AN UPDATED REVIEW ON WRIGHTIA TINCTORIA(Roxb). R Br

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ABSTRACT

Since ancient times human utilized the nature to cure various ailments. The knowledge of medicinal plants resulted in the development of various indigenous systems of medicine worldwide. Serendipitous discovery as well as scientific approach on the reason for medicinal properties of plants gave the knowledge of chemical constituents such as secondary metabolites in plants. *Wrightia tinctoria* which is commonly known as 'Danthapala' is a known potential medicinal plant, the leaves of which is traditionally used in the treatment of psoriasis and non-specific dermatitis in Siddha and Ayurvedic systems of medicine and distributed in tropical region belongs to the family Apocynaceae.. This plant is beneficial for the treatment of dandruff, various scalp and skin disorders. Phytochemical and pharmacological investigation on the various parts of the plant showed anti-ulcer, anti-inflammatory, analgesic, anthelmintic, anti-cancer, anti-dandruff, wound healing and anti-anxiety activity. The current review focus on providing an update on the recent pharmacological and phytochemical investigations on the plant by researchers around the globe with special emphasis on Antisporiatic, Antifungal, Antibacterial, Antiviral, Cytotoxic, Anti-inflammatory, Anti-diabetic, Analgesic, Hepatoprotective, Anthelmintic, and Wound healing activities.

Keywords: [Wrightia tinctoria, pharmacological, phytochemical, Antisporiatic, Wrightial, Indurubin}

1. INTRODUCTION

Wrightia tinctoria (Roxb) R.Br is comparatively a small deciduous tree, bark is scaly and smooth, young parts are glabrous or puberulous. Leaves variable, 7.5-15 cm in length and 2.5-5.7cm in width, shape is elliptic, lanceolate, apex is acuminate, surface glabrous or the young leaves puberulous, base acute or rounded, main nerves 6-12 pairs, petioles are 3-4mm in length. Flowers are white and fragrant, arranged in lax terminal cymes about 12.5cm diameter with slender dichrotomous branches; minute ovate bracts, glabrous calyx, glandular inside; segments 2.5mm in length, oblong, apex rounded and with membranous margins. Corolla is short, tube 3mm long; obtuse; corona of numerous linear scales, some inserts with the filaments & some on the corolla lobes. Fruits, 25-50cm(l) 6-8mm(w), cylindrical, tapering to both ends, glabrous striate, cohering at the tip. Seeds 1.3-2cm in length pointed at apex. From a Scottish physician and botanist William Wright (1740 - 1827) Wrightia is named.

The intention of the current review is to explore different parameters of plant like general description, distribution, chemical constituents, traditional

uses and to highlight pharmacological activities studied in recent literatures. The search strategy adopted for this purpose focused on the databases like pubmed, scopus and web of science from inception to 2021.

Fig.1 *W. tinctoria* flowers and pods.

1.1 Plant Profile

Synonym : Sweet Indrajao, Pala indigo plant, Dyers's oleander, Dantappala, Vetpala

Kingdom : Plantae

Order : Gentianales

Family : Apocyanaceae

Genus : Wrightia

Species : tinctoria

Origin : India, Burma

1.2 Geographical distribution:

Rajputana, Central provinces, Deccan, Konkan, S.M Country, Circars, W. Ghats of Madras presidency, Ceylon, Burma-Tunor[1].

1.3 Seasons

Leaves fall in December or January, renewed in April- may. Flowering happens after the leaves, mid May to late June. Fruit conspicuous in November, ripens by the following summer [2].

1.4 Uses

The bark and the seeds of Wrightia tinctoria used in bilious troubles & flatulence. Seeds are anthelmintic and aphrodisiac. Arthritic fevers are cured by both from the leaves and bark. Decoction prepared from the leaves and bark is stomachic. The dried and ground bark is rubbed over the body in dropsy. The fresh leaves are very pungent and are chewed for relief from tooth ache. The latex produced from the plant is cream in colour and its coagulum is used in code wire insulation, floor furnishings and adhesives. Fresh latex is proteolytic and curdles milk. Bark is especially useful in piles, skin diseases and bilious troubles. Bark is used as tonic[3].

The name Nelempala (Malayalam script) is used as Nelem (From Neelem) means blue & pala refers to plants like 'Alstoniascholaris' that produces milky (pal means milk) latex. When the leaves of this plant is kept in a glass of coconut oil for a few hours, the oil slowly turns blue in colour (after 12-18 hours its colour becomes brown & ultimately black.) The name indicates this property of Wrightia tinctoria. This blue coloured oil is used as an effective medicinal against psoriasis [4].

Decoction prepared from leaves and bark is used as tonic, stomachic and febrifuge (1 in 10) in doses of ½ to 2 ounces. It is given in combination with other vegetable bitters in bowel complaints, during convalescence from fevers and other acute diseases. Seeds are sweet and tonic and are given in seminal weakness [5].

