



**ANTIMICROBIAL EFFECT AND INSILICO ADMET
PREDICTION OF *SANTALUM ALBUM* L.**

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ABSTRACT

Santalum album L. is known as East Indian Sandalwood Plant and is well known for its Heartwood. The Sandalwood heartwood contains two major photochemical i.e. *Santalol* and *Santyl acetate*. For this study different fractions of Heartwood and leaves are extracting in Soxhlet assembly with organic solvents as well as pure Heartwood oil was extracted by steam distillation method in Clavenger assembly. Fractions and Oil were screened against few human pathogens by disc diffusion method. Methanol fraction and Oil resulted in high potential against *Escherichia coli*, *Staphylococcus aureus* and *S.typhi*. In silico prediction of *Santalol* and *Santayl acetate* using PRE-ADMET, shows 100% Human intestinal absorption and plasma protein binding, Positive against carcinogenic cell lines, Positive CNS-Barrier test, which affirms them as potent antibacterial and anticancerus lead compound.

KEY WORDS: *Antimicrobial Effect, Heartwood, ADMET, Clavenger and Soxhlet.*



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INTRODUCTION

Plants have been used in traditional medicine for several thousand years. The knowledge of medicinal plants has been accumulated in the course of many centuries base on different medicinal systems¹. A report by World Health Organization (W.H.O) said approximately 25% of modern drugs used in United States are derived from plants². Up to the present time several traditional African communities still depend on various herbal preparations for their primary health care. However majority of these herbal products are still produced in a crude manner and the active ingredients in them have not been isolated and characterized. In the history of mankind infectious diseases are the leading cause of death. The clinical efficacy of many existing drug is been threatened by the emergence of multidrug-resistant pathogens. The increasing failure of chemotherapeutic agents and antibiotic resistance been encountered in medical practice today has led to the screening of several medicinal plants for their potential antimicrobial activity³. Several secondary metabolites of plant origin have been reported to exhibit various levels of antimicrobial activities. Numerous studies have identified compounds within plants that are effective antibiotics⁴.

The sandal tree, also known as *Chandana* in India, is botanically *Santalum album* L. belonging to family Santalaceae. The plant was mainly exploited for fragrant sandalwood oil obtained by steam distillation. A small evergreen glabrous tree with slender drooping branches the sapwood white and odorless. The plant, *S. album* is a native of the highlands of southern India mainly Coorg, Chennai and Mysore. It generally occurs at altitudes of 2000-3000 feet. The tree attains the height of 60-65 feet and is actually an obligate hemi parasite plant on various hosts- *Cassia siamea*, *Pongamia glabra* and *Lantana acuminata*⁵.

The sandalwood is one of the oldest perfumery and medicinal material, it has over 2000 year of uninterrupted history. The

importance of Sandalwood oil is also mentioned in ancient Epic '*Ramayana* and *Mahabharata*'. The Sandalwood plant is mainly used as coolant, and also sedative effect and astringent activity, making it useful as disinfectant in genitourinary and bronchial tracts, diuretic, expectorant and stimulant. The sweet powerful and lasting odor makes Sandalwood oil useful in perfume industry. The same is also used as tonic for heart, stomach liver, anti-poison, fever, memory improvement and as a blood purifier. Various uses mentioned in *Ayurveda* system about sandalwood are in treatment of various other ailments like diarrhea with bleeding intrinsic hemorrhage bleeding piles, vomiting, poisoning, hiccoughs initial phase of pox, urticaria, eye infections and inflammation of umbilicus^{6,7}. The Sandalwood oil contains two major Phytochemicals *i.e.* Santalol and Santyl acetate both of this responsible for antimicrobial activity. The success of drug's journey through the body is measured the dimensions of Absorption, distribution, metabolism and elimination (ADME). The ADME property of a drug together with its pharmacological properties are conventionally viewed as part of drug development, the process of making molecule as effective as possible as medicine. The toxicology-T in ADMET is the art of making sure that the molecule causes no harm, regardless of what good is Dose.

