Original Article

Bulletin of Environment, Pharmacology & Life Sciences
Volume 1, Issue 1, December 2011: 20-23
Journal's URL: www.bepls.com
[Accepted 10 December 2011]

Chemical Compounds Investigation of *Cassia auriculata* Leaves – A Potential Folklore Medicinal Plant

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ABSTRACT
Medicinal plants, herbs, spices and herbal remedies are integral components of alternative system of medicine since times immemorial. Cassia auriculata Linn. is a potential folklore medicinal plant (Caesalpiniaceae) used for Aurveda nad Siddha systems of medicine. In this study, fatty acid esters, fatty acid amide, triterpene, diterpene alcohols and phytol were identified as the major chemical groups in the methanol fractions of C.auriculata leaf extracts. Their structures were elucidated, on the basis of GC-MS data, 3-O-Methyl-d-glucose (48.50%), α- Tocopherol-β-D-mannoside (14.22%), Resorcinol (11.80%), n-Hexadecanoic acid (3.21%) and 13-Octadecenal,(Z)- (2.18%).

INTRODUCTION

The use of plants with pharmaceutical properties has received increased interest nowadays from both homeopathic and allopathic branches. These medicinal plants play an important role in public health, especially in developing countries, where it is believed that the intense utilization of plants with therapeutic action does not lead to intoxication [1]. The cost of drugs in use today is too expensive for the majority of the population in the third world countries and therefore the search for some cheap sources of antimicrobial substances in nature become inevitable. Plants are good sources for new safe, biodegradable and renewable drugs. The use of plants as therapeutic agents in addition to being used as food is age long. Though the therapeutic uses of plants by the primitive people lack scientific explanations [2], there is a great awareness in the use and significance of these medicinal floras by the World Health Organization in several resource- poor nations [3]. This has led to intensified efforts on the documentation of medicinal plants [4].

Cassia auriculata Linn (Family: Caesalpiniaceae) commonly known as *Tanners Senna*, is distributed throughout hot deciduous forests of India and holds a very prestigious position in Ayurveda and Siddha systems of medicine. The plant has been reported to possess antipyretic [5], hepatoprotective [6], antidiabetic, antiperoxidative and antihyperglyceamic [7] and microbicidal activity [8]. The flowers are used to treat urinary discharges, nocturnal emissions, diabetes and throat irritation [9]. They are one of the constituent of polyherbal formulation 'Diasulin' in the concentration range of 40 mg/dl which is proven to have antidiabetic activity [10]. The dried flower bud powder is used as a substitute for tea in case of diabetic patients and it is also supposed to improve the complexion in women. The present investigation deals with extraction of essential oils, their GC analysis and purification from leaves of *Cassia auriculata* Linn

MATERIALS AND METHODS

PLANT MATERIAL

Lucas aspera was collected in Trichy District, Tamilnadu. The botanical identify of the plant was confirmed by Dr. V. Sampath kumar, Scientist C, Botanical Survey of India (Southern Circle), Coimbatore, Tamilnadu.

PLANT SAMPLE EXTRACTION

50gm powdered plant material is soaked in 200ml of Absolute alcohol overnight and then filtered through whatmann filter paper No.41 along with 2gm sodium sulfate to remove the sediments and traces of water in the filtrate. Before filtering, the filter paper along with sodium sulphate is wetted with absolute alcohol. The filtrate is then concentrated by bubbling nitrogen gas into the solution and reduce the volume to 1ml.The extract contains both polar and non-polar phytocomponents.

GC – MS ANALYSIS

GC-MS analysis was carried out on a GC Clarus 500 Perkin Elmer system comprising a AOC-20i auto sampler and gas chromatograph interfaced to a masss pectrometer (GC-MS) instrument employing the following conditions: Column Elite-1 fused silica capillary column (30mm×0.25mm I.D ×1 µ M df, composed of 100% Dimethyl poly siloxane), operating in electron impact mode at 70 eV; helium (99.999%) was used as carrier gas at a constant flow of 1ml/min and an injection volume of 0.5 µ l was employed (split ratio of10:1) injector temperature 250 °C; ion-source temperature 280 °C. The oven temperature was programmed from 110 °C (isothermal for 2 min), with an increase of 10°C/min, to 200°C, then 5°C/min to 280°C, ending with a 9 min isothermal at 280°C. Mass spectra were taken at 70 eV; a scan interval of 0.5seconds and fragments from 45 to 450 Da. Total GC running time is 46min.

