

Phytochemicals screening in the methanolic leaf extract of plant *Senna italica* by GCMS analysis.

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Abstract

The plant *Senna italica* looks like plant groundnut in its shape and appearance. It is distributed throughout Tamilnadu, mostly in the paddy field after harvest. This plant is a shrub like in appearance. In the rural areas, this plant is used to clear the constipation. Even one gram leaves are enough to induce the dysentery by increasing the bowel movement rapidly. The roots also enter in treatments of indigestion, liver complaints, gall bladder disorders, nausea, vomiting and dysmenorrhoea. Hence, in the present study, the phytochemicals present in the methanolic extract of leaves of plant *Senna italica* was analysed by GCMS. The GCMS results of these plant showed nine peaks which indicates the presence of nine compounds. They are (1)1-butanol, 2-methyl-, acetate (2)Phytol (3) 3-o-methyl-d-glucose (4) N-hexadecanoic acid (5) 9,12,15-octadecatrienoic acid, methyl ester, (z,z,z)- (6) 11,14,17-eicosatrienoic acid, methyl ester (7) Z-5,17-octadecadien-1-ol acetate (8) Dodecyl cis-9,10-epoxyoctadecanoate (9) 1-hentetracontanol.

Key Words : *Senna italica*, GCMS, Constipation, phytochemicals

Introduction

Since the period of time immemorial, human civilization has been used several plants as food, medicine, clothing and shelter. Medicinal plants have been the long-standing backbone of herbalism worldwide and are an important part of biomedical innovation. India has a unique position in the world where a number of recognized Traditional system of medicine *i.e.*, Ayurveda, Siddha, Unani, Homeopathy, Yoga and Naturopathy (Kumar, *et al.*, 2015). Medicinal plants have been recognized as potential drug candidates because they possess drug like properties (Bernhoft, *et al.* 2010). Several herbal medicines are being derived directly or indirectly from plants that are considered as an important medicine currently in use for curing various human diseases (ModK, *et al.* 2007 and Shakya, *et al.* 2011). The World Health Organization (WHO) reported that 4 billion people (80% of the world's population) use herbal medicines

for some aspect of primary healthcare (Fabricant, 2001). Herbal medicine has been recognized by WHO as essential components for primary health care and about 11% of the 252 drugs are derived from plants (Taylor Leslie, 2000). Many therapeutic drugs used in conventional medicine were rediscovered on the basis of traditional knowledge of medicinal herbs, and were developed through multidisciplinary scientific validation (Cragg, *et al*, 1994). Hence in the present study, the methanolic leaf extract of plant *Senna italica* was analysed by GCMS to find their bioactive phytochemicals.

Study plant Description

Senna italica species deciduous, perennial herbs, and shrubs up to 60 cm tall. The plant is woody in nature. Taproots are present in this plant. Stems are solid, usually less than 2 m tall. The stems are having lot of young twigs. Both stem and twigs are glabrous or sparsely glabrous and sparsely to densely hairy. This species has compound leaves with pinnate venation. Each leaf is containing approximately 4-6 leaflets, which are arranged spirally and alternately. The leaflets are elliptical, and shortly hairy on both sides. Each leaf left contain a small reddish gland between. The stipules are usually having lanceolate-triangular shape. They are approximately 3-9 mm long and early spreading -deflexed.

Flowering usually takes place during rainy season, but flowers can appear throughout the year in moist conditions. Flowers of *Senna italica* are actinomorphic and the inflorescence takes the form of axillary racemes, which are about 2-25 cm long. Petals of these flowers are usually yellow or orange in color, up to 13 mm long and are of obovate shape. Rhombic to ovate bracts are conspicuously present, but very small in size (up to 5mm long). The flower of Senegal *senna* is bisexual, zygomorphic and 5-merous. It usually has 10 stamens (lower 2 are the largest; 5 are medium-sized; 3 are short and sterile), Style up to 6 mm long and a superior ovary with short and stiff hair. Senegalsenna has freely dehiscent fruits. Fruits are oblong or ellipsoidal; they are strongly curved, falcate, bent or lunate shaped. The fruit may contain as many as 11 seeds. The seeds have elliptical line or depression and are wrinkled or rugose. Seeds are usually of olive, green or black color. *Senna italica* is usually propagated by seeds. Treating the seed by abrasion improves germination.

