Research Article

Determination of Biologically Active Compounds in Methanolic Extract of Flowers of *Senna alata* through GC-MS Analysis

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Abstract | *Senna alata* (L.) Roxb. (synn. *Cassia alata* L.) is a traditional medicinal legume native to Argentina that grows as a wild plant in Pakistan. The present study was conducted for GC-MS analysis of *C. alata* flowers to identify possible bioactive compounds. Twelve compounds were recorded in the methanolic extract with hexadecanoic acid, 15-methyl-, methyl ester as the most abundant ones with 29.78% peak area. Other compounds were linolenic acid methyl ester (15.84%), guaiacol (9.48%), 2-cyclopenten-1-one, 2-hydroxy-3-methyl- (8.56%), 2-cyclopenten-1-one, 3-methyl- (7.73%), butanal, 3-hydroxy- (6.19%), phenol, 4-ethyl- (4.83%), aziridine, 1-(2-methyl-1-propenyl)- (4.50%), 2-cyclopenten-1-one, 2,3-dimethyl- (3.52%), benzene, 1-ethenyl-4-methoxy- (3.29%), indole (3.64%), benzaldehyde, 2-methyl- (2.57%). A literature survey revealed that some compounds *viz.* linolenic acid, methyl ester; hexadecanoic acid, 15-methyl-, methyl ester; benzaldehyde, 2-methyl-; guaiacol; and 2-cyclopenten-1-one, 2-hydroxy-3-methyl-possess various biological properties such as antibacterial, antifungal, acaricidal, antioxidant, anticancer, nematicidal, antiacne, insectifuge, antiarthritic, antihistaminic, anti-inflammatory and antieczemic hepatoprotective.

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Introduction

Natural products are essential and are in enormous demand because they possess numerous biological activities including antioxidants, anticancer, antimicrobial and anti-inflammatory (Iñiguez-Luna *et al*., 2021; Kwiecień *et al*., 2023). In recent years, many studies have been carried out to explore biologically active compounds from plants growing in Pakistan. Plants namely *Monotheca buxifolia, Ageratum conyzoides, Chenopodium quinoa, Acacia nilotica* and *Sonchus oleraceous* contain potent antifungal compounds against devastating soil-borne plant pathogen, *Macrophomina phaseolina* (Banaras *et al*., 2020, 2021; Khan and Javaid, 2020; Javed *et al*., 2021; Rafiq *et al*., 2024). Likewise, extracts of *Chenopodium album* and *Datura metel* can control the growth of another highly damaging soil-borne fungus, *Sclerotium rolfsii* (Jabeen *et al*., 2022; Javaid *et al*., 2023). Other studies have shown that different Pakistani weed, ornamental and crop plants possess many medicinally and pharmaceutically important compounds (Javaid *et al*., 2021a; Khan and Javaid, 2022; Ferdosi *et al*., 2023).

Senna alata, a plant of the family Fabaceae, is native to Argentina and freely grows in the tropics. It is a plant of medicinal importance that is especially useful for treating skin diseases (Fatmawati *et al*., 2020). Its different parts are used in folk medicine for treatment of variousinfections and diseases (Oladeji *et al*., 2020). It possesses antifungal, antibacterial (Owoyale *et al*., 2005), Senna alata, anti-inflammatory (Pongnimitprasert *et al*., 2018), antitumor, antioxidant (Olarte *et al*., 2010, 2013) and antihelmintic (Kundu *et al*., 2012) activities. A range of bioactive compounds namely alatinone, alatonal (anthraquinones), glycosides, rhein, chrysaphanol, kaempferol (phenolics), sitosterol, campesterol, sitosterol, stigmasterol (terpenoides), and various steroids have been identified in this plant species (Fatmawati *et al*., 2020; Oladeji *et al*., 2020). However, information related to the phytochemical analysis of *S. alata* flowers is rare, hence, this study was carried out to explore phytoconstituents of methanolic flower extract of this plant growing in Lahore, Pakistan.

Materials and Methods

Methanolic extraction

S. alata flowers were collected from Lahore, Pakistan in November 2022. The flowers were washed gently with water to remove the dust and the moisture was removed under the fan. These flowers were completely dried in a hot oven set at 40 °C. That dried material was crushed in a pestle and mortar and 20 g of it was extracted in 200 mL of pure methanol for 14 days. The extract was filtered and its GC-MS analysis was performed to identify possible bioactive compounds from the extract (Ferdosi *et al*., 2021).

