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DETERMINATION OF BIOACTIVE COMPONENTS OF *ANNONA MURICATA* LEAF BY GC MS ANALYSIS

[1]Priscilla Sweetlin G., [2]Packia Lincy.M

[1]Department of Botany, Lady Doak College, Madurai, [2]Department of Botany, Lady Doak College, Madurai

Correspondence: Priscilla Sweetlin George [1]Department of Botany, Lady Doak College, Madurai

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ABSTRACT

Phytochemicals are the chemicals extracted from plants. These organic chemicals are classified as primary or secondary constituents, depending on their role in plant metabolism. GC-MS method used for the analysis of the obtained extract can be an interesting tool for testing the amount ofsome active principles in herbs used in various industries. The aim of this study was to carry out for identification of bioactive compounds from sample by Gas chromatography and Mass spectroscopy (GC-MS). GCMS analysis of ethanolic *Annona muricata* leaf extract was done by standard protocol using the equipment Perkin-Elmer Gas Chromatography– Mass Spectrometry, while the mass spectra of the compounds found in the extract was matchedwith the National Institute of Standards and Technology (NIST) library. The GC-MS analysis of*Annona muricata leaf extract* showed the presence of compounds. The prevailing compounds are 1,2-Benzenedicarboxylicacid, diethyl ester, Ergosta-5,22-dien-3-ol, acetate, (3á,22E), 6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one, 2-[4 - methyl-6-(2 ,6, 6 – trimethylcyclohex -1- enyl)hexa-1,3,5-trienyl]cyclohex – 1 -en -1-carboxaldehyde, Octadecanal, 1,2-Benzenedicarboxylicacid, butyl octyl ester which have wide range of biological activities.

INTRODUCTION

Annona muricata L., commonly known as soursop, graviola, or guanabana, belongs to the Annonaceae family, which comprises around 130 genera and 2300 species [1,2] *A. muricata* is an evergreen, terrestrial tree that grows 5–8 meters tall, with an open, roundish canopy and large, glossy, dark green leaves. Its edible fruits are large, heart-shaped, and green, with diameters ranging from 15 to 20 cm. [4]. All portions of the *A. muricata* tree, similar to other Annona species, including *A. squamosa* and *A. reticulata* are extensively used as traditional medicines against an array of human ailments and diseases, especially cancer and parasitic infections. The fruit is used as natural medicine for arthritic pain, neuralgia, arthritis, diarrhea, dysentery, fever, malaria, parasites, rheumatism, skin rushes and worms, and it is also eaten to elevate a mother's milk after childbirth. The leaves are employed to treat cystitis, diabetes, headaches and insomnia. Moreover, internal administration of the leaf's decoction is believed to exhibit anti-rheumatic and neuralgic effects, whereas the cooked leaves are topically used to treat abscesses and rheumatism [1,3,5]. Phytochemicals, organic compounds from plants, are classified into primary constituents like sugars, amino acids, and chlorophyll, and secondary constituents like alkaloids, terpenes, and phenolics. Research highlights their nutritional benefits and disease-fighting potential. Advances in techniques such as HPTLC, FT-IR, HPLC, GC-MS, and NMR have greatly enhanced the ability to analyze these compounds. [4]. *Annona muricata* leaves are rich in various phytochemicals and minerals, including essential oils and annonaceous acetogenin. These components enhance the plant's nutritional value, supplying essential nutrients like potassium, calcium, sodium, copper, iron, and magnesium to the human body. [6]. Like other Annona species, Annona muricata is frequently used in traditional medicine to treat a variety of illnesses, including parasite infections and cancer. A number of medical ailments, including rashes on the skin, neuralgia, diarrhea, fever, and arthritic pain, are treated using the fruit and other parts of the tree..[7,8]. With advanced automated techniques, GC-MS has become essential in analytical labs, offering enhanced selectivity and multi-compound analysis capabilities. The use of fused silica columns has made GC a leading method for analyzing complex mixtures. This study aims to identify the phytoconstituents in Annona muricata leaf extract using GC-MS.[9]. The aim of this study is to determine the phytoconstituents present in *Annona muricata* leaf extract with the aid of GC-MS technique.

