophyton rubrum* (Jahan *et al.*, 2010).

Antioxidant Activity

Methanol extract (75%) of *V. anthelmintica* was found to be potent inhibitors of lipid peroxide formation and scavengers of hydroxyl and superoxide radicals' in-vitro (Antioxidant Activity of Indian Herbal Drugs in Rats with Aloxan -induced Diabetes, 2003) The chloroform fraction of *V. anthelmintica* demonstrated significant antioxidant activity with DPPH (1, 1-Diphenyl-2-picrylhydrazyl), ORAC (oxygen radical absorbance capacity), and FRAP (Ferric Reducing! Antioxidant Power) assays (Aditya *et al.*, 2012).

Estrogen Biosynthesis Activity

Nine highly oxygenated stigmastane-type steroids, vernoanthelein A-I (1-9), and two stigmastane-type steroidal glycosides, vernoantheleine A and B (10 and 11) have been isolated from the aerial parts of *V. anthelmintica*. Compounds 1, 5, 7, 9 and 10 were tested for their effects on estrogen biosynthesis in human ovarian granulosa-like cells (KGN cells). (Lei *et al.*, 2012)

Immunomodulatory Activity

The orally administered seed of *V. anthelmintica* exhibited suppression of Rh-incompatibility of female during pregnancy. Phytochemical study showed that no harmful steroids are present in seeds. These studies reveals that seeds suppressed

Rh-incompatibility in patients either by suppressing the sensitivity of B and T cells to the antigens or by blocking both T and B cells to undergo blast formations and did not allow them to differentiate into antibodies forming cells. (Shaheen *et al.*, 2006)

Melanogenesis Activity

V. anthelmintica flavone (VAF) stimulated melanogenesis directly on human epidermal melanocytes. Three different concentrations of VAF were incubated with human melanocytes for 72h and then melanin content and tyrosinase activity were respectively measured. The proliferation of melanocytes was most inhibited by VAF at high dose ≥ 100 μ g/ml. In the range of experimental dose, VAF increased the activity of the tyrosinase and melanin content significantly in a dose - dependent manner. The increasing protein expression of TRP 1 was in a dose dependant manner. No effect was observed on protein expression of TYR and TRP2. The studies revealed that VAF may stimulate melanogenesis directly on human epidermal melanocytes, which may be induced by increasing expression of TPR 1 and post-translational modification of TYR. (Hui-jun *et al.*, 2008)

Role as Diuretic agent

This review articles identify the plant extracts which promotes diuresis and also to identify the research needed area. A number of genus and species reporting diuretic effects. *Xylopi aethiopica* (dunal) and *Alepidea amatymbica* eckl. & zeyh, *Steganotaenia araliacea* hochst, *Carissa edulis* (forssk) vahl, *Oxystelma esculentum*, *Tylophora indica*, *Centratherum anthelminticum* (L.) kuntze, *Opuntia ficus indica* (L.) mill., *Spergularia purpurea* pers., *Spilanthes acmella* murr., *Raphanus sativus* var nigra L., *Lagenaria siceraria* (mol.) standi, *Equisetum bogotense* tea (platero herb). (Kumar *et al.*, 2010)

Analgesic and antipyretic activity

Analgesic and antipyretic activities of petroleum ether and alcohol extracts of *Centratherum anthelminticum* (L) Kuntze (family: Asteraceae) seeds (100 and 200 mg/kg, p.o.) evaluated in brewer's yeast-induced fever model in rats, acetic acid-induced writhing and Eddy's hot plate methods in mice. Both petroleum ether and alcohol extracts showed significant decrease in number of writhes in acetic acid-induced writhing and increase in paw licking time to heat stimuli in the hot plate method. The maximum analgesic activity was observed at 90 min after dosing when compared to control. Both the extracts showed significant inhibition of elevated body temperature when compared to corresponding control. These results suggested that the petroleum ether and alcohol extracts possessed analgesic and antipyretic activities (Purnima *et al.*, 2009)

Conclusion

On the basis of Pharmacological and Phytochemical Studies reported, supporting the use of plant for diabetes, Cancer and inflammatory conditions. *V. anthelmintica* is used in traditional system of medicine to treat various physiological conditions. It is an important source of various types of compounds as wet: as pharmacological activities. Further

research in view of fulfilling the need of quality control aspects, standardization of the various constituents and extracts are needed. Also there is a need to explore its maximum potential in the field of medicinal and pharmaceutical sciences for novel and fruitful applications.

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