INTRODUCTION

Each and every plant contains a certain specific type of chemical substances/compounds, which are produced during the normal growth and development of the plant body. These compounds are generally referred to as “phytochemicals” or sometimes also known as “secondary metabolites.” These phytochemicals attribute to the medicinal value of the plant, and that's why, plants are being used to cure various serious diseases such as cancer and Alzheimer’s disease [1]. Medicinal plants are extensively being utilized to screen the phytochemicals in medicinal plant species. Using GC-MS, it is now possible to identify volatile compounds with ease.

Senna angustifolia Vahl. commonly known as “senna” is one of the most important medicinal shrubs belonging to family Fabaceae (Sub family: Caesalpinioideae). It is a native of Saudi Arabia and has been naturalized in India, Pakistan, and Egypt. The plant is a small shrub with usually 5-8 leaflets, lanceolate, glabrous, and axillary erect [2]. Senna is widely used in the Ayurvedic and Unani system of medicine. Leaves and pods are used as a febrifuge in splenic enlargements, curing various diseases such as anemia, typhoid, jaundice, and cholera as well as reported to be an excellent blood purifier. It has also been employed in the treatment of constipation, amoebic dysentery, as an anthelmintic and a mild liver stimulant. Plant contains rhein, chrysophanol, emodin, and aloe-emodin—a water-soluble glycoside, besides these, derivatives of anthraquinone glycosides are also present in the leaves and pods which are commonly referred as “sennosides” [2]. Considering the importance of C. angustifolia, this study was undertaken for the first time to analyze the bioactive compounds present in the methanolic extract of leaves of this plant.

METHODS

Collection of plant samples and extraction procedure
Leaves of C. angustifolia were collected from Botanical Garden of the Department of Botany, AMU, Aligarh. Only mature and healthy leaves were harvested and collected in the early morning, packed in polythene bags, and kept at room temperature for further processing. Leaves were washed with tap water and shade dried for 5-6 days. About 5 g of air dried leaves were ground to fine powder in a mortar pestle. About 5 g of powdered leaves of senna were extracted with 100 ml methanol. It was left for 24 hrs so that terpenoids and other constituents if present will get dissolved. The methanolic extract was then filtered through syringe filter, and the residue was removed.

GC-MS analysis and identification of components
The GC-MS analysis of the methanolic extract of the leaves of C. angustifolia was performed using a GC-MS of Hewlettt-Packard 6890/5973 operating at 1000 eV ionization energy, equipped with Agilent 7890A/5975 C GC HP-5. Capillary column (phenyl methyl siloxane, 25 m × 0.25 mm id.) with Helium (He) was used as the carrier gas with split ratio 1:5. Oven temperature was 80°C (2 minutes) to 280°C at 1-40°C/minutes, detector temperature 250-280°C, and carrier gas He (0.9 ml/minutes). 2.0 µl of respective diluted samples was manually injected in the splitless mode, with split ratio and with mass scan of 50-600 amu. Total running time of GC-MS is 40 minutes, the relative percentage of each extract constituent was expressed as a percentage with peak area normalization.

Interpretation on mass spectrum of GC-MS was done using the database of National Institute of Standards and Technology (NIST) and Wiley library for mass spectra, having more than 62000 patterns. The mass spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular weight (MW), and structure of the components of the test materials were ascertained.
RESULTS
In the present study, a total of 45 different phytocomponents have been found in the methanolic extract of the leaves of *C. angustifolia*. The identified compounds of senna, their retention indices (RT), molecular formulae, molecular structure, MW, and percentage composition (area %) are given in Table 1. The results showed the presence of five major components with maximum percentage (12.3%) of squalene, followed by 6,6-dideutero-nonen-1-ol-3 (10.45%), pentadecanoic acid (9.22%), 1-butanol, 3 methyl-, acetate (7.47%), and phytol (6.49%). The leaf extract also showed the presence of tocopherol (Vitamin E, 3.85% with RT of 30.4 minutes) and gamma-tocopherol (0.58% with RT of 28.6). The GC-MS chromatogram also shows 45 different peaks which confirm the presence of 45 compounds with their respective RT (Fig. 1). On comparison of the mass spectra of each phytochemical with the NIST and Wiley library, 45 phytoconstituents were characterized and identified. The individual fragmentation of five major components is illustrated in Fig 2 (A-E). Some of the identified components possess biological activities which are listed in Table 2. These biological activities are based on Dr. Duke’s Phytochemical and Ethnobotanical Databases by Dr. Jim Duke of the Agricultural Research Service USDA [3].

