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REVIEW ARTICLE **OPEN ACCESS**

A REVIEW ON ETHNO MEDICAL AND BIOLOGICAL ASPECTS OF *RUELLIA TUBEROSE*

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ABSTRACT

Ruellia tuberosa, a perennial herb belonging to the family Acanthaceae, is a member of the third-largest tropical family of dicotyledonous plants, comprising approximately 2500 species, most of which possess medicinal properties. Traditionally, *R. tuberosa* has been utilized for its antiseptic, depurative, diaphoretic, diuretic, emetic, and purgative properties, and has been employed to treat various ailments, including bronchitis, constipation, bladder stones, cystitis, fever, leprosy, gonorrhea, and other venereal diseases. This study aims to provide an overview of the phytochemical, ethnomedicinal, and biological properties of *R. tuberosa*, highlighting its importance and potential applications.

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INTRODUCTION

Ruellia tuberosa, an erect, sub-erect, or diffuse perennial herb growing up to 60-70 cm tall, belongs to the family Acanthaceae. This plant is known by various names, including Minnie Root, Fever Root, Snapdragon Root, Sheep Potato, and is commonly referred to as the "Cracker plant". Typically found in shady, moist areas such as side drains, *R. tuberosa* have been traditionally used by local populations as a diuretic, antipyretic, and anti-hypersensitive agent. The plant has been extensively employed for its antiseptic, depurative, diaphoretic, diuretic, emetic, and purgative properties, and has been used to treat various ailments, including bronchitis, constipation, bladder stones, cystitis, fever, leprosy, gonorrhea, and other venereal diseases (Lans, 2006; Phytochemical and Ethnobotanical Databases, 2007). Phytochemical analyses have revealed a diverse range of secondary metabolites, including long-chain alkane derivatives (Misra *et al.,* 1997), flavonoids (Wagner *et al.,* 1971; Nair & Subramaniyan, 1974), and sterols and terpenoids (Singh *et al.,* 2002; Phytochemical and Ethnobotanical Databases, 2007).

Phytochemical studies: Dorcas Olufunke Moronkola *et al.,* 2015, reported the composition of *Ruellia tuberosa* L. (Acanthaceae) leaf, stem, root, fruit, and flower volatile oils from Nigeria. The five volatile oils were obtained by hydro-distillation and were procured in 0.09 to 0.36% yields. Each was separately examined using GC-MS analysis. The result revealed that the leaf oil contain 24 compounds, which make-up 86.95% of it; stem oil has 15 compounds (accounting for 93.96%); root oil with 42 compounds being 91.49%; fruit oil contain 60 compounds which amount to 89.68% and flower oil has 6 compounds representing 95.06% of the oil. Dominant compounds in each essential oil are (%): leaf (E-phytol 21.06, tributylacetyl citrate 19.44, heptacosane 7.55); stem (m-xylene 33.83, heptacosane 16.57, p-xylene 9.67); root (heptane 22.25, heptacosane 12.89, borneol 12.48); fruit (hexacosane 15.43, sextone 13.12, heneicosane 11.14) and flower (tributylacetyl citrate 67.78, 2-methyl-2-pentanol 10.15, 1-methyl-1- cyclopentanol 6.90). Important classes of compounds in Nigerian *R. tuberosa* volatiles are monoterpenes, monoterpenoids, sesquiterpenes, sesquiterpenoids, hydrocarbons, aromatics, esters, alcohols, sulphur compounds, ketones and aldehydes.109 compounds were identified in the five essential oils of *R. tuberosa*. These compounds have high therapeutic effects and are characteristic of *R. tuberosa*. The oils are good sources of sextone (methylcyclohexane), β-linalool and alcohols. Daya *et al.,* 2012 reported that HPTLC fingerprinting was carried out for various extract of root, stem, and leaf of *R. tuberosa*. From the HPTLC fingerprint the florescent band (under 366 nm) at Rf: 0.56 (mobile phase chloroform: toluene : ethyl acetate (6 : 3 : 1, v/v)) was found in leaf, root, and stem of *R. tuberosa*. So, the florescent band (under 366 nm) at Rf: 0.56 was isolated as marker compound RT-F2 from root of *R. tuberosa*. The marker compound RT-F2 was quantified by using HPTLC technique. The percentage (W/W) amount of RT-F2 was found to 40.0% and

44.6% in petroleum ether and ethyl acetate extract of *R. tuberosa* roots, respectively.