MATERIALS AND METHODS

Collection of Leaves:

Fresh leaves of *Annona muricata* were collected from Kadayanallur , Tenkasi district ,Tamil Nadu between July 2023 and January 2024

Preparation of Plant Extract

The newly harvested leaves underwent a thorough cleasing process using running tap water. Subsequently, they were carefully dried in an oven at a controlled temperature of 40° C. Once completely dried, the leaves were coarsely ground using a grinder. The resulting powder was then subjected to extraction using absolute ethanol in a soxhlet extractor, maintaining a temperature range of 40-50° C. The extracted solution was subsequently dried on a water bath at 60° C. To further analyze the composition of this dried ethanol extract, GC-MS analysis was conducted.

RESULTS AND DISCUSSION

Gas Chromatography-Mass Spectrometry (GC-MS) is a powerful analytical technique that merges the capacities of gas-liquid chromatography and mass spectrometry to discern various compounds present in a given sample [10]. Over the recent years, GC-MS has gained significant recognition as a fundamental technological tool for characterizing secondary metabolities in both plant and non-plant organisms [11]. The Annona muricata leaf powder sample was analyzed using GC-MS chromatography, showing a total of 7 peaks (**Figure 1**). These peaks were then identified by comparing their mass spectra with databases of NIST libraries (**Table 1**).

Benzenedicarboxylic acid, diethyl ester, Ergosta-5,22-dien-3-ol,acetate, (3á,22E)- , 6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one, 2-[4-methyl-6-(2,6,6-trimethy lcyclohex-1-enyl)hexa-1,3,5-t rienyl]cyclohex-1-en-1-carbox Aldehyde, 2-bromo-Octadecanal .The biological activities of selected compounds were listed below **(Table 2).**

Table 1: Identification of phyto-constituents present in leaf sample of *Annona muricata*

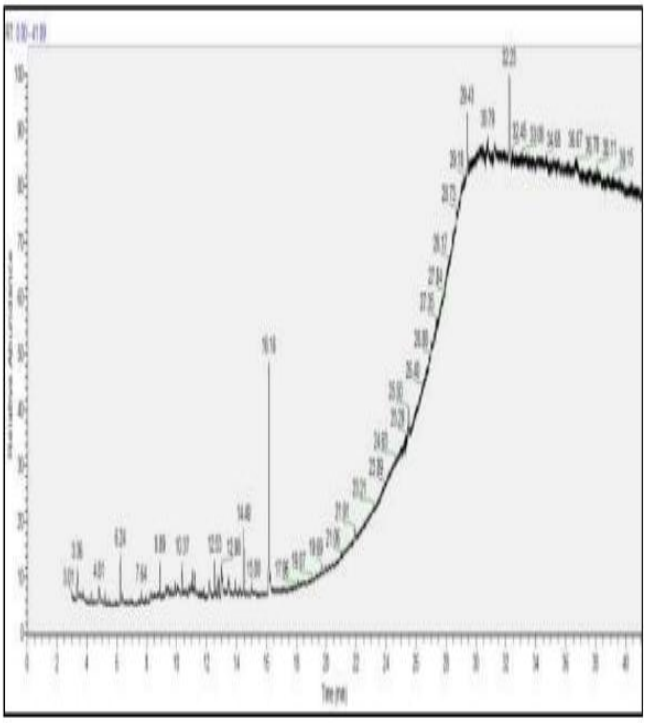
No	RT	Name of the compound	Molecular Formulae	Molecular Weight	Peak Area %
1.	10.37	1,2-Benzenedicarboxylic acid, diethyl ester	C12H14O4	222	5.11
2.	11.20	Ergosta-5,22-dien-3-ol, acetate, (3á,22E)-	C30H48O2	440	5.82
3.	12.54	6-Hydroxy-4,4,7a-trimethyl-5 ,6,7,7a-tetrahydrobenzofuran -2(4H)-one	C11H16O3	196	8.54
4.	12.73	2-[4-methyl-6-(2,6,6-trimethy lcyclohex-1-enyl)hexa-1,3,5-t rienyl]cyclohex-1-en-1-carbox aldehyde	C23H32O	324	4.53
5.	12.99	2-bromo-Octadecanal	C18H35BrO	346	12.57
6.	14.49	1,2-Benzenedicarboxylic acid, butyl octyl ester	C20H30O4	334	11.68
7.	16.16	Phytol	C20H40O	296	51.76

Table 1: Identification of phyto-constituents present in leaf sample of *Annona muricata*

Table 2: Biological activity of phyto-constituents present in the leaf sample of *Annaona muricata*