2. PHARMACOLOGICAL ACTIVITY STUDIES

2.1 Antimicrobial activity

Kannan et al studied *Wrightia tinctoria* leaf extracts screened against skin bacteria and dermatophytes by in vitro. The hexane, methanol and ethanol extracts were tested using agar dilution method and broth micro dilution method. The Methanol and ethanol extracts showed antibacterial activity. The minimum inhibitory concentration of methanol and ethanol extracts were found to be 0.5 mg/ml for Bacillus subtilis and Staphylococcus epidermidis and 0.25 mg/ml for Staphylococcus aureus. The hexane extract was active against Trichophyton rubrum and Trichophyton tonsurans at 2 mg/ml. [6].

N Al Zaqri et al synthesised zirconium oxide nanoparticles using *Wrightia tinctoria* leaf extract. Green synthesis method was used for the synthesis of ZrO2- NPs. ZrO2-NPs formation was confirmed by XRD spectra analysis and DLS. Zeta potential revealed well stabilized ZrO2-NPs and it exhibited 94% degradation for RY 160 dye. ZrO2-NPs using *Wrightiatinctoria* leaf extract showed remarkable antibacterial activity[7].

2.2 Antiulcer activity

Wrightia tinctoria methanolic extract (TM) and *Wrightia tinctoria* 70% ethanolic extract (T70E) were studied for antiulcer activity and wascompared with carboxy methyl cellulose, pylorus control, Aspirin and standard famotidine. Aspirin plus pylorus ligation induced ulcer

model was used for the study. *Wrightia tinctoria* crude extract exhibited excellent antiulcer activity against experimentally induced acute gastric ulcer model [8].

2.3 Anticancer activity

S Ramalakshmi et al studied the anticancer property of the leaves of *Wrightia tinctoria* on HeLa Cells. The methanolic extract was evaluated by in-vitro method for cytotoxic effect by employing MTT assay. The potency of each concentration was calculated in terms of percent decrease in viable HeLa cells and compared to the control value.. At 76.1 µg/ml crude extract showed antiproliferativeactivity(IC50).The extract showed dose dependent anticancer effect[19].

2.4 Antiinflammatory activity

PR Tharkar et al investigated the anti-inflammatory activity of bark of *Wrightia tinctoria* by carrageenan- induced rat paw oedema and cotton pellet induced granuloma method. The various extracts showed inhibition of rat paw oedema at dose of 200mg/kg and also showed granuloma changes when compared to control group. Diclofenac sodium (13.5 mg/kg /b w, p.o) was used as the standard for comparison[10].

NA Aleykutty et al studied the dried leaves of *Wrightia tinctoria* for anti-inflammatory and analgesic effects. Antiinflammatory activity was studied by using HRBC membrane stabilization method and carrageenan induced rat paw oedema model. Ethyl acetate fraction exhibited 67.21% protection in rat paw oedema model at a concentration of 400mg/kg. Ethyl acetate fraction also showed remarkable analgesic potential when studied using hot plate method and acetic acid induced writhing in mice [11].

2.5 Antidiabetic activity

AK Shukla and Papiya Bigoniya studied the effect of total flavonoid isolated from W. tinctoria seed on alloxan induced diabetic model by assessing body weight change, relative organ weight, BG level, and serum lipid parameters. The effect of W. tinctoria seed flavonoid fraction was not significant on hyperglycemia and other disturbed biochemical parameter induced by alloxan, but it has significant effect on normalization of serum creatinine level and lowering of TG and relative weight of liver indicating possible presence of kidney and liver protective property [12].

R Asok Raj et al evaluated petroleum ether extract of Wrightiatinctoria for hypoglycaemic activity in Alloxan-induced diabetic rats. The extract exhibited reduction of serum glucose levels (74.39%) at the dose of 400 mg/kg [13].

2.6. Antifungal activity

K Ponnusamy et al investigated the in vitro antifungal activity of leaf extracts and indirubin, an important constituent of *Wrightia tinctoria*. Leaf extracts showed promising activity against dermatophytic and non-dermatophytic fungi. At dose of 0.5 mg/ml leaf extract was active against Trichophyton rubrum, Epidermophyton floccosum, Aspergillus niger and

Scopulariopsis brevicaulis. Whereas Indirubin, exhibited activity against dermatophytes such as,Trichophyton rubrum,Trichophyton simii, Epidermophyton floccosum, Trichophyton mentagrophytes and Trichophyton tonsurans. Indirubin also exhibited activity against Cryptococcus sp, Aspergillus niger, and Candida albican[14].

2.7. Wound healing activity

M Yariswamy et al evaluated the wound healing potential of *Wrightia tinctoria* latex protease. Excision wound model in mice was used to evaluate the healing potential. Neosporin was used as the standard drug for comparison. The progression of healing was observed using histological examinations, wound contraction, collagen content, catalase and MMP activity. Re-establishment of skin structure, complete epithelialisation and accelerated wound healing were observed by histological examination on day 9 which confirmed the wound healing effect [15].

2.8. Antipsoriatic activity

Antipsoriatic activity of *Wrightia tinctoria* extract was evaluated by mouse tail test. Longitudinal sections of tail skin were prepared and it was stained with hematoxylineosin. Histometrical analysis of specimens showed potent activity of extract (63.94%) than standard isoretinoic acid (48.52%). Both standard and sample increased the epidermal thickness when compared to control[16].