Pharmacognostical studies : Various trichome morphotypes were the unique observation seen in the plant *R. tuberosa* by anatomical as well as powder microscopic studies. Four prominent peaks were detected by UV-Visible spectroscopy and the HPTLC fingerprinting results showed several peaks with different Rf values. Toluene: Ethyl acetate: Formic acid (5: 1.5: 0.1) was the suitable solvent system which resolved various bands on the chromatogram and it indicates various phytochemicals present in the plant (Neethu Kannan *et al*., 2021).

Biological studies

Anti-inflammatory activity: Ashraful Alam *et al.,* 2009 demonstrated that significant anti-inflammatory properties of the ethanol extract of *R. tuberosa*, which was comparable to those of the positive controls, and indicated that this plant could be a potential source for the discovery and development of newer analgesic and anti-inflammatory "leads" for drug development. Daya *et al.,* 2012 was aimed to identification, isolation, and quantification of marker in *R. tuberosa* (Acanthaceae). HPTLC fingerprinting was carried out for various extract of root, stem, and leaf of *R. tuberosa*. From the HPTLC fingerprint the florescent band (under 366 nm) at Rf : 0.56 (mobile phase chloroform : toluene : ethyl acetate (6 : 3 : 1, v/v)) was found in leaf, root, and stem of *R. tuberosa*. So, the florescent band (under 366 nm) at Rf: 0.56 was isolated as marker compound RT-F2 from root of *R. tuberosa*. The marker compound RT-F2 was quantified by using HPTLC technique. The percentage (W/W) amount of RT-F2 was found to 40.0% and 44.6% in petroleum ether and ethyl acetate extract of *R. tuberosa* roots, respectively. Further study is suggested to characterization and biological nature of marker compound.

Antioxidant and anti-proliferative activities: Bo Eng Cheong *et al*., 2013 evaluated the total phenolic constituents, antioxidant and anti-proliferative activities of *Ruellia tuberosa*. The total phenolic and flavonoid contents of the plant extracts were determined by using Folin-Ciocalteau and aluminium chloride colorimetric assays, respectively. The antioxidant activity of the plant extracts was evaluated using DPPH free radical scavenging assay while the anti-proliferative activity was evaluated using MTT assay against the human breast cancer (MCF-7) and cervical cancer (HeLa) cell lines. Significant correlation was found between the total phenolic/flavonoid contents with the total antioxidant activity while weak correlation was found between the total phenolic/flavonoid contents with the inhibition of MCF-7 cell proliferation. The findings indicated that *Ruellia tuberosa* could be a potential source for natural antioxidant as well as chemo-preventive agent against breast cancer in future. Manikandan and Victor Arokia Doss, 2010 evaluated the effect of 50% hydroethanolic leaf extracts of *Ruellia tuberosa* L. on non-enzymic antioxidants, liver glycogen, lipid peroxidation, urea, creatinine and LDH levels in the liver, kidney and serum of alloxan induced diabetic wistar rats. Extracts were orally administered for 30 days at a dosage of 250 and 500 mg/kg bodyweight for alloxan induced diabetic rats. A significant ($\rho < 0.05$) decrease was found in urea, LDH and lipid peroxidation (at 500mg/kg bodyweight) levels in the plant extract treated groups. The level of vitamin A and liver glycogen was significantly ($\rho < 0.05$) increased in the treatment and drug treated groups. The results suggest that the plant extracts treated at 500mg/kg body weight treated groups was found to be effective than the 250mg/kg body weight administration. 50% hydroethanolic leaf extracts of *Ruellia tuberosa* L. is not only useful in controlling the lipid peroxide level but are also helpful in further strengthening the antioxidant potential. Mohan *et al.,* 2014 stated that the extracts from the tuber of *R. tuberosa* were found to possess strong antioxidant activity and scavenging effects on free radicals. These in vitro assays indicate that this plant extract is a significant some of natural antioxidant, which might be helpful in preventing the progress of various oxidative stresses.