MATERIAL AND METHODS

Source of explants: For the present Investigation the explants material collected from the farm of Mr. K. T. Hire, Dist. Jalgaon (MS) India and Shree Shail Medi farms cultivators, Nagpur, India.

Preparation of plant extract: The Heartwood and leaves of plant was collected and converted in powdered form, 20 gm of each powdered material was used for each extraction using Soxhelt apparatus with 4

solvents namely diethyl ether, methanol, hexane and water taken separately and collected fractions were stored in dark amber colored bottles in refrigerator. The essential oil from the heart wood of *S. album* L. was collected by steam distillation method in 'Clavenger' assembly.

Test Microorganisms and inoculums preparation: For the present work the pathogenic microbes were procured from NCL Pune and Department of Microbiology, Ranibai Agnihotri institute of Computer Science and Information Technology, Wardha. The pathogens were characterized by their morphological, biochemical testing after this growth on selective medium. The preparation of bacterial inoculum, pure culture of test organisms was inoculated in 50 ml of nutrient broth and incubated at 37°C for 24 hr till moderate turbidity developed.

Sensitivity test: The antimicrobial assay of Sandalwood fractions and pure oil was performed by disc diffusion method on Muller Hinton agar medium. The dry filter paper discs (0.6mm) were soaked in liquid extracts and then the disc were dried in oven at 60°C. For antimicrobial activity medium was prepared and sterilized by autoclaving at 121°C and 15 lbs for 15 min. The 25 ml of medium was poured in 90 mm Petri dish with spreading 100 micro liters of bacterial culture and place filter paper disc on bacterial lawn. Then the plates were incubated at 37°C for 24 hrs. The disc containing antimicrobial agent showing clear zone of inhibition was considered to be inhibited and test organism shows zone and count with the Himedia scale.

INSILICO ADMET: With the help of ADMET tool of Bioinformatics ADMET Stands for Absorption, Distribution, Metabolism, Excretion and Toxicity. The prediction of the

ADME properties play an important role in the drug design process because these properties account for the failure of about 60% of all, drugs in clinical phases. For the ADME prediction open the <http://www.google.com/>⁸ search engine and simultaneously access the mole data file of Santalol and Santyl acetate download from chemical databases and retrieve the mole files in Pre – ADMET software <http://preadmet.bmdrc.org/>⁹ and results were calculated.

RESULTS

The volatile oil extracted from *S. album* derived from the heartwood is colorless to yellowish, viscous liquid with peculiar heavy sweet odor, the chief constituents of the oil is *Santalol* and *Santyl acetate*. The sandalwood had a mean oil concentration of 1.3–2.3%. In present study antimicrobial efficacy of *S. album* was carried with the four types of solvents used i.e. Diethyl ether, methanol, n-hexane and water these fractions were examined against human pathogenic microbes. In that Diethyl ether, methanol extract shows strong effect against *Escherichia coli*, *Staphylococcus aureus*, *Proteus vulgaris*, *Klebsiella pneumoniae*, *Pseudomonas syringae* and *Xanthomonas malceverum*.

In comparison of leaves and Heartwood fractions the heartwood fractions shows much more effectiveness against pathogens. The diethyl ether and methanol shows maximum activity than hexane and water. In all studied pathogens *Pseudomonas aeruginosa* and *Salmonella typhi* Shows resistance to Heartwood and Leaves fraction. In case of Antimicrobial study of heartwood essential oil shows zone of Inhibition up to 24 mm to 12 mm but *P. aeruginosa* showing resistance.