2.9. Post coital interceptive activity

G Keshri et al worked on post coital interceptive activity in *Wrightia tinctoria*. 250-mg/kg dose of ethanolic extract of the stem bark inhibited pregnancy in 100% of rats on Days 1–7 or 1–5 postcoitum. The hexane, chloroform fractions and water soluble and water-insoluble fractions showed 100% anti-implantation effect. The n-butanol fraction intercepted pregnancy only in 75% of animals.They concluded that estrogen-agonistic activity of the active ethanolic extract and its fractions might be responsible for their contraceptive action [17].

2.10. Antioxidant activity

H Jamshed et al screened *Wrightia tinctoria* leaves and seeds for antioxidant potential. The extracts were evaluated using free radical scavenging assays like DPPH and ABTS. Reducing power abilities of extracts were also noted by fluorescence recovery after photobleaching [FRAP] and TAC. In DPPH method IC50 was 45.4 µg/ml and TAC50 mg GAE/g. In ABTS *Wrightia tinctoria* showed IC50 31.7 µg/ml and FRAP 2.5 mMol Fe+2/g.The results confirmed significant antioxidant potential of *Wrightia tinctoria* compared to other antioxidant-rich medicinal plants [18].

S Ramalakshmi et al reported the antioxidant potential of *Wrightia tinctoria* flower extract. 2,2-Diphenyl-1-Picrylhydrazyl method showed IC50 at 43.16µg/mL and by phosphomolybednum method the extract showed IC50 at 124.07 mg AAE/100g[19].

2.11. Anthelmintic activity

A Sruthi et al reported anthelmintic activity of crude petroleum ether and chloroform extracts of leaves of *Wrightia tinctoria* when studied in Pheretima posthuma. . Piperazine citrate was used as standard drug and normal saline given as control. Time of paralysis and death of the worms were observed using three concentrations (2.5, 5.0, 7.5 mg/ml) of both extracts. The experiment proved the potential benefit of *Wrightia tinctoria* leaves as an anthelmintic agent [20].

2.12. Antinociceptive activity

YS Reddy et al screened the bark of *Wrightia tinctoria* for antinociceptive activity.The ethyl acetate, acetone and methanol extracts were used in the study using acetic acid-induced writhing in mice. The standard drug used was acetyl salicylic acid. The extracts exhibited activity comparable to that of standard drug [21]. P Bigoniya et al reported dose-dependent antinociceptive effects in ethanolic bark extract of *Wrightia tinctoria* p.o. when given in normal rats. The analgesic effect was observed against thermal and chemical noxious stimuli, but not observed against the mechanical stimulus[22].

2.13. Hepatoprotective activity

P Bigoniya et al investigated hepatoprotective effect on isolated triterpene compounds from *Wrightia tinctoria* such as lupeol, b –amyrin and b –sitosterol. The method adopted was CCl 4 –induced hepatotoxicity in the rat. Silymarin was used as the standard drug. Animals were pretreated with triterpene fractions at concentrations of 125, 250 and 400 mg/kg, p.o. once a day for 4 days and then CCl 4 was given. The administration of drug was continued for next 3 days. The results indicated that the CCl 4 –induced acute increase in serum SGOT, SGPT and ALP were attenuated and histopathological alterations were markedly decreased [23].

NV Patil et al evaluated various extracts of *Wrightia tinctoria* leaves for hepatoprotective activity. Hepatotoxicity was induced by using CCl4 and the animals were sacrificed and analysed for biochemical variations like ALP, SGPT, SGOT and bilirubin etc. The methanolic extract exhibited maximum activity whereas aqueous extract found to have minimum activity [24].

2.14. Antiviral activity

P Selvam et al reported anti HIV activity of extracts of *Wrightia tinctoria* and was tested for it's inhibitory effects against the replication of HIV-1(IIIB) in MT-4 cells. Inhibitory effect of extracts were monitored by inhibition of virus-induced cytopathic effect in MT-4 cells and was measured by MTT assay. The different extracts have been evaluated for anti HIV activity in acutely infected MT-4 cells. None of the extracts exhibited anti HIV activity in acutely infected MT-4 cells. At subtoxic concentration extract exhibited a maximum protection of 48% of the MT-4 cells against the cytopathic effect of HIV-1(IIIB) [25].

2.15. Immunomodulatory activity

S Sathianarayanan et al reported the effect of methanol extract of *Wrightia tinctoria* leaves on the primary and secondary antibody responses which were evaluated by the humoral antibody response for a specific immune response. The neutrophil activation was studied by neutrophil adhesion test. *Wrightia tinctoria* showed a marked increase in the primary and secondary humoral antibody responses at doses of 100 and 200mg/kg/bw by increasing the hemagglutinating antibody titre. It also showed a considerable increase in the percentage neutrophil adhesion at doses of 200mg/kg/bw. It also delayed hypersensitivity response in the increasing doses[26].

2.16. Absence of central activity

P Bigoniya and AC Rana evaluated *Wrightia tinctoria* for central activity. The study reported that the ethanolic extract did not possess any significant effect on pentobarbitone-induced hypnosis. So the study concluded that the extract is devoid of any of the protective effect against leptazole- or MES-induced convulsions at any of the tested doses[27].