RESULT AND DISCUSSION

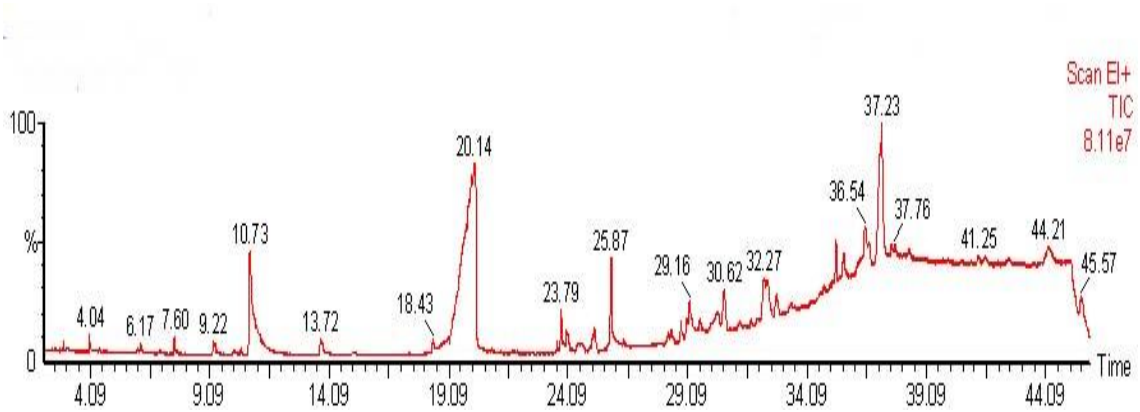
The composition and identification of the main components present in the leaves of *Cassia auriculata* are shown in (Table 1). Twenty-nine compounds were identified in *C. auriculata* by GC-MS analysis. The active principles with their retention time (RT), molecular formula, molecular weight (MW) and concentration (%) are presented in (Table 1 and Fig 1). It was found that the main constituents of leaves 3-O-Methyl-d-glucose (48.50%), α- Tocopherol-β-D-mannoside (14.22%), Resorcinol (11.80%), n-Hexadecanoic acid (3.21%), 13-Octadecenal, (Z)- (2.18%) and 1,2,3,4-Tetrahydroisoquinolin-6-ol-1-carboxylic acid (1.98%) (Fig.1).

Table 1. The Chemical Composition of leaves of *Cassisa auriculata* Linn

S.No	R.T	Name of the Compound	Molecular formula	Molecular wt	Peak area (%)
1	4.04	Glycerin	$C_3H_8O_3$	92	0.16
2	6.06	Thymine	$C_5H_6N_2O_2$	126	0.11
3	6.17	1-Butanol,3Methyl-, formate	$C_6H_{12}O_2$	116	0.17
4	7.60	4H-Pyran-4-one,2,3-dihydro-3,5-dihydroxy-6- methyl	$C_6H_8O_4$	144	0.46
5	9.22	Benzaldehyde,4 methyl-	C_8O_8O	120	0.83
6	10.38	2-Propenoic acid, 4-methylpentyl ester	$C_9H_{16}O_2$	156	0.12
7	10.73	Resorcinol	$C_6H_6O_2$	110	11.80
8	13.72	Sucrose	$C_{12}H_{22}O_{11}$	342	1.20
9	15.07	1,6,Anhydro-β-D-glucopyranose (levoglucosan)	$C_6H_{10}O_5$	162	0.30
10	18.43	β-D-Glucopyranoside, methyl	$C_7H_{14}O_6$	194	0.36
11	20.14	**3-O-Methyl-d-glucose**	**$C_7H_{14}O_6$**	**194**	**48.50**
12	23.79	1,2-Benzenedicarboxylic acid,bis (2-methylpropyl)ester	$C_{16}H_{22}O_4$	278	1.00
13	24.04	Benzenamine,2,3,4,5,6-pentamethyl	$C_{11}H_{17}N$	163	0.87
14	25.20	Unknown	***	-	0.57
15	25.87	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	3.21
16	26.42	Hexadecanoic acid, ethyl ester	$C_{18}H_{36}O_2$	284	0.10
17	28.29	1-Tridecyne	$C_{13}H_{24}$	180	0.30
18	28.41	13-Oxabicyclo[10.1.0] tridecane	$C_{12}H_{22}O$	182	0.42
19	28.80	Phytol	$C_{20}H_{40}O$	296	0.61