Plant distribution

The plant *Senna italica* is grown in grassland of the drier regions. It is found close to streams and in sandy and disturbed habitats, such as, waste places about towns and country dwellings, abandoned gardens, roadsides etc. This plant is adapted to warm temperature and may grow throughout the year annual grows typically in India and China. In India, it grows in all over South India such as Andhra Pradesh, Tamil Nadu, Kerala, and Karnataka. It is also found in Goa, Gujarat, Odisha and some other coastal regions. In Tamil Nadu, it is sown in February during spring rain as an irrigated crop. *Senna* grows in the sun well because it is a sun-loving plant. It should be harvested within 90 days from the time of sowing. Basically, it grows in Asia, Sri Lanka, West Africa, North Africa, Sudan, and Europe. It is also native to Asia, from the Middle East to Iran, Iraq and later, this plant has been introduced to Caribbean and Venezuela.

Medicinal properties

Senna italica plant contains many chemicals, and they are called sennosides. It is safe for a shorter period in case of pregnancy or breastfeeding. *Senna* can cause diarrhoea, dehydration and electrolyte loss. The excess use of it can also cause stomach upset, vomiting and blood thinning. It also has other side effects such as nausea, bloody stool, rectal bleeding, stomach cramp and discoloured urine. So, before the use of *Senna*, the patient must consult with any Ayurveda specialist.. The plant is also used for treatment of hemorrhoids, weight loss, and irritable bowel syndrome, cures leprosy, of bronchitis, cough, cold, and asthma. This is useful in the treatment of skin disorders and also helpful in the treatment of leucoderma, typhoid and cholera and it is proved useful in the treatment of gout and jaundice. Throughout its range the leaves, pods and mature seeds of *Senna italica* are used as a purgative. They are taken, usually as a decoction or maceration, to cure stomach complaints, fever, jaundice, venereal diseases and biliousness, as an abortifacient and against intestinal worms. The leaves, either fresh or dried and pulverized, are used as a dressing for skin problems such as burns and ulcers. A tea made from the flowers is used as a purgative and to induce labour. A maceration of the roots is taken to cure colic and influenza, and boiled roots are used as a wound dressing. A root infusion is used as eye drops for sore eyes. The roots also enter in treatments of indigestion, liver complaints, gall bladder disorders, nausea, vomiting and dysmenorrhoea. In Malawi a root infusion is given to infants to cure diarrhoea. The mature seeds have a purging activity, young seeds are eaten as a snack or as a vegetable in the Sahel region. The seeds are smoked in Mauritania. The leaves, traded as 'neutral henna' or 'blonde henna', are used as a hair conditioner to make the hair glossy. It may impart a yellowish colour rather than a reddish one

Taxonomical classification

Kingdom: *Plantae*

Division: *Angiosperma*

Class: *Dicotyledonae*

Order: *Tubiflorae*

Family: *Caesalpinaceae*

Genus: *Senna*

Species: *italica*

Senna italica names in various language:

Senna, Sanna makkah, sénemúkki, Arabian Senna, Senna, Indian Senna, TinnvellySenna, holoul, Kawaria, Chotatarado, Senegal senna, balibali.

Chemical Constituents

Lot of research works were done by various researchers in the root, stem, leaf, flower, fruit and seeds of the plant *senna italica* have been subjected to chemical investigations extensively (Franz, 1993). They suggested the presence of following phytochemicals.

Ester groups

1-butanol, 2-methyl-, acetate, Pentane, 1-propoxy, acetic acid, pentyl ester, propanoic acid, 3-hydroxy-, methyl ester, acetic acid, 2-ethylbutyl ester, 1-butanol, 4-(1-methylethoxy)-, 1-octanamine, n-methyl-n-nitroso-, isonitrosoacetylacetone, propanal, 2,3-dihydroxy-, (s)-, 2-furanone, 3,4-dihydroxytetrahydro, acetic acid, 2-ethylbutyl ester, 1-butanol, 3-methyl-, formate, 1-butanamine, 3-methyl-n-(3-methylbutyl)-n-nitroso-, -butanol, 2-methyl-, acetate, piperidinemethanol, 1-methyl-, pyrimidine-2,4,6(1h,3h,5h)-trione, 1-octadecyl-.

