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Fatty Acid Composition of Petroleum Ether Extract of Seed of *Terminalia bellirica*

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Abstract *Terminalia bellirica* is a medicinally important plant belonging to the family of Combretaceae. It has been used extensively by ayurvedic practitioner and has antidiabetic, anticancer and antibacterial properties. The fatty acid compositions of the petroleum ether extract of seeds of this plant were determined by gas chromatography-mass spectrophotometer. 5 compounds were identified from the extract of seeds (60.88 %) and they are palmitic acid, linoleic acid, Oleic acid, stearic acid and arachidic acid of methyl ester. Among all fatty acids of palmitic acid showed the highest concentration (13.13%).

Keywords *Terminalia bellirica*, GC-MS, fatty acid compositions.

1. Introduction

Plant products have been part of phytomedicine since time immemorial. These can be derived from any part of the plant like leaves, flowers, bark roots, fruits and seeds etc [1]. Any part of the plant may contain active components. Herbal medicines have become more popular in the treatment of any diseases due to popular belief that green medicine is safe, easily available and with less side effects. Many plants are cheaper and more accessible to most people especially in the developing countries than orthodox medicine, and there is lower incidence of adverse effects after use. These reasons might account for their worldwide attention and use [2]. The medicinal properties of some plants have been documented by some researchers [3-5]. Medicinal plant constitutes are the main source of new pharmaceuticals and healthcare products [6]. Extraction and characterization of several phytocompounds of these green factories have given birth to some high activity profile drugs [7]. Indeed, the market and public demand has been so great that there is a great risk that many medicinal plants today, face either extinction or less of genetic diversity [8]. Knowledge of the chemical constituents of the plant is desirable because such information will valuable for the synthesis of complex chemical substances.

Terminalia bellirica, is known as "Bahera" or Beleric [9], Sanskrit: Vibhitaka, Aksha is a large deciduous tree common on plains and lower hills in Southeast Asia, where it is also grown as an avenue tree. The leaves of the plant *Terminalia bellirica* (Gaertn) Roxb are about 15 cm long and crowded toward the ends of the branches. It is considered a good fodder for cattle. *Terminalia bellirica* seeds are oil bearing. The seeds are called bedda nuts. In traditional Indian Ayurvedic medicine, Beleric is known as "Bibhitaki" (Marathi: "Behadaor Bhenda") (*Terminalia bellirica*). In its fruit form, it is used in the popular Indian herbal rasayana treatment triphala. The nuts of the tree are rounded but with five flatter sides. It seems to be the nuts that are used as dice in the epic poem Mahabharata. A handful of nuts would be cast on a gaming board and the players would have to call whether an odd or even number of nuts had been thrown [10].

Terminalia Billerica (Bahera) is a very important herb. It is large tree with broadly epileptic leaves clustered at the ends of branches [11]. The leaves of the plant possess proteins which makes it promising angiogenic agent [12]. The fruits of this plant are spherical to ovoid. The dry fruit are widely used in Ayurveda, Siddha and Chinese systems of medicine [13]. It has antidiabetic, anticancer and anti microbial properties [14]. The fruit rind is an important ingredient of triphala, an important Ayurvedic formulations [15]. The fruit extracts stimulates the secretion of insulin and enhance its action and inhibits starch digestion [16]. It possesses active compounds which can be used to develop antidiabetic drugs [17]. The fruit pulp possess active compounds phtosterols, triterpenoids, glycosides, tannins and phenolic compounds which accounts for its anti-inflammatory, analgesic, antibacterial, antioxidant and antitumor properties [18].

However many researches have been carried out on *Terminalia bellirica*, is but no systematic research on fatty acid composition of seeds of the plant by GC-MS analysis has been reported. Therefore, the present study was undertaken with an objective to carry out a complete investigation of the compositions of fatty acids of seeds from petroleum ether extract of *Terminalia Billerica* with GC-MS analysis.

2. Materials and Methods

2.1. Collection of Plant Material

Fully matured fresh seeds of *T. bellirica* were collected from local area of Rajshahi district, Bangladesh in the month of April 201**7** and identified by the taxonomist of Bangladesh National Herbarium, Dhaka, where a voucher specimen (No.=4393) has been deposited.

2.2. Preparation of Sample

The mature seeds were washed to remove dirt. Then it was oven-dried at reduced temperature less than 45^0C to make it suitable for grinding purpose. The screened (20 mesh) powder was then stored in air-tight container with marking for future experiment.

2.3. Solvents

Petroleum ether (B.P 40-60 °C, Merck, Germany) and all chemicals are of AR grade, under normal atmospheric pressure was employed for extraction of plant material. Solvent from extract were recovered under distillation and the dried extracts were preserved in a refrigerator.