Antihypercholesteric activity: Krishna Chaitanya *et al*., 2012 evaluated the efficacy of *Ruellia tuberosa* ethanolic extract (RTEE2012) in reducing the cholesterol levels and as an antioxidant in hypercholesterolemic rats. Hypercholesterolemia was induced in normal rats by including high fat diet (cholesterol 25mg/kg in oil). Powdered form of RTEE2012 was administered as feed supplement at 250, 500 and 1000 mg/kg dose levels to the hypercholesterolemic rats. Plasma lipid profile, hepatic superoxide dismutase (SOD) activity, catalase activity and extent of lipid peroxidation in the form of malondialdehyde were estimated using standard methods. Feed supplementation with 250, 500 and 1000 mg/kg of RTEE2012 resulted in a significant decline in plasma lipid profiles. The feed supplementation increased the concentration of catalase, SOD and HDL-c significantly in the experimental groups (250, 500 and 1000 mg/kg). On the other hand, the concentration of malondialdehyde, cholesterol, triglycerides, LDL-c and VLDL in these groups (250, 500 and 1000 mg/kg) were decreased significantly.

Antianxiety activity: Govind shing *et al.,* 2020 revealed that the ethyl acetate and methanolic extract of *Ruellia tuberose* showed effective antianxiety activity. Although methanolic extract at 200 mg/kg improved significantly antianxiety like behavior by using Elevated plus maze & Light and dark models in rats.

Cytotoxic activity: Mamdouh *et al.,* 2015 evaluate various biological effects of the phytochemical constituents as well as extracts of *Ruellia patula* Jacq. and *Ruellia tuberosa* Linn. The study revealed that Rp-MeOH, Rp-Hexane, Rt-MeOH, Rt-Hexane and Rt-13 exhibited marked cytotoxic activity while, the compounds Rp-15, Rp-7, Rt-4 and Rt-12 exhibited moderate cytotoxic activity when compared with the positive control, doxorubicin. The compound Rt-13 showed a noticeable free radical scavenger activity, while RpEtOAc, Rp-7, Rp-15 and Rt-4 exhibited a moderate activity when compared to the reference compound, trolox. The compounds Rt-8, Rt-9 and Rp-14 showed approximately half the radical scavenger activity of the reference compound. The rest of the tested compounds did not exhibit any free radical scavenger activity. Rt-Hexane showed a weak antileishmanial activity while the remaining tested extracts and compounds showed no activity. All the tested extracts and compounds were found to exhibit no antibacterial activity.

Anti-Parkinsonian activity: Catalepsy was induced in rats using Rotenone and HART was administered orally to the treatment group. The behavioral changes were assessed using the hole board test for catalepsy, motor coordination test by rotarod, and locomotor activity by actophotometer. The findings of this study suggest that the hydroalcoholic extract of Ruellia tuberosa leaves possesses antiparkinsonian activity and could be a potential therapeutic agent for the treatment of Parkinson's disease. Further studies are needed to explore the mechanism of action and safety of this extract **(**Gayathri et al*.,* 2024)

Anti inflammatory and antioxidant activity: The ethyl acetate fraction (RTE) exhibited the most effective antioxidant activity against DPPH and ABTS radicals and also contained a high quantity of TFs and TPs. In the anti-inflammatory activity assay, the methanol and ethyl acetate fractions demonstrated the highest activity in the downregulation of the proinflammatory cytokine, IL6 secretion, and NO production at a concentration of 30 μg/mL in LPS-induced RAW 264.7 cells. Eight compounds were isolated and characterized from the RTE, of which three compounds 6–8 exhibited a strong inhibitory effect on NO generation. Physalin D (8) was the most effective compound when it inhibited IL-6 secretion by a factor of 14 at a concentration of 20 μg/mL. Compounds **6** and **7** decreased the level of protein IL6 from 3 to 8 times compared with the LPS control group in a dose-dependent manner. Based on these data, physalin D (8), physalin E (**7**), and hispidulin (**6**) are potentially bioactive compounds for the treatment of inflammation symptoms in type 2 diabetes mellitus from herbal medicines based on *Ruellia tuberosa* (Trinh Nhat Thi Pham *et al*., 2022).

