IJPSR (2021), Volume 12, Issue 4 (Research Article)

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 01 April 2020; received in revised form, 08 September 2020; accepted, 15 September 2020; published 01 April 2021

ANALYSIS OF BIOLOGICALLY ACTIVE COMPOUNDS IN JIMSON WEED LEAFS

A. Ananth [*1], S. Rajan [1] and A. Uma Devi [2]

Department of Microbiology [1], Srinivasan College of Arts & Science, Perambalur - 621212, Tamil Nadu, India.
Department of Microbiology [2], M. R. Government Arts College, Mannargudi - 614001, Tamil Nadu, India.

Keywords:

Jimson weed, GC-MS, Steroids, Campesterol

Correspondence to Author:
Dr. A. Ananth

Assistant Professor,
Department of Microbiology,
Srinivasan College of Arts & Science,
Perambalur - 621212, Tamil Nadu, India.

E-mail: uananth4@gmail.com

ABSTRACT: Jimson weed (*Datura stramonium*) members of the Solanaceae plant family; serve as major folklore medicinal herbs. The biologically active compounds spectrum in leaves of *Datura stramonium* L. was investigated by GC-MS with NMR & FTIR analysis. Steroids, terpenoids, anticholinergic alkaloids, atropine, and scopolamine, are detected. Twenty of them are new constitutions for the species, and eight phytosterol compounds are described for the first time. This is the first report on some other antibacterial heterocyclic compounds, antioxidant flavonoids, antimicrobial phytol, amino acids, and sulfone & fluoro compounds in *Datura stramonium* leave identified by GC-MS analysis.

INTRODUCTION: *Datura stramonium* L. is a plant species distributed throughout the temperate region of the world [1]. This herbaceous annual is an erect plant, is used traditionally in medicine. The leaves of *D. stramonium* are indicating the treatment of cell damage and antioxidant activity [2]. A variety of phytochemicals have been found to occur in *D. stramonium*. These phytoconstituents comprise alkaloids, flavonoids, phenol, tannins, saponins, sterol carbohydrate, and protein [3]. It has been scientifically proven to contain alkaloids, tannins, carbohydrates, and protein [3]. Phytosterol are a large group of compounds that are found exclusively in plants. They are structurally related to cholesterol but differ from cholesterol in the structure of the side chain.

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.12(4).2140-46

This article can be accessed online on www.ijpsr.com

DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.12(4).2140-46

Steroids have been used as blood cholesterol-lowering agents [4], prostatic hyperplasia, rheumatoid arthritis [5], cell damage, allergies, stress related illness, and inhibit the development of colon cancer [6]. Flavonoids are a group of plant metabolites thought to provide health benefits through cell signalling pathways and antioxidant effects [7]. *D. stramonium* is a rich source of tropane alkaloids [8]. The applications of alkaloids are not limited to biological control of herbivores but also have pharmacological, veterinary, and medical importance.

Terpenes are hydrocarbon compounds found in *D. stramonium*, most commonly associated with essential oils and represent many pharmaceutical uses. These chemicals form terpenoids, the basis of some antiseptic, expectorants, gastrointestinal drugs, and pain relievers. Terpenes also show promise as an antioxidant [9]. Heterocyclic, amino acids, phytol, and sulphur compounds are present in many natural habitat, including antitumor, antibiotic, anti-inflammatory, antidepressant, anti-malarial, antimicrobial, antidiabetic, herbicidal,

fungicidal, and insecticidal agents [10]. The present investigation aimed to identify the biologically active compounds in a leaf of Jimson weed using GC-MS with NMR and FTIR technique.

MATERIALS AND METHODS:

Plant Materials: Mature leaves of *Datura stramonium* were collected from the near ponds, River Bridge, Ladapuram village, Perambalur district, Tamil Nadu, India. The leaves of were washed thoroughly three times with water and once with distilled water. The leaves of the plant were air-dried in the shade for several days and powdered.

Extraction of Powdered Plant Material: The powders obtained were extracted separately with ethanol and water at room temperature (25 ± 2 °C). The resulting crude extracts were filtered and evaporated in a shaker water bath maintained at 55-65 °C. The obtained semi-dried crude extracts were contained in plastic containers and labelled

GC-MS Conditions: GCMS-QP 2010 plus was used for identification and quantification of biologically active compounds using MS libraries previously compiled from purchased standards for the acquisition of an electron ionization mass spectrum, an ion source temperature of 250 °C was used. The GC was equipped with a SE-30 capillary column a split injection piece (270 °C), and direct GC-MS coupling (280 °C). Helium (1.2 ml min^{-1}) was used as the carrier gas with a split ration 0f 1:10. The oven temperature program for analyzing the extracts utilized an initial oven temperature of 100 °C, maintained for 2 min, finally ramped to 300 °C at a rate of 7 °C min^{-1} for 10 min. Injection temperature and volume were 250 °C and 1µl, respectively. The total GC running time was about 43.28 min.

