Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2019; 8(4): 380-382
Received: 13-05-2019
Accepted: 15-06-2019

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Chemical characterization of *Pterocarpus santalinus* wood using GC-MS

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Abstract
Pterocarpus santalinus heart wood had various medicinal properties and the deep red coloured heart wood is known to yield dye called santalin. *Pterocarpus santalinus* heart wood are used as a cooling agent, antipyretic, anti-inflammatory, anti-helminthic, tonic, hemorrhage, chronic dysentery and diaphoretic as well as induce vomiting, to improve eye sight, mental aberrations and ulcers. Totally 14 compounds were identified in ethanolic extract of *Pterocarpus santalinus* wood. The major compounds in terms of peak area *viz.*, Tricyclo[4.4.0.0(2,7)]dec-8-ene-3-methanol, à,à,6,8-tetramethyl-, stereoisomer (36.607%), followed by à-Bisabolol (6.468%), 2-Naphthalenemethanol, decahydro-à,à,4atrimethyl-8-methylene-, [2R-(2à,4aà,8aá)] (4.615%), 2-Naphthalenemethanol, 1,2,3,4,4a,5,6,7-octahydro-à,à,4a,8-tetramethyl-, (2R-cis)- (2.698%) and minor compound is 3-Hydroxy-1a,3,6,6-tetramethylhexahydro-2-oxacyclopropa[d]naphthalen-5-one (0.408%) with retention time 18.885, 14.558, 13.898, 13.382 and 21.271 minutes respectively. The above component in the wood is used to treat snake bites and scorpion stings. Its toxicity, wound healing potential also evaluated in animal studies, antioxidant and antimicrobial activity.

Keywords: Red sanders, GC-MS, Dye, *Pterocarpus santalinus*

Introduction
Pterocarpus santalinus commonly called as red sanders or Lal chandan, an endemic species belongs to family fabaceae. It is only found in south India in Kadapa and Chittoor on the Tamil Nadu and Andhra Pradesh border. *Pterocarpus santalinus* heart wood had various medicinal properties which are used as a cooling agent, antipyretic, anti inflammatory, anti-helminthic, tonic, hemorrhage, chronic dysentery (Anonymus, 1969) [2] and diaphoretic as well as induce vomiting, to improve eye sight (Chopra *et al.*, 1956) [4], mental aberrations and ulcers (Kirtikar, 1987) [7]. In combination with other drugs the wood is used to treat snake bites and scorpion stings (Warrier *et al.*, 1995) [17]. Its toxicity, wound healing potential also evaluated in animal studies (Biswas *et al*., 2004) [3], antioxidant and antimicrobial activity. Though the species has more number of active principles and increasing demand. Hence the present study carried out to know the active principle compounds present in ethanolic extract of. heartwood.

Materials and methods
Preparation of the sample
The *Pterocarpus santalinus* Linn. heartwood was collected from Forest College and Research Institute, Mettupalayam. Collected wood samples were chipped into 20mm using chipper and pulverized using wiely mill. Exactly 10 grams of ground powder was defatted with petroleum ether and successively extracted further with ethanol in a Soxhlet extractor for 6 hours (Mohammad, A. *et al*., 2016).

Gas Chromatography and Mass Spectrometer (GC-MS) analysis
The chemical composition of wood extract was analysed by using Perkin Elmer Clarus SQ8C, DB-5 MS capillary non-polar column (30m, ID: 0.25 mm and film thickness of 0.25 µm). Sample size of 1µl ethanolic extract was injected for analysis and Helium was used as a carrier gas at 1mL/minute. The oven temperature was programmed from 80 ºC to 285 ºC (80 ºC for 5 min, 4 ºC rate 260 ºC, and 2 ºC rate 285 ºC hold for 10 minutes). The MS was set to scan from 45-650Da. The Ms also had inbuilt pre-filter which reduces the neutral particles. The data system has two inbuilt libraries for searching and matching the spectrum *viz.*, NIST4 and WILEY9 containing more than million references (Priyanka *et al*., 2016) [13, 16].
The interpretation of mass spectrum of GC-MS was done by using the database of National Institute Standard and Technology (NIST4) and WILEY9 libraries. The relative percentage of extract constituent was expressed with peak area normalization (Vinothini *et al*., 2016) [13, 16].