Table 1
Antimicrobial activity of Heartwood and Leaves Fractions

Microorganism	Heartwood fraction Zone				Leaves fraction zone			
	Die. ether	Meth	Hex	Wat	Die. ether	Meth	Hex	Wat
<i>E. coli</i>	14	14	--	--	12	16	--	--
<i>S. aureus</i>	17	12	--	--	12	11	--	--
<i>P. vulgaris</i>	12	11	--	--	10	8	--	--
<i>K. pneumoniae</i>	--	10	10	--	--	--	--	--
<i>P. aeruginosa</i>	--	--	10	--	--	--	--	--
<i>P. syringae</i>	11	8	--	--	--	--	--	--
<i>X. Malvacearum</i>	--	16	--	--	--	10	--	--
<i>S. typhi</i>	--	13	--	--	--	--	--	--
<i>B. cereus</i>	--	--	--	--	--	--	--	--
<i>S. fecalis</i>	--	--	--	--	--	--	--	--

Note- Die. Ether- Diethyl Ether, Hex- n-Hexane, Metha- Methanol, Wat- Water;
-- means not determined

Table 2
Antimicrobial activity of sandalwood essential oil

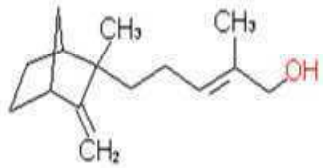
Microorganism	Zone of Inhibition	
	Oil	Standard
<i>E. coli</i>	22	16
<i>S. aureus</i>	17	14
<i>P. vulgaris</i>	14	15
<i>K. pneumoniae</i>	--	12
<i>P. aeruginosa</i>	--	--
<i>P. syringae</i>	12	12
<i>X. malvacearum</i>	24	24
<i>S. typhi</i>	16	14
<i>B. cereus</i>	15	14
<i>E. fecalis</i>	14	14

Note: -- means not determined

Table3
ADMET Properties and Toxicity of Santalol

ADMET

Result of calculation	
Name ^	Value
☒ : Absorption (4 Items)	
Human intestinal absorption (HIA, %)	100.000000
in vitro Caco-2 cell permeability (nm/sec)	22.4713
in vitro MDCK cell permeability (nm/sec)	198.723
in vitro skin permeability (logKp, cm/hour)	-1.00531
☒ : Ames test (7 Items)	
Ames TA100 (+S9)	negative
Ames TA100 (-S9)	negative
Ames TA1535 (+S9)	negative
Ames TA1535 (-S9)	negative
Ames TA98 (+S9)	negative
Ames TA98 (-S9)	positive
Ames test	mutagen
☒ : Carcinogenicity (2 Items)	
Carcinogenicity (Mouse)	positive
Carcinogenicity (Rat)	negative



SANTALOL

TOXICITY PREDICTION
SANTALOL

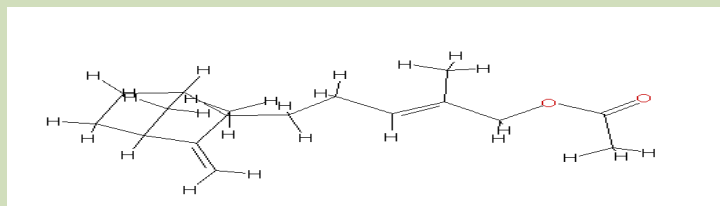
Table 4
CMC and multiple drug resistance Of Santalol

CMC AND MULTIPLE DRUG RESISTANCE
SANTALOL

Result of calculation	
Name ^	Value
☒ : Basic infomation (15 Items)	
CMC like Rule	Qualified
CMC like Rule Violation Fields	
CMC like Rule Violations	0
Lead like Rule	Violated
Lead like Rule Violations	1
Lead-like Rule Violation Fields	SKlogD_value
MDDR like Rule	Mid-structure
MDDR like Rule Violation Fields	No_Rings, No_Rotatable_bonds
MDDR like Rule Violations	2
Rule of Five	Suitable
☒ : Distribution (2 Items)	
in vitro plasma protein binding (%)	100.000
in vivo blood-brain barrier penetration (C.brain/C.blood)	8.90224