DISCUSSION
GC-MS chromatogram of the methanolic extract of the leaves of *C. angustifolia* showed the presence of 45 different phytocomponents. From observations and comparison with the mass spectrum of each constituent with NIST and Wiley library, it was found that squalene, 6,6-Dideutero-Nonen-1-Ol-3, pentadecanoic acid, 1 butanol, 3 methyl acetate, and phytol are the major components (Fig. 2). Besides these compounds, vitamin E (3.85 %), gamma-tocopherol (0.58%), and several other antioxidants are also present.

Squalene, which constitutes about 12.3%, is a hydrocarbon which is used in cosmetics, as an immunologic adjuvant in vaccines and may be used as a chemopreventive substance that protects people from cancer [4,5]. However, pentadecanoic acid, which is 9.22% in the tested extract, is a saturated fatty acid and is rare in nature but found at the level of 1.2% in the milk of cows [6]. Phytol is an acyclic diterpene alcohol that can be used as a precursor for the manufacture of synthetic forms of vitamin...
| Name of the compounds                        | RT  | Molecular formula | Molecular structure | MW  | Area % |
|---------------------------------------------|-----|-------------------|---------------------|-----|--------|
| 1-butanol, 3-methyl-, acetate               | 6.551 | C₇H₁₄O₂          |                     | 130 | 7.47   |
| 5-oxo-pyrroldine-2-carboxylic acid methyl   | 10.456 | C₆H₉NO₃         |                     | 143 | 0.84   |
| 6,6-dideutero-Nonen-1-ol-3                  | 11.808 | C₉H₁₆D₂O        |                     | 144 | 10.45  |
| Phenol, 2,4-bis (1,1-dimethylethyl)-        | 12.183 | C₁₁H₂₂O          |                     | 206 | 0.57   |
| Benzoic acid, 4-ethoxy-, ethyl ester       | 12.348 | C₁₁H₁₄O₃         |                     | 194 | 0.99   |
| Phenol, (1,1-dimethylethyl)-4-methoxy-      | 12.674 | C₁₁H₁₆O₂         |                     | 180 | 0.31   |
| benzene, 1,2,4-trimethoxy-5-(1-propenyl)-, (Z)- | 13.455 | C₁₂H₁₆O₃         |                     | 208 | 0.25   |
| Mome inositol                               | 14.059 | C₇H₁₄O₆          |                     | 194 | 3.93   |
| Mome Inositol                               | 14.574 | C₇H₁₄O₆          |                     | 194 | 2.77   |
| Name of the compounds                                      | RT  | Molecular formula | Molecular structure | MW  | Area % |
|-----------------------------------------------------------|-----|------------------|---------------------|-----|--------|
| Tetradecanoic acid                                        | 15.023 | C\textsubscript{14}H\textsubscript{28}O\textsubscript{2} | ![Molecular structure](image) | 228 | 0.18   |
| 3-Heptadecanol                                            | 15.453 | C\textsubscript{17}H\textsubscript{36}O\textsubscript{2} | ![Molecular structure](image) | 256 | 0.36   |
| Isopropyl tetradecanoate                                  | 15.654 | C\textsubscript{17}H\textsubscript{34}O\textsubscript{2} | ![Molecular structure](image) | 270 | 0.38   |
| (2e)-3,7,11,15-tetramethyl-2-hexadecene                   | 15.761 | C\textsubscript{20}H\textsubscript{40} | ![Molecular structure](image) | 280 | 0.15   |