Identification of Components: GC/MS is a valuable aid for unknown identification peak as well as for confirming the identification of identified phytoconstituents. Identification of components was based on direct comparison of the retention times and mass spectral data with those for standard compounds and computer matching with the library (Wiley Library, NIST data bank, database NIST 98) as well as by comparison of the retention time those reported in the literature [11, 12, 13, 14, 15, 16, 17].

NMR Spectrum Analysis: The ^{1}H NMR and ^{13}C NMR spectra were recorded on the PROBHD PABBO BB-PULPROG spectrometer. 2D ^{1}H decoupled ^{15}N NMR spectra were recorded at 297.4K, operating at 30 MHz using a 5 mm broadband probe head. Spectra were accumulated using a 90° pulse angle, a recycle time of 10 s, and an acquisition time of 1.14 s, for a spectral width of 15 kHz for 32K data points. Before the Fourier transform, a zero filling to 64K was applied, and a line broadening of 0.30 Hz was used to improve the spectral signal-to-noise ratio.

FTIR Spectrum Analysis: Dried powder of methanol extract of plant materials was considered for instrumental analysis. The powdered sample of the plant specimen was treated for FTIR spectroscopy. Scan range from 400 to 4000 cm^{-1} with a resolution of 5 cm^{-1}.

RESULTS AND DISCUSSION: GC/MS is a useful and reliable method for rapid separation and identification of complex mixtures **Fig. 1** of Steroids. In the present study, a GC-MS procedure was applied for the identification of Steroids in the plant leaves of *D. stramonium* L. (Moroccan origin). More than 8 compounds in the Steroids fractions showed the characteristic mass spectral fragmentation of the Steroids and their metabolites.

To our knowledge 1.9% of five compounds of steroids are new for *D. stramonium* L., 5-Cholestene-3-ol, 24-methyl- **Fig. 1**, Ergost-5-en-3-ol, acetate, (3.beta., 24R)- **Fig. 2**, Androst-5-ene 1-acetoxy-16,17-dimethyl-20-oxo- **Fig. 3**, 7-De-hydrocholesterol isocaproate **Fig. 4** and Cam-pesterol **Fig. 5** with a molecular weight 400, 442, 386, 400 & 400 respectively and RT of the base 26 min. As well as 2% of compounds of Cholestan-3-one, 2-bromo-, cyclic 1, 2-ethanediyl acetal, (2. beta) with molecular weight 508 and RT of the base 22 min. As well as 2.7% of compounds of Cholestan-3-one, cyclic 1, 2-ethanediyl aetal, (5.beta.)- **Fig. 6** with molecular weight 430 and RT of the base 19 min. Allopregnane-7 alpha. 11. alpha-diol-3, 20-dione- is one of the steroid were detected at 11 min with molecular weight 348 from leaf of *D. Stramonium*. Steroids are important compounds used in medicine that reduce inflammation. The phytosterol 3-phenyl lactic acid, β-sitosterol, Cholesterol like compounds, Bras-

sterol, Stigmasterol, fucosterol, 5-ergosterol, Stigmasta 5.22-dien-3-ol and Cholestane were identified in leaf and callus of *Datura stramonium* L [18]. Additionally, flavonoids were detected in the intact plant of this species for the first time like Phthalic acid, butyl undecyl ester- **Fig .7** with the molecular weight 376 and RT of the base 20 min. Alkaloids 9 and 20 min possessed different retention times but identical mass spectral fragmentation and were determined as meth scopolamine like N'-(2,4,6(1H,3H,5H)-Trioxopyrimidin-5-y lidene-methyl) - 2 - nitroben **Fig. 8,** Scopolin like Butanamide, 3 - benzoylhydrazono - N - (2-trifluoromethylph enyl)- at 9 min **Fig. 11** and 3', 8, 8 '- Trimethoxy - 3 - piperidyl - 2, 2' - bina-phthalene -1,1',4,4'-tet **Fig. 10** at 20 min. In 25 minutes, alkaloids were detected like 2-Piperidinone, N-[4-bromo-n-butyl] **Fig. 9** with molecular weight 233. The stereochemistry of these alkaloids could not be established solely by MS data. It was suggested on the basis of their retention data reported in the literature. Generally, in Solanaceae, 3α-isomers of the homologous tropine esters occur in a considerably higher amount than the 3β-isomers. Sixty-seven tropane alkaloids were identified in the organs of *Datura stramonium* leaf by GC/MS. Hyoscyamine and scopolamine as the major tropane alkaloids in the plant organs [19].