No	RT	Name of the compound	Biological activity	Reference
1.	10.37	1,2-Benzenedicarboxylic acid, diethyl ester	-	
2.	11.20	Ergosta-5,22-dien-3-ol, acetate, (3á,22E)-	anti-mosquito larvicidal antibacterial activity	[13]
3.	12.54	6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one	-anti-inflammatory activity, antioxidant and anticancer activity	[14]
4.	12.73	2-[4-methyl-6-(2,6,6-trimethylcyclohex-1-enyl)hexa-1,3,5-trienyl]cyclohex-1-en-1-carboxaldehyde	Antimicrobials and anti-virals	[15]
5.	12.99	Octadecanal, 2-bromo-	anti-inflammatory and anti-apoptotic effects	[16]
6.	14.49	1,2-Benzenedicarboxylic acid, butyl octyl ester	Antimicrobial Antifouling	[17]
7.	16.16	Phytol	Antimicrobial, anti-inflammatory, diuretic, Anti-cancer	[16]

Among the identified phytocompounds, 1,2-Benzenedicarboxylic acid and dibutyl phthalate act as plasticizers [18].Ergosta-5,22-dien-3-ol, acetate, (3á,22E)- was the most one of the important a compound have anti-mosquito larvicidal antibacterial activity[13]. 6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one otherwise called Loliolide has various biological activities such as Anti-diabetic, Antidepressive, Antioxidant activity- anti-inflammatory activity, anticancer activity [14]. 2-[4-methyl-6-(2,6,6-trimethylcyclohex-1-enyl) hexa-1,3,5-trienyl] cyclohex-1-en-1-carboxaldehyde have antimicrobial and anti-inflammatory activities[15].Octadecanal, 2-bromo has - anti-inflammatory and anti-apoptotic effects[16]. 1,2-Benzenedicarboxylic acid, butyl octyl ester have biological activities such as Antimicrobial, Antifouling[17]. Phytol possess Antimicrobial, Anticancer, Anti- inflammatory Antioxidant, Anti-diabetic,Anti-diuretic properties[16]. The present investigation clearly indicates the highest percentage of Phytol in the leaf sample of Annona muricata Phytol was used as an Antimicrobial, anti-inflammatory, diuretic, Anti-cancer, Anti-diabetic and Anti-diuretic properties[16]. Using Dr. Duke's phytochemical and ethno-botanical database (online), the biological activity of the identified phytocomponents was ascertained.[18].



From the results, it was observed that1,2-Benzenedicarboxylicacid, butyl octyl ester, Phytol were the major components in the leaf sample of *Annona muricata*. The other phyto-constituents present in the leaf sample are 1,2-

https://data.nal.usda.gov/dataset/dr-dukes-phytochemical-and-ethnobotanical-databases [18]

CONCLUSION

GC-MS analysis is a crucial initial step in understanding the active principles in medicinal plants and determining if the plant species contains specific compounds or groups of compounds. The GC-MS spectrum profile confirms the presence of key components by their retention times. The peak heights indicate the relative concentrations of these components in the extracts. By comparing the mass spectra of the constituents with the NIST library, the phytoconstituents were characterized and identified. In this study, Phytol was identified among the phytochemicals in Annona muricata samples and is known for its antimicrobial, anti-inflammatory, diuretic, anti-cancer, anti-diabetic, and antidiuretic properties [16], Based on the results obtained in the present investigation, it may be concluded that the biological activities of the identified phytocomponents used for anti-microbial, anti- inflammatory, anti-diabetic, antifouling anti-diuretic, anti-apoptotic and anti-cancer activities. From these results, it could be concluded that "***Annona muricata***" contains various bio-active compounds. Therefore, it is recommended as a plant of phytopharmaceutical importance.

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Conflict of interest: None.