Table 5
ADME Properties and Toxicity of Santyl acetate

Santyl acetate



Result of calculation	
Name	Value
Absorption (4 Items)	
Human intestinal absorption (HIA, %)	100.000000
in vitro Caco-2 cell permeability (nm/sec)	38.6995
in vitro MDCK cell permeability (nm/sec)	3.60221
in vitro skin permeability (logKp, cm/hour)	-0.879587
Distribution (2 Items)	
in vitro plasma protein binding (%)	100.000000
in vivo blood-brain barrier penetration (C.brain/C.blood)	4.20921

Table 6
CMC, MDR and Toxicity Values of Santyl acetate

Result of calculation	
Name	Value
Basic information (15 Items)	
CMC like Rule	Qualified
CMC like Rule Violation Fields	
CMC like Rule Violations	0
Lead like Rule	Violated
Lead like Rule Violations	1
Lead-like Rule Violation Fields	SKlogD_value
MDDR like Rule	Mid-structure
MDDR like Rule Violation Fields	No_Rings, No_Rotatable_bonds
MDDR like Rule Violations	2
Rule of Five	Suitable
Ames test (7 Items)	
Ames TA100 (+S9)	negative
Ames TA100 (-S9)	negative
Ames TA1535 (+S9)	negative
Ames TA1535 (-S9)	negative
Ames TA98 (+S9)	negative
Ames TA98 (-S9)	positive
Ames test	mutagen
Carcinogenicity (2 Items)	
Carcinogenicity (Mouse)	positive
Carcinogenicity (Rat)	positive

**CMC AND
DRUG LIKENESS**

TOXICITY

In Insilco ADMET prediction of Phytochemicals from Sandalwood oil i.e. Santalol and Santyl acetate shows 100% human intestinal absorption, with the carcinogenicity parameter it cross the cell permeability of two cell lines namely CaCO₂ and MDCK with this molecules showing permeability with they have 100 % *In-vitro* plasma protein binding. So it indicated that the both of the molecule can be use as drug molecule.

DISSCUSION

Medicinal plants have been an important source of drugs for the treatment of diseases for thousands of years. Ayurveda is traditional system of medicine widely practice in India. Ayurveda was most popular before the advent of modern medicine. In antimicrobial study the four types of solvents used in that Diethyl ether and methanol showing strong antimicrobial effect than n-hexane and water. In this study it is found that pure heartwood oil shows more antimicrobial potential than leaves and other organic fractions. Inouye and Yamaguchi reported that the *H. influenzae* to be most susceptible to most essential oil but in present study the *P. aeruginosa* is more susceptible to the sandal wood oil¹⁰. Hammer and Riley reported that the essential oil from

lemongrass, oregano and bay inhibited all organisms at concentration of $\leq 2.0(v/v)$ ¹¹. Rama Prabha and Vasantha says the maximum antibacterial activity was showed against *S. aureus* and *Bacillus subtilis* while studying the different extracts of plant material¹². In present study the essential oil shows maximum activity against *E. coli*, *S. aureus*, *Proteus vulgaris*, *K. Pneumoniae*, *P. syringae*, *X. malvacearum*, *S. typhi*, *B. cereus* and *S. fecalis*.

CONCLUSION

In conclusion the body of literature identifying the antimicrobial potential of many traditional and medicinal plants was grows. In this study eight extracts from Heartwood and leaves were studied from *S. album* in that Diethyl ether and methanol fraction inhibited the growth of most of Bacteria and in case of oil shows very responsive results against most of the bacteria but only *P. aeruginosa* so it can be concluded the Natural products such as essential oil can be exploited new medicinal drug. The present investigation can be seen as potential source of useful drugs in various infectious diseases concluded that the Indian Sandalwood proves the therapeutic potential against various infectious organisms and it also predicted that Insilco ADMET prediction.

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