D. stramonium L. is a plant with both poisonous and medicinal properties. The neurotoxicity is attributed to the presence of tropane alkaloids which contain a methylated nitrogen atom (N-CH_3) and include the anticholinergic drugs atropine and scopolamine as well as the narcotic cocaine [20]. It contains a variety of toxic tropane alkaloids such as atropine, Hyoscyamine, and scopolamine [21].

The two Terpenoids were detected in the intact plant of this species for the first time, like R-Limonene **Fig. 12** and Isopinocarveol with the molecular weight 184 & 152 and RT of the base 11 & 10 minutes, respectively. Amino acids were detected like Phenylserine, 2-fluoro-4, 5-dimethoxy-.bet. beta-didehydro and Sulfones compounds was detected like Sulfone, (3-amino-3-oxopropyl) (4-chlorophenyl)-with the MW 271 & 247 and RT of the base 13 min. Fluoro compounds like Ben-zamide, 3 - chloro - 4 - ethoxy - N - (6-fluorobenzothiazol-2-yl)- with MW 350 at 13 minutes. Sulphur compounds like 5, 9, 13-trimethyltetradecanoic acid 2, 2, 2- trifluoroethyl ester with MW 352 at 22 min. The detected the 2 Phytol compounds with MW 296 at 15 & 17 min. Finally, many heterocyclic compounds was detected like Lupan-3-one, cyclic 1, 2-ethanediyl acetal-, Cyclohexanone, 2-(1-methyl-2-nitroethyl)-, 2-Oxazolamine, 4, 5-dihydro-5-(phenoxymethyl)-N-[(phenyl mino) ca- and 4, 5, 6, 7-Tetrahydro-3H-cyclopenta [b] pyran-2-one. Heterocycles are present in a wide variety of drugs, most vitamins, many natural products, biomolecules, and biologically active compounds, including anti-tumor, antibiotic, anti-inflammatory, anti-depressant, anti-malarial, anti-HIV, antimicrobial, antibacterial, antifungal, antiviral, antidiabetic, herbicidal, fungicidal and insecticidal agents.

NMR Spectral analysis of the ^1H nmr spectrum a doublet peak is found at 7.685 & 7.705 ppm. It corresponds to p-substitited phenyl ring. The sharp peaks at 5.315 are due to OH group in the phenyl ring. The broad but small peaks from 5.2-3.8 ppm besides multiple at 4.222-4.236 ppm & 4.135ppm are assigned to proton in OH, NH & OCH_3 signal. At 2.761 ppm the NCH_3 signal is present. At 2.256 ppm & 2.012 ppm several broad peaks are found which may be due to azabicyclic ring protons. Multiple peaks from 1.63-0.835 correspond to CH_2 & CH_3 protons **Fig. 16.** Weak signals at 192 ppm can be assigned to C-OH carbon atom in phenyl ring. The signal at 167.6 ppm correspond to aliphatic C-OH or C=O carbon atom. At 132.3 & 129.4 ppm the peaks correspond to the ring C atom in ortho & para position to C-OH group the ring C atom para to C-OH group resonates to give a signal. At 68.4 & 71.6 ppm, the O-CH_3 & N-CH_3 carbon atoms give signals. The multiple in the region 40-14 ppm are due to aliphatic C atoms **Fig. 15**.

FTIR Spectral analysis of the broad peak centering at 3408.3 cm^{-1} corresponds to -OH and -NH stretching vibration modes. At 2929.6 cm^{-1}, a sharp stretching vibration peak corresponds to CH groups. At 2372.2 cm^{-1} C=N stretching vibration band is found. The broad at 1571.96 cm^{-1} can be attributed to >C=O stretching in COOH & >C=C- group in aromatic ring. The peaks at 1269, 1219.8 & 1127.1 cm^{-1} is due to C-O & C-C stretching vibrations. At 921.3cm^{-1}, the C-C ring vibration occurs, another peak at 831.88 cm^{-1} corresponds to

heterocyclic ring. The sharp vibration peak at 770.99 cm^{-1} is characteristic of bicyclic ring vibration. Several spectral peaks at 694.65, 619.92 & 433.7cm^{-1} correspond to bending vibrations of substitutes like OH, NCH_3 & NH_2 *etc*. **Fig. 13**.