GC-MS analysis

Gas chromatograph (GC) model 7890B equipped with Mass spectrometer (MS) model 5977A, was used (Agilent Tech. USA). The flower extract was analyzed following the procedure described by Ferdosi *et al*. (2021) as per set conditions. The standard GC column DB-5, with dimensions of length 30 m, inner diameter 0.25 mm and film thickness 0.25 µm with fused silica GC tubing was used. The injection volume analyzed was 1 µL in split less mode with helium as carrier gas. Oven ramping was started at 80 °C and increased by 10 °C per min up to a maximum of 300 °C. Inlet temperature was 280 °C. A 50–500 m/z scan range with a solvent delay time of 5 minutes was employed. The source temperature was 230 °C. The NIST library was surveyed to identify the compounds found in this analysis.

Literature survey

An exhaustive literature hunting was done by using different data bases such as Science Direct, Google Scholar, Crossref, PubMed, SciELO, Scopus, JSTOR and Directory of Open Access Journals was also consulted to collect information regarding bioactivities of these identified compounds as reported by other scientists (Javaid *et al*., 2021b).

Results and Discussion

There were 12 compounds in the extract of *S. alata* flowers as shown in Table 1 and Figure 1. Hexadecanoic acid, 15-methyl-, methyl ester was the most abundant compound in the extract with 29.78% peak area. Earlier, this compound was reported in flowers of pink *Nerium oleander* (Seed *et al*., 2023), leaves of *Leucaena leucocephala* (Zayed and Samling, 2016), and methanolic extract of *Cycas pectinata* (Tareq *et al*., 2020). It has antioxidant, antiandrogenic, pesticidal and nematicidal properties (Zayed and Samling, 2016). The 2nd most abundant compounds was linolenic acid, methyl ester, (Z, Z, Z)- (15.84%). This compound has been reported in many plant species such as in leaves of *Tabernaemontana divaricata* (Javaid and Khan, 2022), and flowers of *Beaumontia grandiflora*

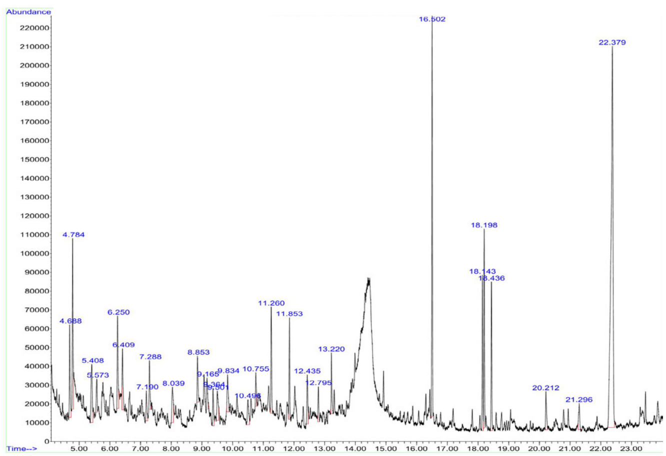

Figure 1: *GC-MS chromatogram of methanolic flower extract of Senna alata.*

Table 1: *List of compounds in methanolic flower extract of Senna alata identified by GC-MS analysis.*

S. No.	Names of compounds	Molecular formula	Molecular weight	Retention time (min)	Peak area (%)
1	2-Cyclopenten-1-one, 3-methyl-	C_6H_8O	96.12	4.688	7.73
2	2-Cyclopenten-1-one, 2-hydroxy-3-methyl-	$C_6H_8O_2$	112.12	5.408	8.56
3	2-Cyclopenten-1-one, 2,3-dimethyl-	$C_7H_{10}O$	110.15	5.573	3.52
4	Guaiacol	$C_7H_8O_2$	124.13	6.250	9.48
5	Butanal, 3-hydroxy-	$C_4H_8O_2$	88.10	6.409	6.19
6	Benzene, 1-ethenyl-4-methoxy-	$C_9H_{10}O$	134.17	7.190	3.29
7	Phenol, 4-ethyl-	$C_8H_{10}O$	122.16	7.288	4.83
8	Benzaldehyde, 2-methyl-	$C_8H_{10}O$	120.14	8.039	2.57
9	Indole	C_8H_7N	117.14	9.165	3.64
10	Aziridine, 1-(2-methyl-1-propenyl)-	$C_6H_{11}N$	97.15	9.501	4.50
11	Hexadecanoic acid, 15-methyl-, methyl ester	$C_{18}H_{36}O$	284.47	16.502	29.78
12	Linolenic acid, methyl ester	$C_{19}H_{32}O_2$	292.45	18.198	15.84

Table 2: *Biological activities of compounds present in flower extract of Senna alata.*