2.17. Antispasmodic and antidiarrhoeal activity

P Bigoniya et al screened *Wrightia tinctoria* bark for antispasmodic and antidiahhoeal activity. The bark extract and isolated steroidal alkaloid from ethanol extract was investigated on different experimentally induced diarrhoea models of rats such as isolated rat ileum, and on enteric bacterium. The extract at 500 and 1000 mg/kg dose inhibited the frequency and wetness of faecal droppings in castor oil-induced diarrhea. The isolated steroidal alkaloid at 50 and 100 mg/kg dose exhibited the effect. Both extract and steroidal alkaloid decrease propulsion of charcoal meal and as well reduced prostaglandin E2-induced enteropooling. The frequency,amplitude and tone of spontaneous gut movement were reduced by the alkaloid.It also inhibited acetylcholine (Ach)- induced contraction of rat ileum[28].

2.18. Larvicidal activity

M Sakthivadivel et al worked on larvicidal potential of *Wrightia tinctoria* fruits and leaves.The crude aqueous and petroleum ether extracts were tested on filarial vector, Culex quinquefasciatus at concentrations of 0.06%, 0.12%, 0.25%, 0.50% and 1.00%. Mortality of Larvae was observed for 24 and 48h. The aqueous fruit extract exhibited highest larvicidal activity with LC50 values of 0.17% and 0.09% followed by aqueous leaf extract at 0.21% and 0.11%[29].

Table 1: Pharmacological activities of various parts of *W.tinctoria.*

Activity studied	Plant parts used	References
Antisporiatic activity	leaves	SP Dhanabal et al [30]
Antifungal	leaf	K ponnusamy et al [15]
Antifungal	leaf	KV Devika et al [31]
Antibacterial and	leaf	M Rajani et al [32]

antifungal		
Antibacterial	leaf	P Kannan et al [7]
Antibacterial	leaf	K Moorthy et al [33]
Antibacterial	bark	MS Kyade et al [34]
Antibacterial	leaf	S Sridhar [35]
Antibacterial	leaf	S Ravisankar et al [36]
Cytotoxic	Stem bark	Chaudhary S et al [37]
Cytotoxic	Flowers	S Ramalakshmi et al [38]
Cytotoxic	Leaves	Sophiya R et al [39]
Anti-inflammatory	Stem bark	Tharkar et al, 2010 [11]
Anti-inflammatory	Stem bark	Jain and Bari, 2010 [40]
Anti-inflammatory	Leaf	Aleykutty et al, 2011[12]
Anti-inflammatory	Leaf	Rajalakshmi and Harindran [41]
Anti-diabetic	Leaf	RA Raj et al, 2010 [14]
Anti-diabetic	leaf	RajaniSrivastava [42]
Anti-diabetic	Pods	MS Rani et al [43]
Analgesic	leaf	Aleykutty et al, 2011[12]
Analgesic	Stem ark	Reddy et al, 2002 [22]
Hepatoprotective	Stem bark	Bigoniya and Rana, 2010 [24]
Hepatoprotective	Seed and leaves	H Jamshed et al [19]
Helmintholytic	leaf	SR Dore et al [44]
Anthelmintic	leaf	A Sruthi et al [21]
Anthelmintic	leaf	GR Rajalakshmi et al [45]
Wound healing	latex	M Yariswami et al [16]
Wound healing	latex	VP Veerapur et al [46]

3. PHYTOCHEMISTRY

The phytochemical constituents of pods without seeds are cycloartenone, cycloartanes, cycloeucalenol besides alpha and beta amyrin, terpene wrightial, oleanolic acid,ursolic acid

and the betasitosterol. Beta amyrin is also present in leaves and stem bark. Stem bark also contains lupeol and beta sitosterol[47].

3.1 Phytochemical studies

Preliminary phytochemical analysis of *Wrightia tinctoria* methanolic extract showed that it contains alkaloids and flavones.The analysis of *Wrightia tinctoria* methanolic extract was conducted using different analytical instruments like UV, HPLC,TLC and GC revealed that indole derivatives like indurubine and isatin were present. The outcome of Gas Chromatographic analysis showed the presence of myristic acid, behenic acid and palmitoleic acid[48].

The phytochemical investigation of the bark of *Wrightia tinctoria* showed the existence of alkaloids, phenolics, saponins, tannins terpenoids, steroids, triterpenoids, flavonoids and carbohydrates[49].

Similarly S Sridhar et al found out that carbohydrates, steroids, phenols, saponins, flavonoids,tannins and proteins were present in the leaves of *Wrightia tinctoria* [36].

A study investigated by SR Sankar et al indicated that alkaloids, terpenoids, glycosides, flavanoids, saponins and phlobatannins were present on the leaves[37].