The phytochemical analysis of the plant revealed that *D. stramonium* contained saponins, tannins and alkaloids, and glycosides. The secondary metabolites identified in the plant materials showed antimicrobial activity [22]. The compound is mostly Secondary metabolites such as alkaloids, steroids, tannins, and phenol compounds which are synthesized and deposited in the leaf of the *D. Stramonium* [23].

The rest of the biologically active compounds had been identified according to their fragmentation pattern reported in the literature, as indicated in **Table 1.**

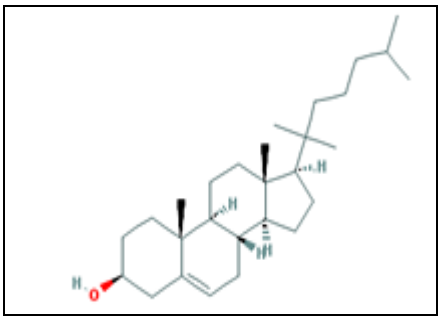

FIG. 1: 5-CHOLESTENE-3-OL

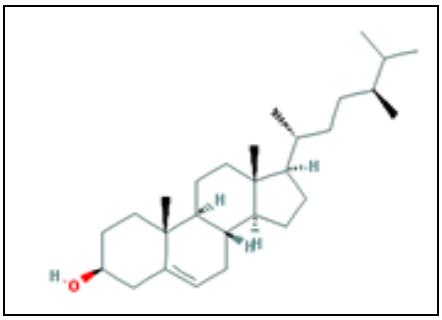

FIG. 2: ERGOST-5-EN-3-OL

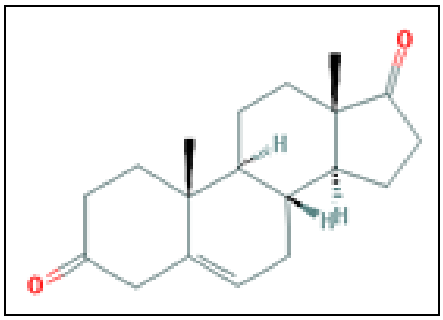

FIG. 3: ANDROST-5-ENE

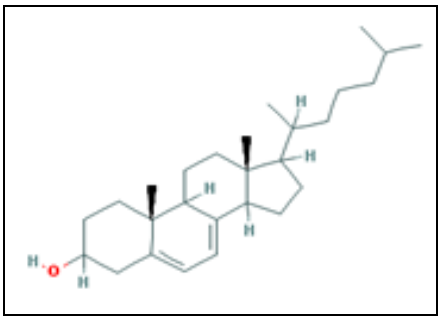

FIG. 4: 7-DEHYDROCHOLESTEROL

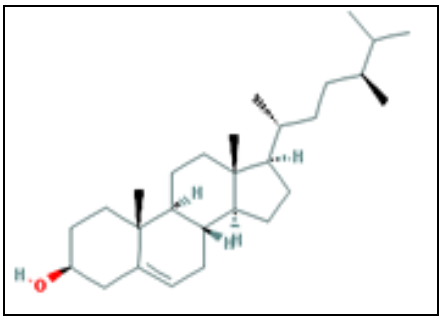

FIG. 5: CAMPESTEROL

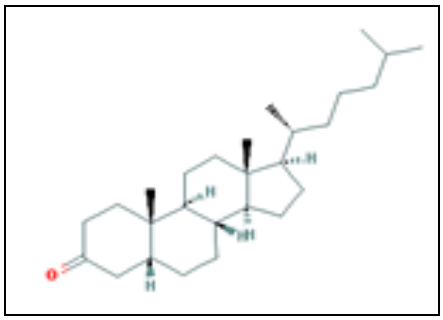

FIG. 6: CHOLESTAN-3-ONE, 5. BETA

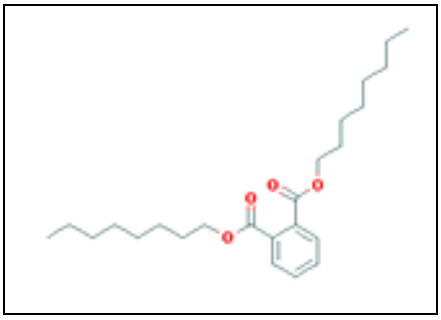

FIG. 7: PHTHALIC ACID

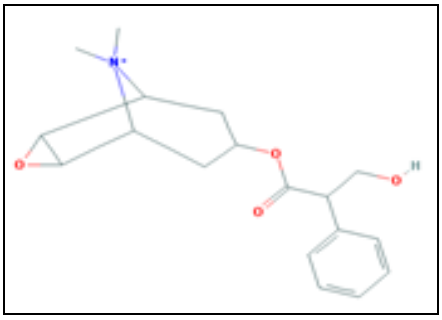

FIG. 8: METHSCOPOLAMINE

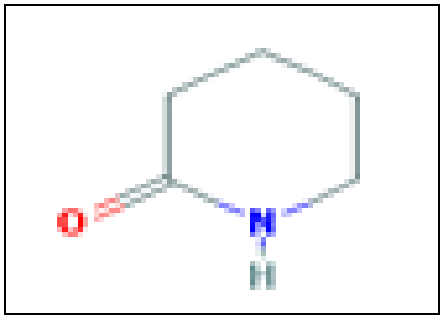

FIG. 9: PIPERIDINONE