Anticancer activity: Several bioactive constituents within Ruellia tuberosa L. methanolic extract (RTME) have significant anti-cancer potential against TNBC cells in vitro. Notably, the extract induces cellular apoptosis by targeting mitochondria-dependent intrinsic pathways via induction of intracellular reactive species (ROS) and promotion of G0/G1 cell cycle arrest. These findings suggest that RTME may serve as promising adjunct to conventional anti-cancer therapies, potentially enhancing treatment outcomes for triple-negative breast cancer. This serves a significant and may represent as a new beacon of light in the field of cancer therapeutics in the near future (Subhabrata Guha *et al.,* 2024). Anticancer activity of only *R. tuberosa* is reported so far. Arun *et al.,* 2008 showed that methanol extract of aerial part of herb *R. tuberosa* possessed cytotoxicity. Its minimum inhibitory concentration (IC50) for methanol extract was found to be 3.5 and 1.9 µg/mL in H460 and MDAMB231 cancer cells, respectively. They have also isolated Tylocrebrine from *R. tuberosa* through bioassay directed elucidated its column chromatography and anticancer and anti-inflammatory potential.

Gastroprotective and analgestic activity: Aqueous extract of *R. tuberosa* roots showed a dose dependent and robust gastroprotective activity in an alcohol induced gastric lesion model of rats. The extract also had mild erythropoietic and moderate analgesic activities and was well tolerated even with subchronic treatment (Suji and Velavan, 2021)

Antimicrobial activity: Senthilkumar *et al.,* 2013 reported that the methanol leaf extracts of *Ruellia tuberosa* showed significant antibacterial activity against *Escherichia coli, Pseudomonas aeruginosa, Klebsiella pneumonia, Bacillus subtilis, Proteus mirablis* and antifungal activity against *Aspergillussp, Mucorsp, Penicilliumsp and Fusarium sp.* The antibacterial potential of *Ruellia tuberose* methanol extract was tested by using Agar well diffusion method. The (100mg/ml) leaf extract showed maximum inhibition against *Proteus mirablis* (7mm). Further the extract showed maximum zone of inhibition against the fungus of Aspergillus sp (8mm). Phytochemical tests were performed and showed that the antibacterial activity of plant *Ruellia tuberosa* leaves was due to the presence of phytochemical compounds like alkaloids, tripenoid, tannins, glycosides, saponins. GC-MS analysis revealed the presence of 27 compounds. Ramadhan *et al.*, 2020 investigated that *R. tuberosa* L. roots extract was determined to act as a reducing agent and to form capping layers around the nanoparticles. The particle sizes of Fe-NPs were dependent on pH condition, pH 9 contributed to the most homogenous surfaces and the smallest size. FTIR spectra result demonstrated that carboxylate and hydroxyl groups of the extracts interacted with the iron nanoparticles and lead surface stabilization. The inhibition zones shown in the antibacterial screening test indicating that the Fe-NPs prepared in the current work has the effective antibacterial activity against pathogenic *S. aureus* and *E. coli*. In contrast, pH 3 resulted in the highest inhibition zones of Fe-NPs. The Fe-NPs that prepared biologically could be of convenient use in their proficient antibacterial activities.

Ethnomedicinal uses: In Siddha system of medicine, leaves are given with liquid copal as remedy for gonorrhea and ear diseases (Suseela and Prema, 2007), used in stomach cancer (Reddy *et al*., 1991). Dried and ground roots in dose of two ounces cause abortion and also used in sore eyes (Kirtikar and Basu, 1935). The herb also exhibits emetic activity and employed substitute of ipecac, also used in bladder stones and decoction of leaves used in treatment of Bronchitis (Anonymous, 1972). In folk medicine, it has been used as diuretic, antipyretic, antidiabetic, antidotal, thirst-quenching agent and analgesic and anti-hypertensive activity (Chiuand and Chang, 1995; Chen *et al.,* 2006). *Ruellia tuberosa* is used as cooling in urinary problem, uterine fibroids (Lans, 2001; Lans, 2006). It has recently been incorporated as a component in a herbal drink in Taiwan (Balik *et al.,* 2000). In Vietnam, the aerial parts of *Ruellia tuberosa* L. are used to treat stress oxidation and inflammatory symptoms in diabetes mellitus (Trinh Nhat Thi Pham *et al*., 2022).

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