In another study the researchers isolated and detected indirubin, rutin, tryptanthrin, Indigotin, isatin, and anthranillate as vital constituents of *Wrightia tinctoria* by HPTLC, HPLC, UV-VIS, IR and EI-MS. Indigotin is present in fresh leaves of live plants and indirubin is formed during drying after collecting of the leaves. This conversion is due to hydrolytic reaction and auto-oxidation. The variation of chemical constituents of leaves due to seasonal changes was investigated using HPTLC and HPLC techniques, and showed a steady rise in indigotin-indirubin concentration from August to November, whereas isatin and anthranillate concentration rose during December and January. Autoxidation of indigotin resulted in the production of isatin[50].

Along with the phytocompounds, *W.tinctoria* is also found to contain important enzyme. Proteases are commercially important class of enzymes and the hydrolytic property of the enzyme is exploited in various biotechnological processes.The researchers isolated Wrightin, serine protease, from the sap of *Wrightia tinctoria*, which is an economical source of protease for commercial use. The plant also has Wrightial, a triterpenoid, besides, Cycloartenone, Cycloeucalenol, β amyrin, and β-sitosterol as phytocompounds[51].

By comparing a synthetic genuine molecule to a novel sterol isolated from the unsaponifiable lipid of Wrightia tinctoria seed, it was determined to be 14-methylzymosterol. Desmosterol, clerosterol, 24-methylene-25-methylcholesterol, and 24-dehydropollinastanol, four rare sterols, were extracted and identified [52].

To concentrate the milk clotting proteases, a study conducted by Rajagopalan et al isolated proteases from Wrightia tinctoria bark and partially purified them using a non-chromatographic approach called three phase partitioning (TPP). The interfacial phase (IP)

with 60 percent ammonium sulphate and 1:1 crude enzyme to t-butanol yielded the highest recovery and purification fold of protease activity. The enzyme fraction's optimal pH and temperature were found to be 7.5 and 50 degrees Celsius, respectively. Inhibition studies revealed its serine nature. Non-denaturing PAGE, Zymography, and 2D PAGE of IP revealed the existence of three caseinolytic proteases with molecular weights of 95.62 kDa, 91.11 kDa, and 83.23 kDa, respectively, and pI values of 3.89, 5.45, and 5.43. IP in both aqueous and lyophilized form was exceptionally stable, retaining full activity for 3 weeks at 4 °C[53].