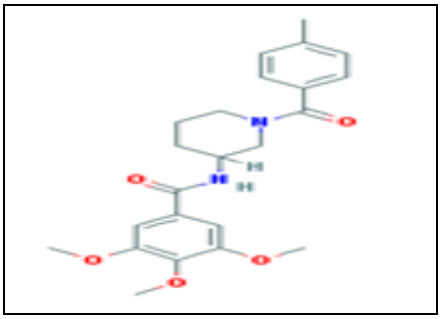

FIG. 10: 3-PIPERIDYL

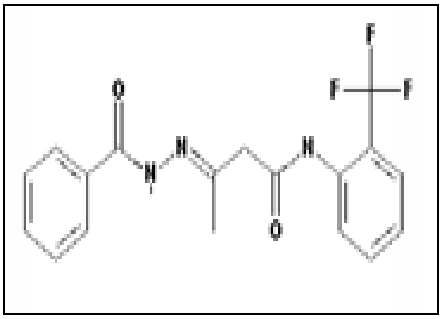

FIG. 11: BUTANAMIDE SCOPOLIN

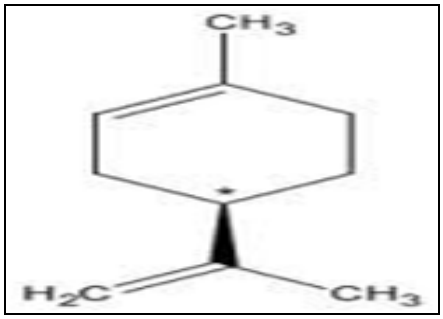

FIG. 12: R-LIMONENE

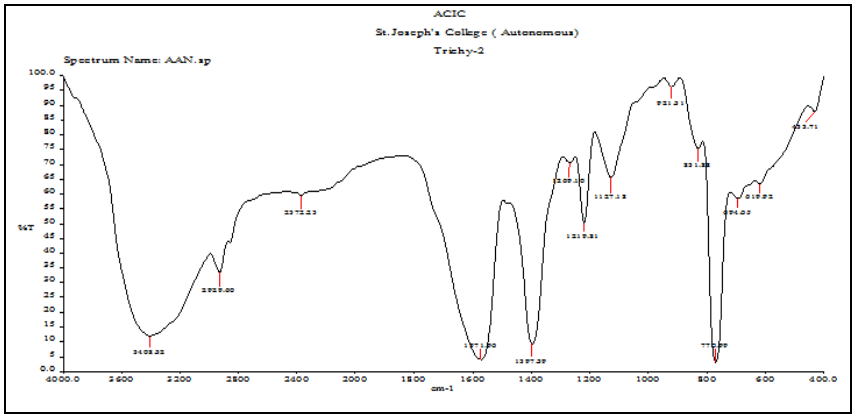

FIG. 13: FTIR ANALYSIS

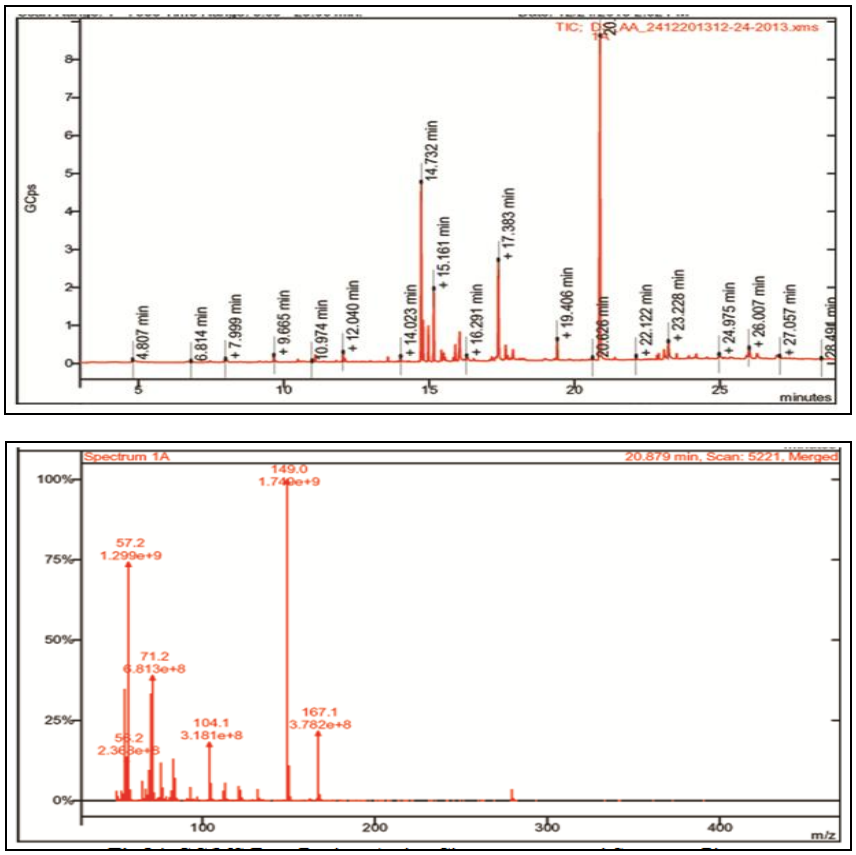

FIG. 14: GC/MS DATA REVIEW ACTIVE CHROMATOGRAM AND SPECTRUM PLOTS SCAN RANGE: 1 - 7563 TIME RANGE: 3.00 - 29.00 MIN

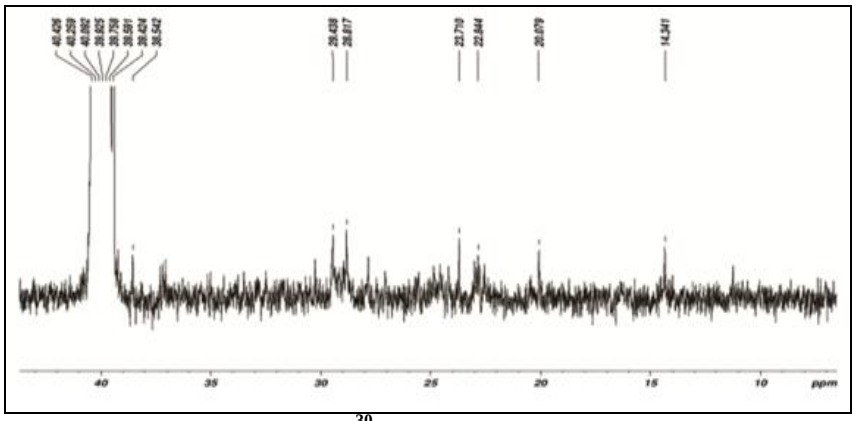

FIG. 15: ^{30}C NMR ANALYSIS

TABLE 1: BIOLOGICALLY ACTIVE COMPOUNDS IDENTIFIED IN *DATURA STRAMONIUM* LEAF PRESENTED AS A PERCENTAGE OF TOTAL ION CURRENT

