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RESEARCH ARTICLE

Antifungal Activity of Oxidized Essential Oil of *Chloroxylon Swietenia* Roxb. Corom

T. Telang*, S. K. Awasthy and P. Oswal

Ex. H.O.D. & Associate Prof. Chemistry, R.N. Modi Engineering college, Ranpur, Kota, (Rajasthan), India.

ABSTRACT:

The essential oil from the leaves of *C. swietenia* has been oxidized using Hydrogen peroxide and studied for antifungal efficacy against thirteen fungi viz. *Aspergillus oryzae, A.terreus, A.niger, Curvularia prasadii, Rhizopus nodosus Candida albicans* etc. The oxidized essential oil was found effective against all fungi but found more effective against *Aspergillus oryzae, A.terreus, Curvularia prasadii, Candida albicans* and *Trichoderma viridi.*

KEY WORDS: Hydro distillation, essential oil, Thin Layer Chromatography (TLC), Gas Chromatography-Mass Spectroscopy (GC-MS), chemical composition, antifungal evaluation.

INTRODUCTION:

The interest of the essential oil bearing plants is increasing because of necessity of finding safer microbicides in combination with the need of preventing environmental degradation and pollution (Sivropoulou et al 1995)[1].

C. swietenia (Ghiria or Satinwood) is cultivated in dry deciduous forest throughout India at an altitude of 1000 to 1500 m. It is 9 to 15 m high tree and found in Ceylon and India. The decoction of leaves is reported to use as a lotion for ulcer and for healing abrasion of the skin. The leaves are also prescribed in rheumatism (Anonymous 1950)[2].

The antifungal efficacy of essential oil from the leaves of *C. swietenia* was studied (Garg and Oswal 1982)[3] but no study had been reported on the antifungal efficacy of oxidized essential oil. The components identified in the oxidized essential oil using Column chromatography and GC-MS are α-pinene, α-terpinene, limonene, Δ_3-carene, β-phellandrene, camphene, α-terpeniol, linalool, β-caryophyllene oxide, geraniol, geranyl acetate, methyl cynnamate copaene, naphthalene hexahydro dimethyl and cyclo buta 1,2 : 3,4 dicyclopentene (Telang et al 2003)[4]. As oxidized essential oil contain more oxygenated compounds, diseases cannot exist in oxygen rich environment. With this intention the oxidized essential oil was screened for its antifungal efficacy.

Material and method:-

The shade dried leaves of C. swietenia were hydrodistilled in the modified Clevenger's apparatus. The oil was dried over anhydrous sodium sulphate and oxidized using 30 volumes Hydrogen peroxide. The unoxidsed and oxidized essential oils of C. *swietenia* were studied by different chromatographic techniques i.e. Thin Layer Chromatography (TLC), Column Chromatography, Gas Liquid Chromatography (GLC), and Gas Chromatography – Mass Spectroscopy (GC-MS). The components and their percentages of concentration presents in unoxidised and oxidised essential oils are recorded in the **Table –1**.

The sample of oxidized essential oil was tested in the Pathological Laboratory Betul for its antifungal efficacy using Culture Media Sabouraud's medium[5] by adapting following procedure.

PROCEDURE:

Culture media Sabouraud's 60 medium was used for the preparation of inoculums given as below ;-

Sr. No.	Component	Composition gm./liter (in hot distilled water)
1	Dextrose	40.00
2	Peptone	10.00

PREPARATION OF SLANTS AND MEDIUM:

2% Agar was added to sabouraud's medium and used for the preparation of slants and medium. The composition of Agar medium was as follows:

Sr. No.	Component	Composition gm./litre(in hot distilled water)
1	Dextrose	40.00
2	Peptone	10.00
3	Agar	20.00

Medium, culture tubes, petrifies and other materials in use were sterilized in an autoclave at 15lbs/sq.inch steam pressure for 30 minutes.

INCUBATION:

Culture tubes are seeded agar plates were kept in an incubator at 30^0C.The patridishes were kept in a cool

***Corresponding author**: Dr. Tulika Telang **/Email**: bhatt.tulika9@rediffmail.com

room at 25^0C for seven days till the optimum growth of fungi take place.

PREPARATION OF PLATES:

The different pathogenic fungal species were subcultured on sterililized sabouraud 's Agar medium for every test. Suspension of subcultured organism in Sabourud's medium was prepared at 20 ml. of it were added two each petridish and mixed uniformly.