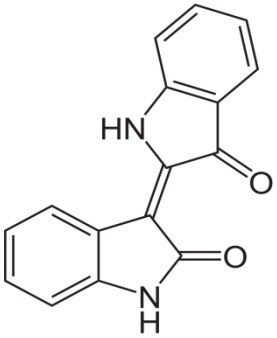

Fig 2: Indirubin

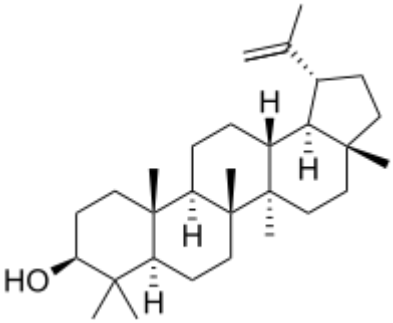

Fig 3: lupeol

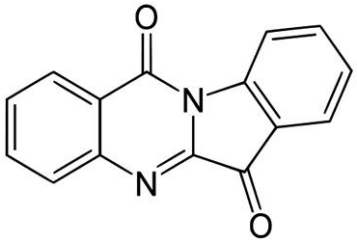

Fig 4: Tryptathrin

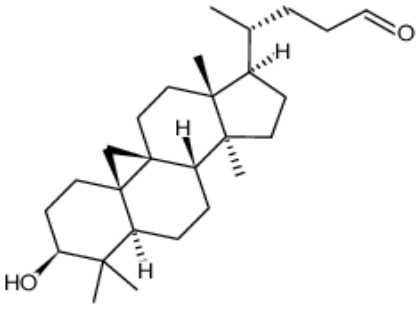

Fig 5: Wrightial

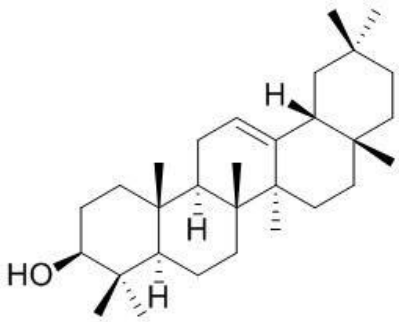

Fig 6: Beta amyrin

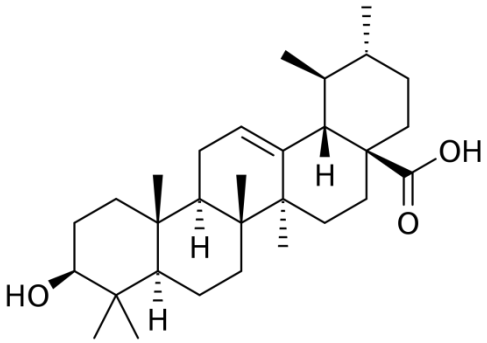

Fig 7: Ursolic acid

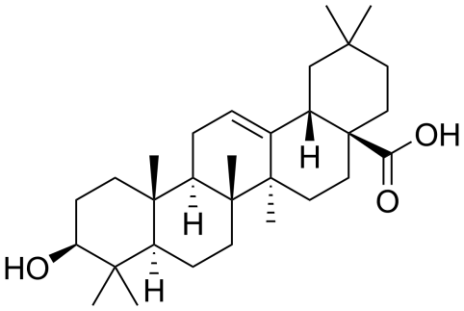

Fig 8: Oleanolic acid

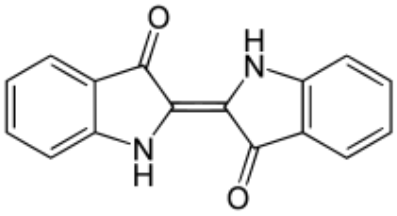

Fig 9: Indigotin

4. CONCLUSION

Use of*Wrightiatinctoria*inayurvedic and siddha system of medicine for its effects against psoriasis and epidermal thickening and drying problems. It is added in hair oil preparations as it effectively minimises dandruff. The pharmacological studies proves its pharmacological significance such as antiviral, anti-inflammatory, cytotoxic, hepatoprotective, wound healing, post coital interceptive, anthelmintic, antinociceptive, antioxidant, antiviral, antifungal, antibacterial , antidandruff and antipsoriaticactivity.Total flavonoid isolated from W. tinctoria seed lack hypoglycemic effect.

Alkaloids, saponins, terpenoids, flavones,triterpenoids, tannins, steroids, carbohydrates, glycosides, Indole derivatives like isatin and indurubine, and fixed oils like myristic acid,palmitoleic acid, behenic acid, acid indigoid compounds reflects its phytochemical abundance. So the present study suggests that the proved phytochemical and biological characteristics makes *Wrightia tinctoria* a promising drug to the pharmaceutical industries and a good candidate for more exploration to the future.

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DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

REFERENCE

1. Kirtikar, Basu. Indian medicinal plants, п nded. Allahabad: Book sellers and publishers; 1987. 2: p. 1581-1583.
2. Krishnan P. Trees of Delhi, a field guide. Delhi tourism: 2006. p-137.
3. Warrier PK, Nambiar YPK, Ramankutty C. Indian Medicinal Plants a compendium of 500 species.Madras: Orient Longman Limited;2006.5: p.417-419.
4. Van rheede's `hortusmalabaricus' (Malabar garden); English ed. 9: Thiruvanadapuram, University of Kerala, 2003; p. 7-10
5. Dr. K M Nandakarni's Indian MateriaMedica. Vol. 1. p. 1297.
6. P. Kannan , B. Shanmugavadivu , C. Petchiammal , and W. Hopper. In vitro antimicrobial activity of *Wrightiatinctoria* leaf extracts against skin microorganisms. ActaBotanicaHungarica. Volume 48: Issue 3-4.Pages: 323–329
7. Al-Zaqri N, Muthuvel A, Jothibas M, Alsalme A, Alharthi FA, Mohana V. Biosynthesis of zirconium oxide nanoparticles using *Wrightiatinctoria* leaf extract: Characterization, photocatalytic degradation and antibacterial activities. Inorganic Chemistry Communications. 2021 May 1;127:108507.
8. Madhu.C. Divakar and Lakshmi Devi. S. Antiulcer activity of *Wrightiatinctoria*(Roxb.) R.Br. Der Pharmacia Sinica, 2011, 2 (2): 355-360.
9. Ashish Dixit*, A.K. Jain and MukulTailang. An in-vitro evaluation of cytotoxic activity of *Wrightiatinctoria.* Int J Pharm 2017; 7(4): 14-18.
10. P. R. Tharkar*, A.U. Tatiya, S.J. Surana, N. S. Bhajipale, S. R. Deore. Anti-inflammatory Study of *Wrightia tinctoria*R.Br Stem Bark in Experimental Animal

Models. International Journal of PharmTech Research CODEN (USA): IJPRIF ISSN : 0974-4304.Vol. 2, No.4, pp 2434-2437.

11. N.A. Aleykutty, A.R. Bindu *, S. Sangeetha, G. Jiljit. Evaluation of Anti-inflammatory and Analgesic Activity of *Wrightiatinctoria*Leaves. Journal of Biologically Active Products from Nature (JBAPN)1 (1) 2011 pp 33 – 41.

12. Shukla AK, Bigoniya P. Lack of hypoglycemic activity in total flavonoid fraction of *Wrightiatinctoria* on alloxan induced hyperglycemia. Journal of Pharmaceutical Negative Results. 2014 Jan 1;5(1):34-8.

13. Raj RA, Kumar AS, Gandhimathi R. Hypoglycemic and hypolipidemic activity of *Wrightiatinctoria* L. in alloxan induced diabetes in albino wistar rats. Pharmacologyonline. 2009;3:550-9.

14. Ponnusamy K, Petchiammal C, Mohankumar R, Hopper W. In vitro antifungal activity of indirubin isolated from a South Indian ethnomedicinal plant *Wrightiatinctoria* R. Br. Journal of ethnopharmacology. 2010 Oct 28;132(1):349-54.