| 2,6,10-trimethyl, 14-ethylene-14-pentadecene              | 15.819 | C\textsubscript{17}H\textsubscript{33} | ![Molecular structure](image) | 278 | 4.24   |
| (2e)-3,7,11,15-tetramethyl-2-hexadecene                   | 15.885 | C\textsubscript{20}H\textsubscript{40} | ![Molecular structure](image) | 280 | 0.36   |
| 3,7,11,15-Tetramethyl-2-Hexadecen-1-Ol                    | 16.077 | C\textsubscript{20}H\textsubscript{40}O\textsubscript{2} | ![Molecular structure](image) | 296 | 0.99   |
| 2-Hexadecen-1-ol, 3,7,11,15-tetramethyl-[R-[R*,R*-[E]]]   | 16.271 | C\textsubscript{20}H\textsubscript{40}O\textsubscript{2} | ![Molecular structure](image) | 296 | 1.42   |
| Z-9-Tetradecen-1-Ol formate                               | 16.660 | C\textsubscript{20}H\textsubscript{40}O\textsubscript{2} | ![Molecular structure](image) | 240 | 0.13   |
| Hexadecanoic acid, methyl ester                           | 16.707 | C\textsubscript{17}H\textsubscript{34}O\textsubscript{2} | ![Molecular structure](image) | 270 | 2.46   |
| Benzene, 1,1′-sulfonylbis-                                 | 17.014 | C\textsubscript{16}H\textsubscript{32}O\textsubscript{5} | ![Molecular structure](image) | 218 | 0.61   |
| Pentadecanoic acid                                        | 17.136 | C\textsubscript{15}H\textsubscript{30}O\textsubscript{2} | ![Molecular structure](image) | 242 | 9.22   |
| 9,12-Octadecadienoic acid (Z, Z)-, Methyl Ester           | 18.387 | C\textsubscript{18}H\textsubscript{32}O\textsubscript{2} | ![Molecular structure](image) | 294 | 1.42   |
| 9,12,15-Octadecatrienoic acid, methyl ester, (Z, Z, Z)-   | 18.458 | C\textsubscript{18}H\textsubscript{32}O\textsubscript{2} | ![Molecular structure](image) | 292 | 5.32   |
| Phytol                                                    | 18.573 | C\textsubscript{20}H\textsubscript{40}O\textsubscript{2} | ![Molecular structure](image) | 296 | 6.49   |
| Methyl stearate                                           | 18.650 | C\textsubscript{18}H\textsubscript{34}O\textsubscript{2} | ![Molecular structure](image) | 298 | 0.63   |
| 9,12-Octadecadienoic Acid (Z, Z)-                         | 18.797 | C\textsubscript{18}H\textsubscript{34}O\textsubscript{2} | ![Molecular structure](image) | 280 | 1.17   |
| 9,12,15-octadecatrienoic acid, (Z, Z, Z)-                 | 18.878 | C\textsubscript{18}H\textsubscript{34}O\textsubscript{2} | ![Molecular structure](image) | 278 | 6.32   |
| Octadecanoic acid                                         | 19.024 | C\textsubscript{18}H\textsubscript{36}O\textsubscript{2} | ![Molecular structure](image) | 284 | 0.79   |
| 9,12-Octadecadienoic acid (Z, Z)-, methyl ester           | 19.226 | C\textsubscript{19}H\textsubscript{38}O\textsubscript{2} | ![Molecular structure](image) | 294 | 0.20   |
| 2-Ethylhexyl (2e)-3-(4-methoxyphenyl)-2-propenoate        | 20.498 | C\textsubscript{18}H\textsubscript{38}O\textsubscript{3} | ![Molecular structure](image) | 290 | 0.17   |
| 3-Cyclopentylpropionic acid, 2-dimethylaminoethyl ester   | 21.599 | C\textsubscript{18}H\textsubscript{32}NO\textsubscript{3} | ![Molecular structure](image) | 213 | 0.27   |
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Table 1: (Continued)