Phytol groups

Oxirane, tetradecyl-, octadecanal, oxirane, heptadecyl-, oxirane, hexadecyl-, exadecanaloxirane, hexadecyl-, oxirane, tetradecyl-, 15-tetramethyl-2-hexadecen-1-ol, oxirane, tridecyl-, tetradecanal, octadecanal, 16-heptadecenal, tetradecanal, n-nonadecanol-1 tetradecanal, oxirane, tetradecyl.

Methyl group

3-o-methyl-d-glucose, myo-inositol, 4-c-methyl-, d-manno-l-gluco-octonic acid, myo-inositol, 2-c-methyl-, scyllo-inositol, 1-c-methyl-, .alpha.-d-mannofuranoside, methyl, .beta.-d-mannofuranoside,

methyl,3-methylmannoside, .alpha.-methyl mannofuranoside,-o-methyl-d-mannopyranosa,d-epi-inositol, 4-c-methyl,-o-methylmannose, alpha.-d-6,3-furanose, methyl-.beta.-d-glucohexodialdo-1,4-furanosi,d-glycero-d-gulo-heptonic acid, .gamma.-lactone,methyl d-lyxofuranoside, alpha.-d-mannofuranoside, 1-o-decyl-, methyl(methyl 4-o-methyl-.alpha.-d-mannopyranoside)urate.

Long-Chain compounds:

(4-(24-Hydroxy-1-oxo-5-n-propyltetracosanyl)-phenol);28-ydroxypentatriacontan-7-one;7-Hydroxydotriacontan-2-one;1-Hydroxytetatriacontan-4-one;32-Methyltetatriacontan-8-ol; Nonatriacontane; 5-Acetoxytriacontane.

Flavonoid compounds:

Catechin; Acacetin; Apigenin 7-O-[60-O-(p-coumaroyl)-b -D-Glucoside; Chrysoeriol; Apigenin.

Sterols and Fatty compounds:

3-Sitosterol; 9,12,15-Octadecatrienoic acid methyl ester; n-Hexadecanoic acid; Linoleic acid; oleic acid; Stearic acid; Linolenic acid; Ceryl alcohol; Dotriacontanol.

Fig. 1. The plant *Senna italica* with leaves and flowers



Materials and method

The fresh and healthy leaves of plant *Senna italica* was collected from the waste land near Vallimalai Murugan Temple. The collected plants were identified and authenticated by local siddha practitioner and traditional heals in the Serkadu Village. The collected leaves were spread on the newspaper and sun dried at room temperature in shade place. After ensure the complete drying. The dried leaves were ground well using grinding machines. The known amount of powder (100gm) *Senna italica* powder was packed inside the thimble of soxhlet apparatus. Extraction was performed with adequate amount of methanol for 48 h at a temperature not exceeding the boiling point of the solvent. The extraction process was continued upto the disappearance of green colour of packed powder. The extract obtained was filtered through a 45 µm filter. The resulting solution was air dried by kept in open condition. Then semisolid extract was further concentrated in vacuum to dryness to give methanol extract. The solid extract was stored in a refrigerator at 4°C for further use.

GC – MS Analysis

The quantitative and qualitative analysis of phytochemicals in the plants *Senna italica* were analysed by Gas chromatography- Mass spectrometry (Turbo Mass).

Results and discussion

Plants are used for traditional medicine to treat chronic as well as infectious diseases (Alam *et al*, 2009). The common view in the society and the medical community is that plant based products are healthier, safer, and more reliable than synthetic products (Benli *et al*, 2008). The extract of *Senna italica* has served as good sources of antibiotics against various bacterial and fungal pathogens (Falodun, *et al*, 2006). Addition to this, *Senna italica* was used to treat constipation both adult and children. The present GC-MS analysis of *Senna italica* leaves revealed the existence of nine compounds (phytochemical constituents) that could contribute the medicinal quality of the plant. The identification of the phytochemical compounds was confirmed based on the peak area, retention time and molecular formula. They are (1) 1-butanol, 2-methyl-, acetate (2) Phytol (3) 3-O-methyl-D-glucose (4) N-hexadecanoic acid (5) 9,12,15-octadecatrienoic acid, methyl ester, (Z,Z,Z)- (6) 11,14,17-eicosatrienoic acid, methyl ester (7) Z-5,17-octadecadien-1-ol acetate (8) Dodecyl cis-9,10-epoxyoctadecanoate (9) 1-hentetracontanol.