S. No.	Names of compounds	Biological activity	Reference
1	2-Cyclopenten-1-one, 3-methyl-	-	
2	2-Cyclopenten-1-one, 2-hydroxy-3-methyl-	Antifungal, antibacterial	Makinen *et al.* (1982), Millet-Clerc *et al.* (1989)
3	2-Cyclopenten-1-one, 2,3-dimethyl-	-	
4	Guaiacol	Antioxidant, antifungal	Gao *et al.* (2021)
5	Butanal, 3-hydroxy-	-	
6	Benzene, 1-ethenyl-4-methoxy-	-	
7	Phenol, 4-ethyl-	-	
8	Benzaldehyde, 2-methyl-	Acaricidal	Yang *et al.* (2014)
9	Indole	-	
10	Aziridine, 1-(2-methyl-1-propenyl)-	-	
11	Hexadecanoic acid, 15-methyl-, methyl ester	Antioxidant, pesticide, nematicide	Zayed and Samling (2016)
12	Linolenic acid, methyl ester	Anticancer, nematicide, antiacne, antiarthritic, insect repellent, antieczemic hepatoprotective, anti-inflammatory, antihistaminic	Zayed and Samling (2016)

(Ferdosi *et al.*, 2022). It is recognized as a remedy for numerous bioactive properties including anti-inflammatory, antiacne, anticancer, hepatoprotective, antiarthritic, insect repellent, nematicide, antihistaminic and antieczemic (Zayed and Samling, 2016). Guaiacol (9.48%) was found to be the third major compound. It is a phenolic compound identified in Guaiac resin for the first time. So far, it has been found in many plants (Burdock, 1995). It possesses antioxidant properties. In addition, it also exhibited antifungal activity by inhibiting the production and germination of conidia of *Fusarium graminearum* (Gao *et al.*, 2021). It is also used in industrial applications like eugenol and vanillin production (Hiscox and Boddy, 2017). In addition, it is widely used as an anesthetic and antiseptic agent, to treat indigestion problem, and for the preparation of guaiacol benzoate and oral solutions for expectoration (Araujo *et al.*, 2018). Other significant compounds were 2-cyclopenten-1-one, 2-hydroxy-3-methyl (8.56%) and 2-cyclopenten-1-one, 3-methyl- (7.73%). The former compound has been described earlier in tea, cocca and coffee (Gianturco and Friedel, 1963; Aesbacher *et al.*, 1989). It showed antifungal property against *Aspergillus fumigatus* and different dermophytes (Millet-Clerc *et al.*, 1989), in addition to antibacterial activity against different species of *Salmonella* (Makinen *et al.*, 1982) (Table 2).

Less abundant compounds included butanal, 3-hydroxy- (6.19%), phenol, 4-ethyl- (4.83%), aziridine, 1-(2-methyl-1-propenyl)- (4.50%), 2-cyclopenten-1-one, 2,3-dimethyl- (3.52%), benzene, 1-ethenyl-4-methoxy- (3.29%), indole (3.64%), benzaldehyde, 2-methyl- (2.57%). Phenol, 4-ethyl-, a volatile phenolic compound, is produced by non-spore forming yeast, *Brettanomyces* spp. and gives aroma to wine (Cheynier *et al*., 2010). Indole has been reported in *Cannabis sativa* and is responsible for its aroma as reported by Oswald *et al*. (2023). Indole nucleus has a prominent position in many compounds of biological importance (Kaushik *et al*., 2013). The indole derivatives are very active biological molecules exhibiting anticancer, antiviral, antioxidant, anti-inflammatory, anti-HIV, antidiabetic, antimicrobial and antimalarial activities (Kumar and Ritika, 2020). Benzaldehyde, 2-methyl- showed acaricidal activity against *Dermatophagoides pteronyssinus, D. farina* and *Haemaphysalis longicornis*. Its acaricidal activity was up to 40 times higher than the commercial acaricide DEEP against these microorganisms (Yang *et al*., 2014).

Conclusions and Recommendations

Flowers of *S. alata* contain several bioactive compounds such as linolenic acid, methyl ester; hexadecanoic acid, 15-methyl-, methyl ester; benzaldehyde, 2-methyl-; guaiacol; and 2-cyclopenten-1-one, 2-hydroxy-3-methyl. These compounds possess antioxidant, antibacterial, acaricidal, antifungal, anticancer, nematicidal, antiacne, antiarthritic, insectifuge, anti-inflammatory, antihistaminic and antieczemic hepatoprotective properties.

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Novelty Statement

Senna alata is a weed plant possessing many biological activities. Various studies regarding its biological activities and chemical composition have been carried out in different countries. However, studies about chemical constituents of flowers of this plant species growing in Lahore, Pakistan are lacking. Since chemical composition of plants changes with the change in environmental factors, therefore, the present study was undertaken to explore chemical compounds of flowers of this plant growing in Lahore, Pakistan.

Author's Contribution

MFHF supervised the whole work, arranged experimental materials and did collection of plant samples. AJ did literature survey and wrote the manuscript. MS did experimental work. ZM did editing and TH supervised the GC-MS related part of the study.

Conflict of interest

The authors have declared no conflict of interest.

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