S. no.	Retention Time	Compounds	Chemical Name	Molecular Weight
1	26 min	Steroids	5-Cholestene-3-ol, 24-methyl-	400
2			Ergost-5-en-3-ol, acetate, (3.beta.,24R)-	442
3			Campesterol	400
4			Androst-5-ene,1-acetoxy-16,17-dimethyl-20-oxo-	386
5			7-Dehydrocholesterol isocaproate	400
6	25 min	Breakdown compounds of Alkaloids	2-Piperidinone, N-[4-bromo-n-butyl]	233
7	22 min	Steroids	Cholestan-3-one, cyclic 1,2-ethanediyl aetal, (5.beta.)	430
8			Cholestan-3-one, 2-bromo-, cyclic 1,2-ethanediyl acetal, (2.beta	508
9		Sulfur compounds	5,9,13-trimethyltetradecanoic acid 2,2,2-trifluoroethyl ester	352
10		Heterocyclic compounds	Lupan-3-one, cyclic 1,2-ethanediyl acetal	470
11		Esters	3-Hydroxyhex-4-enethioic acid, S-t-butyl ester	202
12	20 min	Alkaloids	3',8,8'-Trimethoxy-3-piperidyl-2,2'-binaphthalene-1,1',4,4'-tet	487
13		Breakdown products of Flavonoids	Phthalic acid, butyl undecyl ester	376
14	19 min	Steroids	Cholestan-3-one, cyclic 1,2-ethanediyl aetal, (5.beta.)-	430
15	17 min	Phytol	Phytol	296
16	15 min	Phytol	Isophytol	296
17	14 min	Fluoro compounds	Acetic acid, trifluoro-, 3,7-dimethyloctyl ester	254
18	13 min	Sulfones	Sulfone, (3-amino-3-oxopropyl)(4-chlorophenyl)-	247
19		Aminoacids	Phenylserine, 2-fluoro-4,5-dimethoxy-.beta.,.beta.-didehydro-	271
		Fluoro compounds	Benzamide, 3-chloro-4-ethoxy-N-(6-fluorobenzothiazol-2-yl)-	350
20	11 min	Terpenoids	R-Limonene	184
21		Cyclic hydrocarbon	Cyclohexanone, 2-(1-methyl-2-nitroethyl)	185
22		Heterocycle	2-Oxazolamine, 4,5-dihydro-5-(phenoxymethyl)-N-[(phenylamino)ca	311
23		Steroids	Allopregnane-7.alpha.,11.alpha.-diol-3,20-dione	348
24	10 min	Terpenoids	Isopinocarveol	152
25	9 min	Alkaloids	N'-(2,4,6(1H,3H,5H)-Trioxopyrimidin-5-ylidenemethyl)-2-nitroben	319
26		Hydrozone of Alkaloids	Butanamide, 3-benzoylhydrazono-N-(2-trifluoromethylphenyl)-	363
27		Acids	3,6-Dinitro-4-cyclohexene-1,2-dicarboxylic acid	260
28		Quinolines	2,3-Bis(4-acetylanilino)-6,7-dinitroquinoxaline	486
29	8 min	Heterocycle	4,5,6,7-Tetrahydro-3H-cyclopenta[b]pyran-2-one	138

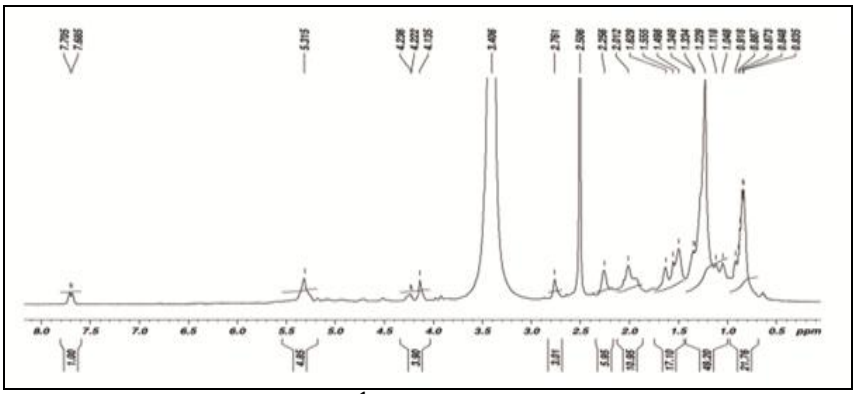

FIG. 16: ^{1}H NMR ANALYSIS

CONCLUSION: The leaf extracts of *Datura stramonium* showed the presence of eight Phytosterol like Campesterol, Sitosterol, *etc*., two Terpenes like R-Limonene, Isopinocarveol, Amino-acids like Phenylserine and some breakdown compounds of Alkaloids, Flavonoids were newly isolated compared with other biologically active compounds.

The ability of these compounds to inhibit the growth of microbes and also treatment of various human inflammations caused due to these pathogenic strains.

ACKNOWLEDGEMENT: The authors are grateful to Dr. Saravanan, Dept. of Chemistry, National College, Trichy, for providing us all Laboratory facilities. The authors would like to thank Dr. Radha, Government of Chemistry, SR College, Trichy, Mr. Mohanraj, and Mr. Vegatesh, Government of Chemistry, SCAS, Perambalur, for providing the necessary idea to carry out this research. My heartful thanks to My Parents for their financial support and continuous encouragement during the work.

CONFLICTS OF INTEREST: The author declares no conflicts of interest.