Instrumentation

The chemical composition of defatted ethanolic wood extract was analysed using Clarus SQ 8C Gas Chromatography - Mass Spectrometer - Perkin Elmer. With non - polar column (DB-5 MS capillary with dimensions 0.25mm OD x 0.25µm ID x 30 meters length procured from Agilent Co., USA. Helium was used as the carrier gas with flow rate at 1 ml/min. The instrument was programmed as follows, Injector port temperature was 220 ºC, Interface temperature was 250 ºC, ion source kept at 220 ºC. The oven temperature programmed with 75 ºC for 2 mins, 150 ºC @ 10 ºC/min, up to 250 ºC @ 10 ºC/min. with split ratio 1:12 and the injected in splitless mode. The MS was set to scan from 50 to 600 Da. The ionization energy was -70eV. The MS was also having inbuilt pre-filter which reduced the neutral particles. The data system has inbuilt libraries for searching and matching the spectrum. NIST MS Search 2.2v contains more than five lakh references.

Identification of compounds

The interpretation of mass spectrum of GC – MS was done using the database of National Institute Standard and Technology (NIST14). The spectrum of the known component was compared with the spectrum of the known components stored in the inbuilt library (Priyanka *et al*., 2016) [13, 16].

Results

Table 1: Chemical constituents detected by GC-MS in ethanolic extract of *Pterocarpus santalinus* heart wood.

Sl. No.	Retention time (minutes)	Compound name	Peak Area (%)
1.	13.382	2-Naphthalenemethanol, 1,2,3,4,4a,5,6,7-octahydro-à,à,4a,8-tetramethyl-, (2R-cis)-	2.698
2.	13.898	2-Naphthalenemethanol, decahydro-à,à,4atrimethyl-8-methylene-, [2R-(2à,4aà,8aá)]-	4.615
3.	14.558	à-Bisabolol	6.468
4.	14.968	2,4-Dimethoxy-3-methylbenzaldehyde	0.415
5.	15.273	6-(p-Tolyl)-2-methyl-2-heptenol, trans-	0.471
6.	15.678	(S,Z)-2-Methyl-6-(p-tolyl)hept-2-en-1-ol	0.419
7.	16.374	(1R,4aR,7R,8aR)-7-(2-Hydroxypropan-2-yl)-1,4adimethyldecahydronaphthalen-1-ol	1.197
9.	17.194	(1R,4aR,7R,8aR)-7-(2-Hydroxypropan-2-yl)-1,4adimethyldecahydronaphthalen-1-ol	1.817
10.	18.885	Tricyclo[4.4.0.0(2,7)]dec-8-ene-3-methanol, à,à,6,8-tetramethyl-, stereoisomer	36.607
11.	19.300	(4aS,7R)-7-(2-Hydroxypropan-2-yl)-1,4a-dimethyl-4,4a,5,6,7,8-hexahydronaphthalen-2(3H)-one	0.606
12.	21.271	3-Hydroxy-1a,3,6,6-tetramethylhexahydro-2-oxacyclopropa[d]naphthalen-5-one	0.408
13.	22.031	6-Isopropenyl-4,8a-dimethyl-1,2,3,5,6,7,8,8aoctahydronaphthalene-2,3-diol	0.747
14.	22.411	Acetic acid, 3-hydroxy-6-isopropenyl-4,8adimethyl-1,2,3,5,6,7,8,8a-octahydronaphthalen-2-yl ester	0.842
15.	22.756	Oxirane, 2,2-dimethyl-3-[3,7-dimethyl-9-(phenylthio)-3,7-nonadienyl]	1.048

Totally 14 compounds were identified in ethanolic extract of *Pterocarpus santalinus* wood presented in Table 1 and the chromatograph depicted in Figure 1.

Among the total fourteen compounds, the major compounds in terms of peak area *viz.*, Tricyclo [4.4.0.0(2,7)]dec-8-ene-3-methanol, à,à,6,8-tetramethyl-, stereoisomer (36.607%), followed by à-Bisabolol (6.468%), 2-Naphthalenemethanol, decahydro-à,à,4atrimethyl-8-methylene-, [2R-(2à,4aà,8aá)] (4.615%), 2-Naphthalenemethanol, 1,2,3,4,4a,5,6,7-octahydro-à,à,4a,8-tetramethyl-, (2R-cis)- (2.698%) and minor compound is 3-Hydroxy-1a,3,6,6-tetramethylhexahydro-2-oxacyclopropa[d]naphthalen-5-one (0.408%) with retention time 18.885, 14.558, 13.898, 13.382 and 21.271 minutes respectively.