The identified all these nine compounds possess many biological properties. The first compound is 1-Butanol, 3-methyl, Acetate, the other names of this compound are 2-Methyl-1-butyl acetate; 2-Methylbutyl acetate; Acetic acid 2-methylbutyl ester; 2-Methylbutyl acetate; 2-Methyl-1-butanol acetate; 2-Methylbutanol acetate; Active amyl acetate.

The -2-Methyl-1-butanol is found in fruits, isolated from grapes, apples, tomatoes etc, belongs to the family of Primary Alcohols. These are compounds comprising the primary alcohol functional group, with the general structure RCOH possesses anti-inflammatory, insectifuge, hypocholesterolemic, cancer preventive, nematocide, hepatoprotective, antihistaminic, antieczemic, antiacne, 5-alpha reductase inhibitor, antiandrogenic, antiarthritic and anticoronary properties. N-Hexadecanoic acid - palmitic acid (R/T 17.25) can be an antioxidant, hypocholesterolemic, nematocide, pesticide, lubricant activities and hemolytic 5-alpha is a reductase inhibitors. Phytol- Diterpene (R/T 19.67) is an antimicrobial, anticancer, anti-inflammatory and diuretic agent (Praveen kumar, *et al.*, 2010).










Phytol is one among the nine compounds of the present study. Maria Jancy Rani *et al.* (2011) observed the presence of phytol in the leaves of *Lantana camara* and Sridharan, *et al.* (2011) in *Mimosa pudica* leaves, in the leaves of *Lantana camara* (Sathishkumar and Manimegalai, 2008). Phytol has antibacterial activities against *Staphylococcus aureus* by causing damage to cell membranes as a result there is a leakage of potassium ions from bacterial cells (Inoue *et al.*, 2005). Phytol, Phenol, 2, 4-bis (1-phenylethyl) - which are all have medicinal properties. Phytol is a key acyclic diterpene alcohol that is a precursor for vitamins E and K1. It is used along with simple sugar or corn syrup as a hardener in candies. Mangunwidjaja, *et al.* (2006) reported that the phytol is the main components of 9, 12 octadecadienoic acid, Octadec- 9enoic acid and 9,12-actadecadienoic acid present in *Croton tiglium* seed. These compounds were found to have potential antioxidant and anticancer activities.

Hexadenoic acid has earlier been reported as a component in alcohol extract of the leaves of *Kigelia pinnata* (Grace *et al.*, 2002) and *Melissa officinalis* (Sharafzadeh *et al.*, 2011). (Parasuraman *et al.*, 2009) identified 17 compounds with n-Hexadecanoic acid and Octadecanoic acid as the major compounds in the leaves of *Cleistanthu scollinus*. GC-MS analysis of ethyl acetate extract of *Goniothalamus umbrosus* revealed the presence of n-Hexadecanoic acid (Siddig Ibrahim *et al.*, 2009). N-hexadecanoic acid, Hexadecanoic acid, Phytol, 9, 12 - Octadecadienoic acid, 9, 12, 15-Octadecatrienoic acid and Squalene were identified in the ethanol leaf extract of *Aloe vera* (Arunkumar and Muthuselvam, 2009) and *Vitex*

negundo (Praveen kumar *et al.*, 2010). Squalene is used in cosmetics as a natural moisturizer. (Devi *et al.* 2009) reported that *Euphorbia long an* leaves mainly contained n-hexadecanoic acid and 9, 12-Octadecadienoic acid. These reports are in accordance with the result of this study. 9, 12, 15-Octadecatrienoic acid, methyl ester, (Z,Z,Z)-, n-Hexadecanoic acid, 1,2-Benzenedicarboxylic acid and diisooctyl ester were present in *Caesalpinias appan* ethanol extract (Sarumathy *et al.*, 2011). Similar types of compounds were identified among the nine compounds of this present study. 11,14,17-eicosatrien-1-01acetate, antiarthritic, anticoronary, antiinflammatory, antimicrobial, anticancer, antiinflammatory, diuretic. antiarthritic, anticoronary and antiinflammatory.Z-5, 17-octadecadien-01 acetate, Octadecanoic acid, n-Non aldehyde and Nonanoic acid has found to possess antioxidant activity and 2- methyltetracosane is found to be a free radical Scavenger.