| Name of the compounds                                           | RT    | Molecular formula | Molecular structure | MW   | Area % |
|-----------------------------------------------------------------|-------|-------------------|---------------------|------|--------|
| Oxalic acid, monoamide, N-allyl-, tetradecyl ester              | 21.690| C_{19}H_{35}NO_{3} |                     | 325  | 0.24   |
| Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl) ethyl ester     | 22.006| C_{19}H_{34}O_{4}  |                     | 330  | 1.49   |
| 1,2-Benzenedicarboxylic acid                                    | 22.310| C_{19}H_{34}O_{4}  |                     | 390  | 0.48   |
| Pentacosane                                                     | 23.534| C_{25}H_{52}       |                     | 352  | 1.24   |
| 9-octadecenamide                                               | 24.545| C_{18}H_{35}NO     |                     | 281  | 1.41   |
| Squalene                                                       | 25.056| C_{30}H_{50}       |                     | 410  | 12.30  |
| Tetracontane                                                   | 25.839| C_{26}H_{50}       |                     | 562  | 0.41   |
| Nerylinalool isomer                                             | 26.499| C_{20}H_{34}O      |                     | 290  | 0.45   |
| Gamma-tocopherol                                                | 28.698| C_{30}H_{50}O_{2}  |                     | 416  | 0.58   |
| 1-Heptacosanol                                                 | 29.375| C_{27}H_{56}O      |                     | 396  | 1.59   |
| Vitamin E                                                       | 30.404| C_{29}H_{50}O_{2}  |                     | 430  | 3.85   |
| Ergost-5-En-3-Ol (3.Beta.,24r)-                                | 32.958| C_{29}H_{50}O      |                     | 400  | 0.69   |
| Stigmasta-5,22-Dien-3-Ol                                        | 33.820| C_{29}H_{50}O      |                     | 412  | 1.67   |
| Stigmast-5-En-3-Ol (3.Beta.)-                                  | 35.579| C_{29}H_{50}O      |                     | 414  | 2.74   |

RT: Retention time; MW: Molecular weight, *C. angustifolia: Cassia angustifolia*

E [7] and vitamin K [8]. It is used in shampoos, cosmetics, toilet soaps, fragrance industry, household cleanser, and detergents [9]. Some of the antioxidants found to be present in senna leaf extract are tetradecanoic acid (synonym: Myristic acid), hexadecanoic acid, heptadecanoic acid, squalene (synonym: 2,6,10,14,18,22-tetracosahexaene), vitamin E (synonym: D-alpha-tocopherol), gamma-tocopherol (synonym: 7,8-dimethyltocol), campesterol (synonym: Ergost-5-en-3.beta.-ol), and beta-stigmasterol (synonym: Stigmast-5-En-3-Ol, [3.Beta.]). In our study, the leaf extract of senna contains 2.74% beta-sitosterol. However, Rastogi and Mehoratra [10] reported that the plant contains only 0.33% beta-sitosterol. Some of the components such as hexadecanoic acid methyl ester, phytol, 9,12-octadecanoic acid (Z,Z)-, 9,12,15-octadecatrienoic acid (Z,Z,Z)-, octadecanoic acid, and squalene have also been reported in the methanolic leaf extract of *Cassia alata* [11]. The methanolic leaf extract of *Lannea kerstingii* and *Nauclea diderrichii* also reported to contain hexadecanoic acid methyl ester and pentacosane [12].
CONCLUSION

Screening of leaf extract of *C. angustifolia* by GC-MS revealed the presence of 45 different phytochemicals. The study was undertaken for the first time, till date no information was available on GC-MS analysis of senna. The analysis showed the presence of highly valuable compound which could be used by various pharmaceutical or drug companies for the development of new medicines.

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| Name of compounds | Biological properties* |
|-------------------|------------------------|
| 2,6,10,14,18,22-tetraicosahexaene, 2,6,10,15,19,23-hexamethyl-squalene | Antibacterial, antioxidant, cancer-preventive, antitumor, immunostimulant, perfumery, pesticide, sunscreen |
| Pentadecanoic acid/myristic acid | Antioxidant, lubricant, hypercholesterolemic, cancer-preventive, cosmetic |
| 3,7,11,15-tetramethyl-2-hexadecen-1-ol 2-pentadecanone, 6,10,14-trimethyl/phytol | Cancer preventive |
| Gamma-tocopherol | Antihepatotoxic, antiviral, antioxidant, cancerpreventive, hypercholesterolemic |
| Vitamin E | Antioxidant, hypocholesterolemic |
| Ergost-5-en-3.beta.-ol/campesterol | Antioxidant, hypocholesterolemic |
| Stigmasta-5,22-dien-3.beta.-ol (3.beta.,22E)-/stigmasterol | Antioxidant, hypocholesterolemic |

*Source: Dr. Duke’s phytochemical and ethnobotanical database www.ars.gov/cgi-bin/duke/[3]. *C. angustifolia: Cassia angustifolia. GC-MS: Gas chromatography-mass spectrometry