The compounds identified in ethanolic extract found to have more pharmaceutical importance to treat various ailments. à-Bisabolol is essential oil extracted from *Matricaria chamomilla* and *Populus balsamifera*. Tricyclo [4.4.0.0(2,7)] dec-8-ene-3-methanol, à,à,6,8-tetramethyl-, stereoisomer found in *Pterocarpus marsupium* (Maruthupandian and Mohan, 2014) anti-oxidnat and anti-inflammatory. 2-Naphthalenemethanol, decahydro-à,à,4atrimethyl-8-methylene-, [2R-(2à,4aà,8aá)] found in *Populus nigra* (Dongli *et al*., 2014) anti-inhibition against microbes.

Other group of compound found to have varied biological activities. The results are similar with literatures such as Luis *et al.,* (2016) evaluated in *Eucalyptus globulus* stump wood methanolic extract; Marcelo *et al.,* (2012) [9], Rui *et al.,* (2013) [14] & Pasakorn *et al.,* (2016) [11] in *E. globulus* bark; Saravanan *et al.,* (2014) [15] in *Ficus religiosa*; Domingues *et al.,* (2011) [5] reported by Inner and outer bark fractions of *E. grandis* and *Eucalyptus urograndis* (*E. grandis* x *E. urophylla*) and *Eucalyptus maidenii*; Patinha *et al.,* (2013) [12] in *E. grandis x globulus*. Ashraf *et al.,* (2015) [1] in *E. camaldulensis* leaves extracts of methanol, ethanol, chloroform and hexane.

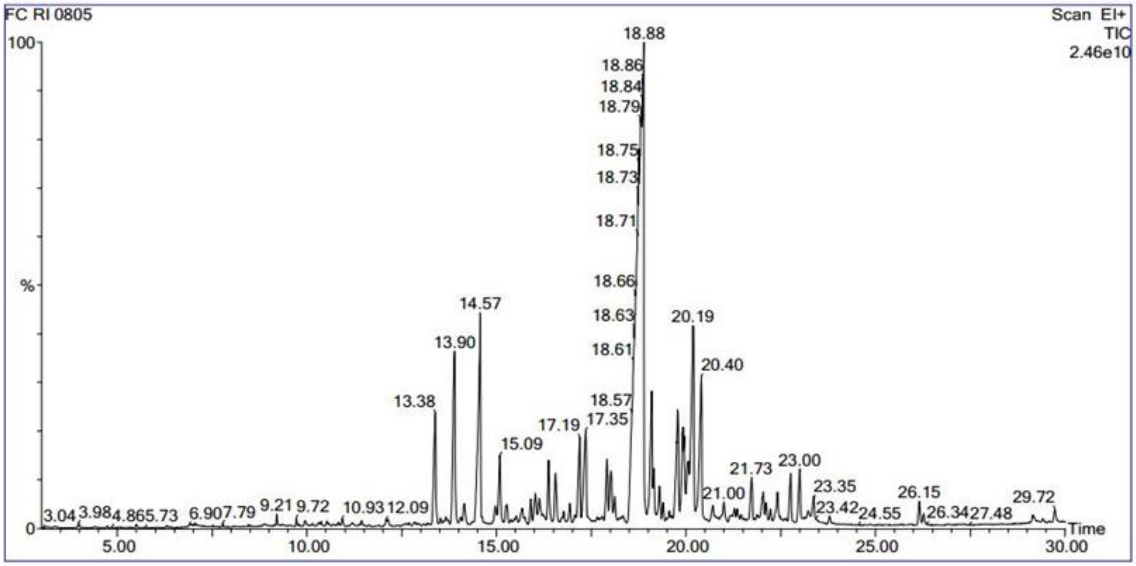

Fig 1: Chromatograph for *Pterocarpus santalinus* heartwood ethanolic extract

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