15. Yariswamy M, Shivaprasad HV, Joshi V, Urs AN, Nataraju A, Vishwanath BS. Topical application of serine proteases from *Wrightiatinctoria* R. Br.(Apocyanaceae) latex augments healing of experimentally induced excision wound in mice. Journal of ethnopharmacology. 2013 Aug 26;149(1):377-83.

16. Raj BA, Muruganantham N, Praveen TK, Raghu PS. Screening of *Wrightiatinctoria* leaves for Anti psoriatic activity. Hygeia.J.D.Med. April2012 –Sept 2012.vol.4 (1): 73-78.

17. Keshri G, Kumar S, Kulshreshtha DK, Rajendran SM, Singh MM. Postcoital interceptive activity of *Wrightiatinctoria* in Sprague–Dawley rats: a preliminary study. Contraception. 2008 Sep 1;78(3):266-70.

18. Jamshed H, Siddiqi HS, Gilani AU, Arslan J, Qasim M, Gul B. Studies on antioxidant, hepatoprotective, and vasculoprotective potential of Viola odorata and *Wrightiatinctoria*. Phytotherapy Research. 2019 Sep;33(9):2310-8.

19. Ramalakshmi S, Edaydulla N, Ramesh P, Muthuchelian K. Investigation on cytotoxic, antioxidant, antimicrobial and volatile profile of *Wrightiatinctoria* (Roxb.) R. Br. flower used in Indian medicine. Asian Pacific Journal of Tropical Disease. 2012 Jan 1;2:S68-75.

20. Shruthi A, Latha KP, Vagdevi HM, Vaidya VP, Pushpa B, Shwetha C. In vitro anthelmintic activity of leaves extract of *Wrightiatinctoria*. International Journal of ChemTech Research. 2010;2(4):2043-5.

21. Reddy YS, Venkatesh S, Ravichandran T, Murugan V, Suresh B. Antinociceptive activity of *Wrightiatinctoria* bark. Fitoterapia. 2002 Aug 1;73(5):421-3.

22. Bigoniya P, Shukla A, Agrawal GP, Rana AC. Pharmacological screening of *Wrightiatinctoria* bark hydro-alcoholic extract. Asian J Exp Sci. 2008;22(3):235-44.

23. Bigoniya P, Rana AC. Protective effect of *Wrightiatinctoria* bark triterpenoidal fraction on CCl4-induced acute rat liver toxicity. Iranian Journal of Pharmacology and Therapeutics. 2010 Sep 10;9(2):55-0.

24. Patil NV, Bhosale AV, Ubale MB. EVALUATION OF HEPATOPROTECTIVE ACTIVITY OF *WRIGHTIA TINCTORIA* IN CARBON TETERACHLORIDE INDUCED RATS. Advances in Pharmacology & Toxicology. 2012 Dec 1;13(3).

25. Selvam P, Murugesh N, Witvrouw M, Keyaerts E, Neyts J. Studies of antiviral activity and cytotoxicity of *Wrightiatinctoria* and Morindacitrifolia. Indian journal of pharmaceutical sciences. 2009 Nov;71(6):670.

26. Sathianarayanan S, Rajasekaran A. Immunomodulatory activity of ethanolic extract of *Wrightiatinctoria* leaves. immunity. 2012 Aug 1;5:8.

27. Sathyanarayanan S, Selvam P, Jose AS, George RM, Revikumar KG, Neyts J. Preliminary phytochemical screening and study of antiviral activity and cytotoxicity of *Wrightiatinctoria*. Int J Chem Sci. 2009;7(1):1-5.

28. Bigoniya P, Rana AC. Antidiarrheal and antispasmodic activity of *Wrightiatinctoria* bark and its steroidal alkaloid fraction. Pharmacologyonline. 2009;3:298-310.

29. Sakthivadivel M, Gunasekaran P, Annapoorani JT, Samraj DA, Arivoli S, Tennyson S. Larvicidal activity of *Wrightiatinctoria* R. BR.(Apocynaceae) fruit and leaf extracts against the filarial vector Culexquinquefasciatus Say (Diptera: Culicidae). Asian Pacific Journal of Tropical Disease. 2014 Jan 1;4:S373-7.

30. Dhanabal SP, Raj BA, Muruganantham N, Praveen TK, Raghu PS. Screening of *Wrightiatinctoria* leaves for anti psoriatic activity. Hygeia-Journal for Drugs and Medicine. 2012;4(1):73-8.

31. Devika KV, Sabarinathan T, Shamala S. Antifungal Efficacy of *Wrightiatinctoria* (Roxb.) R. Br on Candida Species Isolated from the Oral Cavity: an Invitro Study. Journal of Orofacial Sciences. 2021 Jan 1;13(1):67.

32. Ranjani M, Deepa S, Kalaivani K, Sheela P. Antibacterial and antifungal screening of ethanol leaf extract of *Wrightiatinctoria* against some pathogenic microorganisms. Drug Invention Today. 2012 May 1;4(5).

33. Moorthy K, Aravind A, Punitha T, Vinodhini R, Suresh M, Thajuddin N. In vitro Screening of antimicrobial activity of *Wrightiatinctoria* (Roxb.) R. Br. Asian J Pharm Clin Res. 2012;201(5):4.