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REFERENCES

- Mishra S., Ahmad S., Kumar N., Sharma B.K. *Annona muricata* (the cancer killer): A review. *Glob. J. Pharm. Res.* 2013;2:1613–1618.
- Leboeuf M., Cavé A., Bhaumik P., Mukherjee B., Mukherjee R. The phytochemistry of the annonaceae. *Phytochemistry.* 1980;21:2783–2813.
- Adewole S.O., Caxton-Martins E.A. Morphological changes and hypoglycemic effects of *Annona muricata* Linn. (Annonaceae) leaf aqueous extract on pancreatic B-cells of streptozotocin-treated diabetic rats. *Afr. J. Biomed. Res.* 2006;9:173–187.
- De Souza R., Benassi E., da Silva R.R., Afonso S., Scarminio I.S. Enhanced extraction yields and mobile phase separations by solvent mixtures for the analysis of metabolites in ***Annona muricata*** L. Leaves. *J. Sep. Sci.* 2009;32:4176–4185.
- De Sousa O.V., Vieira G.D.-V., de Pinho J.D.J.R., Yamamoto C.H., Alves M.S. Antinociceptive and anti-inflammatory activities of the ethanol extract of *Annona muricata* L. leaves in animal models. *Int. J. Mol. Sci.* 2010;11:2067–2078.
- Gyamfi K., Sarfo D., Nyarko B., Akaho E., Serfor-Armah Y., Ampomah-Amoako E. Assessment of elemental content in the fruit of graviola plant, ***Annona muricata***, from some selected communities in ghana by instrumental neutron activation analysis. *Elixir Food Sci.* 2011;41:5671–5675.
- Chukwunonso Agu K., Eluehike N., Oseikhumen Ofeimun R., Abile D., Ideho G., Olukemi Ogedengbe M., Omozokpea Onose P., Olalekan Elekofehinti O. Possible Anti-Diabetic Potentials of *A. muricata* (Soursop): Inhibition of α-Amylase and α-Glucosidase Activities. *Clin. Phytoscience.* 2019;5:21. 0116-0.
- Sokpe A., Mensah M.L.K., Koffuor G.A., Thomford K.P., Arthur R., Jibira Y., Baah M.K., Adedi B., Agbemenyah H.Y. Hypotensive and Antihypertensive Properties and Safety for Use of *A. muricata* and Persea Americana and Their Combination Products. *Evid. Based Complement. Altern. Med.* 2020;2020:8833828.
- 9. Hubschmann, H. (2015). Handbook of GC-MS: Fundamentals and Applications. Germany: Wiley.
- 10.Kell, D.B., Brown, M., Davey, H.M., Dunn, W.B., Spasic, I., Oliver, S.G., (2005) Metabolic footprinting and systems biology: The medium is the message. *Nat Rev Microbiol.*; 3:557-65.
- 11.Fernie, A.R., Trethewey, R.N., Krotzky, A.J., Willmister, L. (2004)., Metabolite profiling: From diagnostics to systems biology. *Nat Rev Mol Cell Biol.* 5:763-9.
- 12. Krishnamoorthy, K., & Subramaniam, P. (2014). Phytochemical profiling of leaf, stem, and tuber parts of Solena amplexicaulis (Lam.) Gandhi using GC-MS. *International scholarly research notices*, *2014*.
- 13. Irrusappan, H., Gokulakrishnan, J., Elumalai, K., Senthilmurugan, S., Vijayan, P., & Baranitharan, M. (2022). Entomotoxicity properties of eco-friendly methanol extract fractions from Phyla nodiflora (L.) Greene leaf exhibits mosquito larvicidal, pupicidal and antimicrobial activity. *World News of Natural Sciences*, *42*, 107-122.
- 14. Silva, J., Alves, C., Martins, A., Susano, P., Simões, M., Guedes, M., ... & Pedrosa, R. (2021). Loliolide, a new therapeutic option for neurological diseases? In vitro neuroprotective and anti-inflammatory activities of a monoterpenoid lactone isolated from codium tomentosum. *International Journal of Molecular Sciences*, *22*(4), 1888.
- 15. Hussein, H. J., Hadi, M. Y., & Hameed, I. H. (2016). Study of chemical composition of Foeniculum vulgare using Fourier transform infrared spectrophotometer and gas chromatography-mass spectrometry. *Journal of Pharmacognosy and Phytotherapy*, *8*(3), 60-89.
- 16.Kumar, D., Karthik, M., & Rajakumar, R. (2018). GC-MS analysis of bioactive compounds from ethanolic leaves extract of Eichhornia crassipes (Mart) Solms. and their pharmacological activities. *Pharma Innov J*, *7*(8), 459-462.
- 17. Beulah, G. G., Soris, P. T., & Mohan, V. R. (2018). GC-MS determination of bioactive compounds of Dendrophthoe falcata (LF) Ettingsh: An epiphytic plant. *Int. J. Health Sci. Res*, *8*, 261-269.
- 18. Dukes. Phytochemical and Ethnobotanical Databases. Phytochemical and Ethnobotanical Databases.www.ars-gov/cgi-bin/duke/. 2019.