REFERENCES:

1. Berkov S, Zayed R and Doncheva T: Alkaloid patterns in some varieties of *Datura stramonium*. Fitoterapia 2006; 77: 179-82.
2. Bellakhdar J: La pharmacopée marocaine traditionnelle (medicine arabe ancienne et savoirs populaires). Saint-Étienne Ibis Press 1997; 494-6.
3. Philipov S and Berkov S: GC-MS investigation of tropane alkaloids in *Datura stramonium*. Z Naturforsch C 2002; 57: 559-61.
4. Ostlund JR, Racette SB and Stenson WF: Inhibition of cholesterol absorption by phytosterol-replete wheat germ compared with phytosterol-depleted wheat germ. Am J Clin Nutr 2003; 77: 1385-89.
5. Oomah BD and Mazza G: Health benefits of phytochemicals from selected Canadian crops. Trends Food Sci Technol 1999; 10: 193-98.
6. Bradford PG and Awad AB: Phytosterol as anticancer compounds. Mol Nutr Food Res 2007; 52: 161-70.
7. Havsteen BH: The biochemistry and medical significance of the flavonoids. Pharmacol Ther 2002; 96(2-3): 67-202.
8. Berkov S, Doncheva TS, Philipov S and Alexandrov K: Ontogenetic variation of the tropane alkaloids in *Datura stramonium*. Biochem Syst Ecol 2005; 33: 1017-29.
9. Ayoola GA: Phytochemical Screening and Antioxidant Activities of Some Selected Medicinal Plants Used for Malaria Therapy in Southwestern Nigeria. Tropical Journal of Pharmaceutical Research 2008; 7(3): 1019-24.
10. Katritzky AR: Handbook of heterocyclic chemistry. Oxford Oxfordshire, Pergamon Press, First Edition 1985.
11. Song YS, Jin C and Park EH: Identification of metabolites of phytosterols in rat feces using GC/MS. Arch Pharma Res 2005; 23: 599-604.
12. Holser RA, Bost G and Van Boven M: Phytosterol composition of hybrid *Hibiscus* seed oils. J Agric Food Chem 2004; 52: 2546-48.
13. Lembcke J, Ceglarek U, Fiedler GM, Baumann S, Leichtle and Thiery J: Rapid quantification of free and esterified phytosterols in human serum using APPI-LC-MS/MS. J Lipid Res 2005; 46: 21-26.
14. Phuruengrat A and Phaisansuthichol S: Preliminary study of steroids in *Sericocalyx schomburgkii* (craib) Bremek by GC-MS. Songklanakarin. J Sci Technol 2006; 28: 39-44.
15. Chen QL, Steinhauer, Hammerlindl J, keller W and Zou J: Biosynthesis of phytosterol esters: Identification of a sterol O-acetransferase in arabiodopsis. Plant Physiol 2007; 145: 974-84.
16. Delazer A, Nazifi E, Movafeghi A, Nazemiyeh H, Hemmati S, Nahar L and Sarker SD: Analysis of phytosterol and free radical scavengers in the bulbs of *Ornithogalum cuspidatum* Bertol. Boletin Latino americandel Caribe De Plantae Medicinales Aromatics 2010; 9: 87-92.
17. Winkler Moser J: Gas chromatographic analysis of plant sterols. The AOAC Lipid Library 2011; 1-18.
18. Bhardwaj R, Yadav A, Sharma P and Sharma RA: *In-vitro* and *in-vivo* GC-MS profile and Antimicrobial activity of Phytosterols of *Datura stramonium*. Res J of medicinal Plant 2014; 8(3): 112-20.
19. Ahmed El Bazaoui AB, My Ahmed Bellimam A and Abdelmajid Soulaymani B: Nine new tropane alkaloids from *Datura stramonium* L. identified by GC/MS. Fitoterapia 2011; 82: 193-97.
20. Maibam Rasila Devi1, Meenakshi Bawari1, Paul SB and Sharma GD: Neurotoxic and Medicinal Properties of *Datura stramonium* L. - Review. Assam University Journal of Science & Technology Biological and Environmental Sciences 2011; 7(1): 139-44.
21. Gaire1 BP and Subedi L: A review on the pharmacological and toxicological aspects of *Datura stramonium* L. J of Integrative Medicine 2013; 11(2): 73-79.
22. Banso A and Adeyemo S: Phytochemical screening and antimicrobial assessment of Abutilon mauritianum, Bacopa monnifera and *Datura stramonium*. Biokem 2006; 18(1): 39-44.
23. Kumar A, Garg BR, Rajput G, Chandel D, Muwalia A, Bala I and Singh S: Antibacterial activity and quantitative determination of protein from leaf of *Datura stramonium* and *piper betle* plants. Pharmacophore 2010; 1(3): 184-95.

How to cite this article:
Ananth A, Rajan S and Devi AU: Analysis of biologically active compounds in Jimson weed leafs. Int J Pharm Sci & Res 2021; 12(4): 2140-46. doi: 10.13040/IJPSR.0975-8232.12(4).2140-46.