ACTIVITY MEASUREMENT:

The sterilized Whatman's filter paper no,1 disc was thoroughly moistened in the pure oxidized essential oil. The three different sterilized Whatman's filter paper no.1 discs were moistened with different dilute solutions- 1:50, 1:100 and 1:200. The solvent used to prepare different dilution was Tween 80. Four test discs along with control disc previously moistened in 2% Resorcinol solution in water were placed on each seeded Agar plate and incubated in cool room for 72 hours. The experiments were performed in triplicate and average Zones of inhibition were recorded in Table no.2. Table no.3 indicates the comparative record of antifungal activity of pure unoxidised and pure oxidized essential oils of C .swietenia, when compare to control (2%Resorcinol).

OBSERVATIONS: " Table 1, 2 & 3 here**"**

Table 1: Comparativ Study of Components of Unoxidsed and Oxidized Essential Oils of C. *swietenia*

Sr. No.	Name of Component	Percentage concentration of Component in Unoxidised Essential Oil	Percentage concentration of Component in Oxidised Essential Oil
1	α-Pinene	0.11	0.40
2	Camphene	0.78	0.42
3	Limonene	2.78	28.95
4	β-Pinene	0.08	-
5	Δ^3-Carene	3.17	1.94
6	Myrecene	0.83	-
7	β-Phellandrene	0.10	1.76
8	P-Cymene	0.93	-
9	α-Terpinene	9.29	4.30
10	α-Terpincol	12.5	4.34
11	Mythyl heptenone	12.29	-
12	Citral-a	4.05	-
13	Citral-b	2.22	-
14	Garaniol	1.05	1.97
15	Linalool	1.75	0.42
16	β-Caryophyllene oxide	18.40	47.73
17	Nerol	6.54	-
18	Geranyl acetate	3.34	0.22
19	β- Caryophyllene	5.40	-
20	α-Caryophyllene	2.83	-
21	Methyl cinnamate	3.22	0.07
22	α-Cadinene	6.20	-

Table 2: *In-vitro* antifungal activity of oxidized essential oil of *C. swietenia*

Sr. No.	Fungai	Zone of Inhibition (mm)				
		Pure Oil	1:50	1:100	1:200	Control (2%Resorci-nol)
1	**Trichoderma viride**	17	15	12	08	10
2	**Fusarium solani**	20	17	16	09	18
3	*Rhizopus nodosus*	34	24	16	10	30
4	*Aspergillus niger*	15	10	08	08	12
5	*A. fumigatus*	14	0	0	0	10
6	*A. flavous*	15	15	13	09	16
7	*A. oryzae*	42	28	23	10	21

8	*A. terreus*	30	29	22	16	20
9	*Trichophyton rubrum 5S*	10	06	05	05	08
10	*T. rubrum 12S*	22	19	15	10	22
11	*Curvularia prasadii*	37	30	22	10	17
12	*Candida utilus*	12	08	06	06	10
13	*C. albicans*	22	10	10	06	13

Table 3: Antifungal activity: Comparison of Oxidized and Unoxidised Essential Oils of C. swietenia with respect to control (2% Resorcinol)

Sr. No	Fungi	Maximum Zone of Inhibition (mm.) of test Fungi Against pure Oxidised Essential Oil	Maximum Zone of Inhibition (mm.) of test Fungi Against pure Unoxidised Essential Oil
1	*Aspergillus oryzae*	42	25
2	*Curvularia prasadii*	37	14
3	*Rhizopus nodosus*	34	12
4	*Aspergillus terreus*	30	30
5	*Trichophyton* 12S	22	11
6	*Candida albicans*	22	20
7	*Fusarium solani*	20	10
8	*Trichoderma viride*	17	09
9	*Aspergillus niger*	15	07
10	*Aspergillus flavus*	15	15
11	*Aspergillus fumigates*	14	09
12	*Candida utilus*	12	12
13	*Trichophyton rubrum* 5S	10	06

RESULTS AND DISCUSSION:

The data revealed (Table-2) that oxidized essential oil of *C. swietenia* had inhibitory action on the growth of test fungai. The activity of oxidized essential oil on serial dilution decreases against test fungai. Pure oxidized oil was found to be most effective against *Aspergillus oryzae,* and *Curvularia prasadii*. Similarly the pure oxidized essential oil exhibited significant activity against *A.terreus, Candida albicans and Trichoderma viridi.*the oxidized oil was quite active against *A.terreus* and *Trichoderma viridi* even at dilution 1:50. Comparative study of oxidized and unoxidised essential oils of C. swietenia (Table-3) indicats that oxidized essential oil had more antifungal activity against maximum test fungi.

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