2.4. Extraction of fatty acids and preparation of methyl ester (FAMEs)

The matured seeds of *T. bellirica* was collected and washed individually from running tap water to remove soil particles and other dust. Then they were dried at room temperature and powdered by Fritsch mortar grinder, Germany. The natural fatty acids were extracted separately from the seed powder (100 gm) of the plant with petroleum ether (B.P 40 °C – 60 °C) in a Soxhlet apparatus for 72h. The extracts were concentrated under reduced pressure in a rotary evaporator. The extracts were filtered using Whatman No.1 filter paper and then vacuum distilled to remove solvent completely. The extracts from the seeds of *T. bellirica* was 6.38 gm (6.38% w/w) Petroleum ether extracts for seeds of *T. bellirica* were kept in a nitrogen atmosphere in a refrigerator. The fatty acids present in the extracts were converted to fatty acid methyl esters (FAMEs) first and analyzed according to the method reported by Griffin [19] for GC-MS analysis.

The fatty acid composition was determined by analysis of their methyl esters. The fatty acid methyl esters (FAMEs) were prepared by esterification reaction by using BF_3-MeOH complex according to AOAC method [20]. 10 mg of extract of seeds was taken in a screw capped glass tube. 1 ml of BF_3-MeOH complex were added and then heated at 100 °C for 1 hour in a water bath. After that it was cooled at room temperature and 1ml of deionized water & 2ml of hexane were added. The glass tube was vortexed and centrifuged at low RPM for two minutes. The upper layer was collected by means of syringe and kept in closely tight glass vial in refrigerator. Then the prepared FAMEs were ready to analyze.

2.5. Gas Chromatograph-Mass Spectrum Analysis

GC-MS analysis of seeds of *T. bellirica* from petroleum ether extracts were carried out on Agilent 7890A system equipped with mass Spectrophotometer detector and split less injection system. The GC was fitted with a HP-5MS capillary column (30 m X 0.25mm: film thickness: 0.25µm). The temperature program was as follows: injector temperature 260 °C, initial oven temperature at 70 °C, then increased at 10 °C/min to 150 °C for 5 min., then 12 °C/min to 200 °C for 15 min. and then12 °C/min to 220 °C for 15 min. Helium was used as the carrier gas at 17.69 psi pressure with flow 0.6 ml/min. Samples were dissolved in methanol and 1µl aliquot was injected automatically. MS was set in scan mode. The ionization was electron ionization. The mass range was set in the range of 50-550 m/z. MS spectra of separated components were identified on NIST libraries for fatty acid compositions.

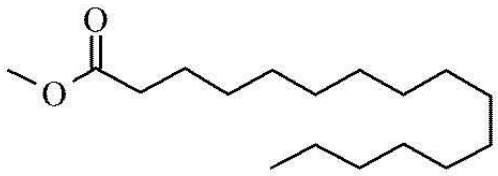
3. Result and Discussion

GC-MS analysis of fatty acids of seeds of *T. bellirica* petroleum ether extract showed the presence of 5 compounds for seeds of *T. bellirica.* GC analyzed results which include the active principles with their retention time, molecular formula, molecular weight and composition of the fatty acids of seeds of *T. bellirica* from petroleum ether extract are presented in table-1 & figure -1 respectively.

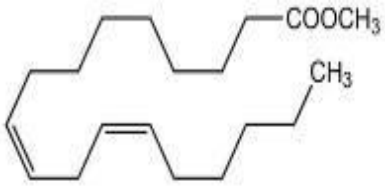
Table 1: GC-MS analysis of fatty acids from petroleum ether extract of seeds of *T. bellirica*

Sl No.	Retention time (min)	Name of the compound	Molecular weight	Molecular Formula	Conc. (%)
1.	13.64	Methyl Palmitate	270.45	$C_{17}H_{34}O_2$	13.13
2.	16.08	Methyl Linoleate	294.47	$C_{19}H_{34}O_2$	12.13
3.	16.21	Methyl Oleate	296.49	$C_{19}H_{36}O_2$	12.92
4.	16.48	Methyl Stearate	298.50	$C_{19}H_{38}O_2$	11.56
5.	19.07	Methyl Arachidate	326.56	$C_{21}H_{42}O_2$	11.14

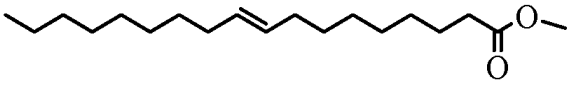
Total 5 fatty acids were identified as their methyl esters in the case of seeds of *T. bellirica*. The major constituent was Methyl Palmitate (13.13%) with retention time 13.64.