20	29.06	1-E,11,Z-13-Octadecatriene	$C_{18}H_{32}$	248	0.56
21	29.16	13-Octadecenal,(Z)-	$C_{18}H_{34}O$	266	2.18
22	29.58	1 Octadecanoic acid	$C_{18}H_{36}O_2$	284	0.46
23	30.62	1,2,3,4-Tetrahydroisoquinolin-6-ol-1-carboxylic acid	$C_{11}H_{36}NO_3$	207	1.98
24	32.26	Unknown	***	**	3.29
25	32.44	Unknown	***	**	2.61
26	32.82	α- Tocopherol	$C_{28}H_{48}O_2$	416	1.16
27	35.28	N-Acetltyramine	$C_{10}H_{13}NO_2$	179	1.24
28	35.60	Unknown	***	**	1.14
29	37.23	α- Tocopherol-β-D-mannoside	$C_{35}H_{60}O_7$	592	14.22

Fig.1. GC MS of leaves of *Cassisa auriculata* Linn



Our results showed the presence of aliphatic acid esters, terpene and diterpene alcohol in the leaves of *C.auriculata*. Diterpene alcohol was the major chemical group in of *C.auriculata* fractions. 3-0-Methyl-D-glucose (3-0MG), a nontoxic nonmetabo lizable derivative of glucose, is effective in reducing the toxicity of streptozotocin (SZ). It has been found to possess antitumor, oncogenic, and diabeto genic properties [11]. In the last decades, α-tocopherol has been consecrated as being one of the most efficient antioxidant and radical scavenger. This remarkable biochemical and physiological function is due, at least partially, to the shielding of its phenolic group by hydrophobic methyl groups and its lateral chain. The antioxidant and radical scavenger function of α-tocopherol is essentially dependent on the free state of its hydroxyl group. However, the use of α-tocopheryl glycosides makes sense and constant efforts have been made for their chemical and biochemical syntheses. This phenomenon is due to the large widespread of hydrolytic enzymes cleaving their glycosides, produced either by mammalian host or the comensal microorganisms populating their digestive tract. Moreover, spectacular antiallergic and antiinflamatory activities have been attributed to DL-α- tocopheryl-α-D-mannopyranoside and DL-α-tocopheryl-β-D-galactopyranoside [12].
Hexadecanoic acid methyl ester, also known as Methyl palmitate, in the methanol fraction is an aliphatic acid ester reported to cause growth inhibition and apoptosis induction in human gastric cancer cells [13] (Daniet *et al*., 2011). The n-hexadecanoic, methyl/ethyl ester of hexadecanoic acids are considered as fatty acids and these play important role in biological process [14]. Like other plants, *Litsea glutinosa* [15], *Suaeda maritime* [16], *Alpinia hainanesis* and *Alpinia katsumadai* [17], *Macrotyloma uniflorum* was also found to contain n – hexadecanoic acid*C.auriculata* is a potential folklore medicinal plant used for many diseases and infections. Phytochemical analysis by GC-MS revealed presence of fatty acid esters, fatty acid amide, terpenoids, diterpene alcohols and phytol as major compound groups in the methanol fractions. Compositional variation in quantities, qualities and structural features may influence compounds behavior on GC-MS, as well as bioactivities of their precursor fractions.

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