Table.1. Retention time and related bioactive phytochemicals in the methanolic leaf extract of *Senna italica*

S. NO	R/T	Name of the Compound	Molecular Formula	Area%	MW	Structure
1.	10.822	1-butanol, 2-methyl-, acetate	C ₇ H ₁₄ O ₂	13.09	130	
2.	16.374	Phytol	C ₂₀ H ₄₀ O	1.84	296	
3.	17.589	3-o-methyl-d-glucose	C ₇ H ₁₄ O ₆	61.12	194	
4.	18.260	N-hexadecanoic acid	C ₁₆ H ₃₂ O ₂	6.12	256	
5.	18.970	9,12,15-octadecatrienoic acid, methyl ester, (z,z,z)-	C ₁₉ H ₃₂ O ₂	2.48	292	
6.	19.030	11,14,17-eicosatrienoic acid, methyl ester	C ₂₁ H ₃₆ O ₂	4.33	320	
7.	19.900	Z-5,17-octadecadien-1-ol acetate	C ₂₀ H ₃₆ O ₂	4.19	308	
8.	24.707	Dodecyl cis-9,10-epoxyoctadecanoate	C ₃₀ H ₅₈ O ₃	1.63	466	
9.	26.268	1-hentetracontanol	C ₄₁ H ₈₄ O	5.14	592	

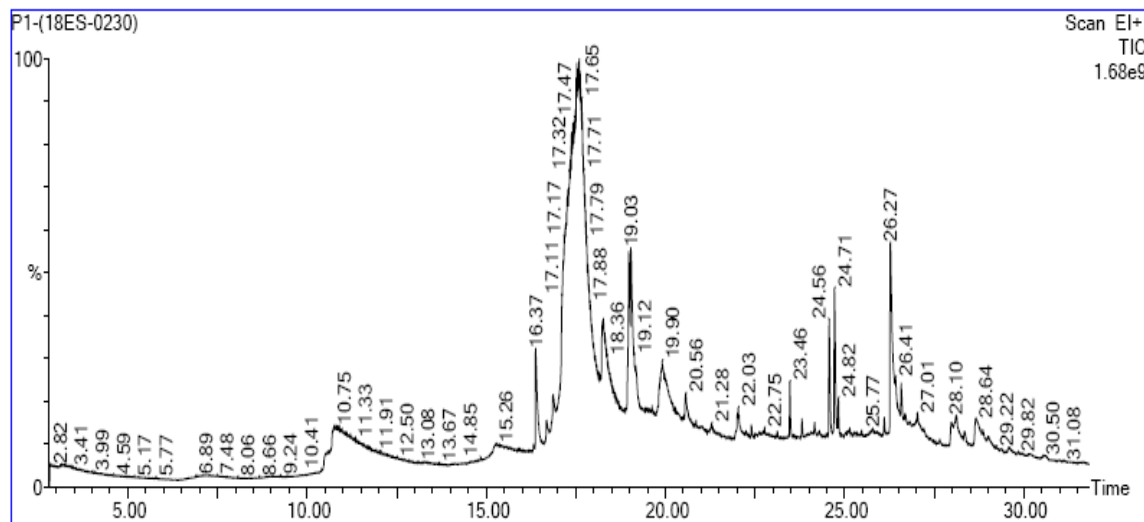
Qualitative Report

File: C:\TURBOMASS\2016.PRO\Data\P1-(18ES-0230).raw
 Acquired: 08-Mar-18 07:14:10 AM
 Description:
 GC/MS Method: GC: METHOD-1.mth MS: METHOD-1.EXP
 Sample ID: P1-(18ES-0230)

Printed: 20-Mar-18 10:23 AM

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Vial Number: 56



#	RT	Scan	Height	Area	Area %	Norm %
1	10.822	1604	172,726,416	170,112,864.0	13.094	21.42
2	16.374	2714	392,898,752	23,944,758.0	1.843	3.02
3	17.589	2957	1,383,892,736	794,032,704.0	61.120	100.00
4	18.260	3091	358,550,720	80,201,864.0	6.173	10.10
5	18.970	3233	619,380,672	32,200,138.0	2.479	4.06
6	19.030	3245	633,529,792	56,211,688.0	4.327	7.08
7	19.900	3419	213,922,752	54,394,840.0	4.187	6.85
8	24.707	4380	558,152,832	21,190,594.0	1.631	2.67
9	26.268	4692	744,320,384	66,843,104.0	5.145	8.42



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