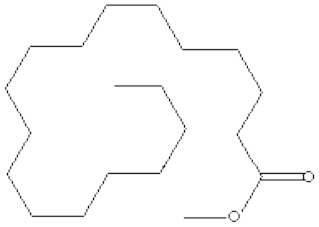
Methyl Palmitate



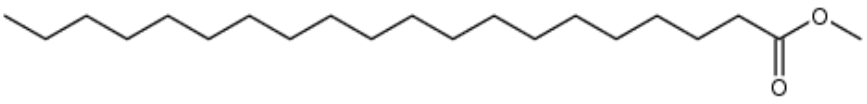
Methyl Linoleate



Methyl Oleate



Methyl Stearate



Methyl Arachidate

Figure 1: Structure of the identified fatty acid esters from GC-MS analysis of petroleum ether extract of seeds of T. bellirica

4. Conclusion

The present study found 5 constituents from seeds of petroleum ether extract of *T. bellirica* by Gas Chromatography-Mass spectroscopy (GC-MS) analysis. The presence of these chemical compounds justified the extensive uses of seed of the plant by traditional practitioner to treat various ailments. It could be concluded that *T. bellirica* contains various chemical constituents that can be bioactive compounds of medical importance. However, further studies are needed to evaluate its bioactivity and toxicity profile.

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References

1. Cragg, G. M., Newman, D. J. (2001). Natural Product drug discovery in the next millenium. *Pharm Biol* , 1:8-17.
2. Sofowora A. (1993). Medicinal plants and traditional medicine in Africa: Spectrum Books Ltd, Ibadan, Nigeria, pp 289.
3. Mensah, J. K., Okoli, R.I., Ohaju-Obodo, J.O. and Eifediyl, K. (2008). Phytochemical, Nutritional and medical properties of some leafy vegetables consumed by Edo people of Nigeria. *African Journal of Biotechnology*, 7(14): 2304-2309.
4. Gill, L.S. (1992). Ethanobotanical uses of plants in Nigeria: University of Benin Press, Benin city, pp 350.
5. Banso, A. and Adeyemo, S.O. (2007). Evaluation of antibacterial properties of tannins isolated from Dichrostachys cinerea. *African Journal of Biotechnology*, 6: 1785-1787.
6. Ivanova, D., Gerova, D., Chervenkov, T., Yankova, T. (2005). Polyphenol and antioxidant capacity of Bulgarian medicinal plants. *J Ethanopharmacol*, 96:145-150.
7. Mandal, V., Mohan, Y., Hemalatha, S. (2007). Microwave assisted extraction-An Innovative and promissing Extraction tool for Medicinal Plant research. *Pharmacog Rev*, 1: 7-18.
8. Misra., A. (2009). Studies on biochemical and physiological aspects in relation to phyto-medicinal qualities and efficacy of the active ingredients during the handling, cultivation and harvesting of the medicinal plnats. *Journal of Medicinal plants research*, 3: 1140-1146.
9. Raymond, Arveill Addenda au FEW XIX (Orientalia). (1999). Max Niemeyer Verlag. Tübingen.
10. Judd, S.W., Christopher, C.A., Elizabeth, K.J., Michael, D. and Peter, S. (2002). Plant Systematics: A Phylogenetic Approach, 2nd ed., Suderland.
11. Meena, A.K., Ajay, Y., Uttam, S., Sandeep, K., Rao, M.M. (2010). Evaluation of physiochemical parameters on the fruit of Terminalia bellirica Roxb. *Int. J of pharmacy Pharm*, 2: 97-99.
12. Vinoth, P.V., Chidambaranathan, N., Gopal, V. (2012). Evaluation and quantification of angiogenesis activity of *Terminalia bellirica* Roxb, by mice sponge implantation method. *Pharmacology*, 4(1):22-27.
13. Saraswathi, M.N., Karthikeyan, M., Kannan, M., Rajasekar, S. (2012). *Terminalia belerica* Roxb-A phytopharmacological Review. *IJRPBS*, 3(1):96-99.
14. Kumudhavalli, M.V., Vyas, M., Jayakar, B. (2010). Phytochemical and Pharmacological evaluation of the plant fruit of *Terminalia belerica* Roxb. *IJPLS*, 1(1):1-11.
15. Kamali, S.H., Khalaj, A.R., Shirin, H.R., Esfehani, M.M., Kamalinejad, M., Larijani, B.A. (2013). Systematic review of the antioxidant, anti- diabetic, and anti-obesity effects and safety of triphala herbal formulation. *Journal of Medicinal Plants Research*, 7(14):831-844
16. Violet, K., Peter, R.F., Taseer, H. (2010). *Terminalia bellirica* stimulates the secretion and action of insulin and inhibits starch digestion and protein glycation *in vitro*. *British Journal of Nitro*, 212-217.

17. Latha, R.C.R., Daisy, P. (2010). Influence of *Terminalia belirica* Roxb. Fruit extracts on Biochemical parameters in Streptozotocin Diabetic Rats. *International Journal of Pharmacology*, 6(2):89-96.
18. Monahar, V.R., Chandrashekar, R., Rao, S.N. (2012). Phytochemical analysis of *Terminalia belirica* fruit pulp extracts. *World journal of pharmacy and Pharmaceutical Sciences*, 194:1376-1383.
19. Association of Official Analytical Chemists. (1995) "Official Methods of Analysis". 16th Ed: Washington DC, ch -41: pp. 14 -15.
20. AOAC. Association of Official Analytical Chemist.(1984). Official Methods of Analysis, 14th Edn. Official methods 28.059. Sydney, Williams. Arlington, USA.