34. Khyade MS, Vaikos NP. Comparative phytochemical and antibacterial studies on the bark of *Wrightiatinctoria* and Wrightiaarborea. Int J Pharm Bio Sci. 2011;2(1):176-81.

35. Sridhar S, Kamalakannan P, Elamathi R, Deepa T, Kavitha R. Studies on antimicrobial activity, physio-chemical and phytochemical analysis of *Wrightiatinctoria* R. Br. IJPRD. 2011;3(8):139-44.

36. Shankar SR, Rangarajan R, Sarada DV, Kumar CS. Evaluation of Antibacterial Activity and Phytochemical Screening of *Wrightiatinctoria* L. Pharmacognosy Journal. 2010 Sep 1;2(14):19-22.

37. Chaudhary S, Devkar RA, Bhere D, Setty MM, Pai KS. Selective cytotoxicity and pro-apoptotic activity of stem bark of *Wrightiatinctoria* (Roxb.) R. Br. in cancerous cells. Pharmacognosy magazine. 2015 Oct;11(Suppl 3):S481.

38. Ramalakshmi S, Edaydulla N, Ramesh P, Muthuchelian K. Investigation on cytotoxic, antioxidant, antimicrobial and volatile profile of *Wrightiatinctoria* (Roxb.) R. Br. flower used in Indian medicine. Asian Pacific Journal of Tropical Disease. 2012 Jan 1;2:S68-75.

39. Sophia R, MohanaRao K, LekshmiNath R, Shabna A, Murty MS, Saikia M, Chandran H, Paul A, Antony J, Joseph SM, Vinod V. DW-F-5: A novel formulation against malignant melanoma from *Wrightiatinctoria*.

40. Jain PS, Bari SB. Anti-inflammatory effects of wood stem extracts of *Wrightiatinctoria*. Asian J Tradit Med. 2010 Aug 20;5:132-7.

41. Rajalakshmi GR, Harindran J. Anti-inflammatory activity of Samaderaindica leaves by membrane stabilization. International Journal of Pharmaceutical Sciences and Research. 2013 Feb 1;4(2):721.Srivastava R. A review on phytochemical, pharmacological, and pharmacognostical profile of *Wrightiatinctoria*: Adulterant of kurchi. Pharmacognosy reviews. 2014 Jan;8(15):36.

42. Srivastava R. A review on phytochemical, pharmacological, and pharmacognostical profile of *Wrightiatinctoria*: Adulterant of kurchi. Pharmacognosy reviews. 2014 Jan;8(15):36.

43. Rani MS, Pippalla RS, Mohan GK, Raju AB, Kumar VH. In vitro study of methanolic extracts of Dodonaeaviscosa Linn and *Wrightiatinctoria* R. Br. on glucose uptake by isolated rat hemi-diaphragm. Int J Chem Sci. 2012;10:1724-30.

44. Tare HL, Gore MS, Deore SR, Bidkar JS, Dama GY. Comparative hemintholytic potential of extracts obtained from Cymbopogoncitratus and *Wrightiatinctoria* leaves. International Journal of Pharma and Biosciences. 2011;2(1):321-6.

45. Rajalakshmi GR, Harindran J. In vitro anthelmintic activity of *Wrightiatinctoria*. In vitro. 2013 Apr;5(2):308-10.

46. Veerapur VP, Palkar MB, Srinivasa H, Kumar MS, Patra S, Rao PG, Srinivasan KK. The effect of ethanol extract of *Wrightiatinctoria* bark on wound healing in rats. Journal of Natural Remedies. 2004 Jun 1;4(2):155-9.

47. Khare CP. Indiam Medicinal plants An Illustrated Dictionary. New Delhi: Springer; 2007.p.720.

48. Khyade MS, Vaikos NP. Comparative phytochemical and antibacterial studies on the bark of *Wrightiatinctoria* and Wrightiaarborea. Int J Pharm Bio Sci. 2011;2(1):176-81.

49. Rao B, Rajeswari D, Devarakonda R, Battu H. Phytochemical and Pharmacological Studies on *Wrightiatinctoria*. World J. Pharm. Pharm. Sci. 2019.

50. Muruganandam AV, Bhattacharya SK, Ghosal S. Indole and flavanoid constituents of *Wrightiatinctoria*, W. tomentosa and W. coccinea. nopr.niscair. Feb-2000. Vol.39B(02): 125-131.

51. Tomar R, Kumar R, Jagannadham MV. A stable serine protease, wrightin, from the latex of the plant *Wrightiatinctoria* (Roxb.) R. Br.: purification and biochemical properties. J Agric Food Chem. 2008 Feb 27;56(4):1479-87.

52. Akihisa T, Ahmad I, Singh S, Tamura T, Matsumoto T. 14α-Methylzymosterol and other sterols from *Wrightiatinctoria* seeds. Phytochemistry. 1988 Jan 1;27(10):3231-4.

53. Rajagopalan A, Sukumaran BO. Three phase partitioning to concentrate milk clotting proteases from *Wrightiatinctoria* R. Br and its characterization. International journal of biological macromolecules. 2018 Oct 15;118:279-88.