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Murraya koenigii (Linn.) Spreng.: An opulent source of fatty acid

Shahin Aziz*

Chemical Research Division, BCSIR Laboratories Dhaka, Bangladesh Council of Scientific and Industrial Research, Dhaka-1205, Bangladesh

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ABSTRACT

Murraya koenigii L., a medicinal plant utilized in traditional folk medicine, possesses anti-diabetic, anti-microbial, anti-inflammatory attributes and also is widely used for the treatment of hemorrhoids, itching, leukoderma, and hematological disorders. From the present work, total 8 fatty acids were identified by Gas Chromatography–Mass PSpectrometry (GC-MS) technique where the level of saturation in the fatty acid derived from the petroleum ether extract of the aerial section of *Murraya koenigii* L. is significantly higher compared to unsaturated part. The saturated portion includes capric acid, myristic acid, palmitic acid, stearic acid and arachidic acid while palmitic acid is obtained at higher concentration (35.07%). Conversely the unsaturated portion comprises oleic acid, linoleic acid, α-linoleic acid where oleic acid covers a significant concentration (17.31%). In connection to the above findings, the current study indicates that the significant presence of fatty acids such as palmitic acid, oleic acid, α-Linolenic acid may contribute to the recognition of the potential pharmacological significance of this plant in the treatment of illnesses.

***Corresponding author:** Shahin Aziz ✉ shaziz2408@yahoo.com

INTRODUCTION

Plant parts such as the flower, leave, root; bark, seed, and fruit have been used in herbal and ayurvedic medicine since time immemorial (Basit *et al.,* 2023; Shahin *et al.*, 2016). Continuous and extensive research revealed that medicinal plants are the prime source of new bioactive compounds and healthcare products with therapeutic qualities (Banso *et al.,* 2007; Ivanova *et al.,* 2005; Shahin *et al*, 2019). Through the successive extraction and characterization of numerous phytochemicals from the vast natural repository, several drugs with high activity profiles were identified and industrially synthesized (Mandal *et al.,* 2007, Misra *et al.,* 2009).

Murraya koenigii L., a member of the Rutaceae family, is frequently referred to as "kamini, Kariaphuli, Gandhal, Curry Pata" in Bangladesh. It is strongly aromatic deciduous shrub or small tree having clusters of small white flowers, small ovoid black fruits, and fragrant leaves grown across the different regions of Bangladesh as well as tropical and sub-tropical areas in the world (Abdelwahab *et al.,* 2023). The leaves of *M. koenigii* are used as spice to season a variety of meals, though they are most frequently employed in curries. Curry leaves are exceptionally abundant in chemical constituents such as essential oil, tannins, resin and crystalline glucoside, koenigin (Ghani, 1998) with a variety of pharmacological and biological activities, for an instance, antidiabetic (Arulselvan and Subramanian, 2007), antioxidant (Baliga *et al.,* 2003), antimicrobial (Abhishek *et al.,* 2010), hepatoprotective (Pande *et al.,* 2009), antiinflamatory (Muthumani *et al.,* 2009), antihyper cholesterolemic (Iyer *et al.,* 1990), effective action against colon carcinogenesis (Iyer *et al.,* 1990), increasing of digestive secretions, relief from nausea, indigestion, vomiting, diarrhea, dysentery, fever and snakebite as well as nutritional and fragrant properties (Ghani, 1998; Abdelwahab *et al.,* 2023, Shashank *et al.,* 2020). Furthermore, girinimbin was obtained from the stem-bark part while the flowers contain a significant quantity of mono- and sesquiterpenoids.

The primary terpenoids found in the flowers are beta-caryophyllene, beta-ocimene, and linalool (Ghani *et al.,* 1998). In addition, both free and complex lipid-bound fatty acids are essential for metabolism because they function as a metabolic fuel, storing and transferring energy, as a building block of all membranes, and as a gene regulator. Fatty acids are essential for mechanical protection, electrical and thermal insulation, and complex lipids. Fatty acids' amphipathic qualities and ability to form micelles additionally provide them a variety of industrial uses as soaps and detergents (Furuhashi *et al.,* 2008). Three most abundant unsaturated fatty acids (UFAs) in plants are oleic acid (918:1), linoleic acid (918:2), and α-linoleic acid (18:3), all of which consist of 18 carbon atoms. These relatively simple compounds serve as constituents and regulators of glycerolipids, triacylglycerols as a carbon and energy reservoir, stores of constituents of the extracellular barrier such as cutin and suberin, predecessors of different biologically molecules like nitro alkenes and jasmonates, and regulators of stress signaling. However, they have the ability to cause oxidative stress (He *et al.,* 2020).

According to literature study, *M. koenigii* has been subjected to numerous investigations. Many of the chemical components of *M. koenigii* show signs of pharmacokinetic response. As per plant science, different geographic locations, climatic circumstances and environmental influences produce non-identical plant secondary metabolites linked to physiological variances in plants.

For this reason, a number of thorough scientific investigations on the effectiveness of the entire plant or specific parts in various extract have been accomplished for medical purposes. However, fatty acid profiling by GC-MS analysis of aerial sections of *M. koenigii* has not been reported at all. Therefore the current study focuses a comprehensive GC-MS assessment of the fatty acid compositions in the aerial sections of the petroleum ether extract of *M. koenigii*, a species native to Bangladesh.

MATERIALS AND METHODS

Fresh and matured *M. koenigii* plant was gathered from Munshiganj district, Bangladsh. The taxonomist recognized the plant at the Bangladesh National Herbarium, Dhaka where a voucher specimen (DACB accession number - 43203) was recorded.

The flower, leaf, stem, and seed of the plant were dried and cleaned before being crushed into a powder using a German Fritsch mortar grinder and screened through a 20 mesh filter.

The petroleum ether at standard atmospheric pressure used as a solvent for extraction part was recovered using distillation and the dried extracts were stored in a refrigerator. The reagents utilized were sourced from Merck (Darmstadt, Germany) and Sigma Aldrich (Buchs, Switzerland).

The powdered components of aerial parts were separately immersed in petroleum ether using a soxhlet apparatus for duration of 72 hours. This method was based on the procedure described by Aziz *et al.* (2016). The extracts was undergone filtration using Whatman No.1 filter paper as well as the vacuum distillation process was followed to entirely remove the solvent. The extracts were condensed using a rotary evaporator under reduced pressure. The plant's petroleum ether extract (5.82% w/w) obtained was stored in a refrigerator under an inert environment.

By analyzing the respective fatty acid methyl esters, the composition of fatty acids was determined. Firstly, the fatty acids were transformed into fatty acid methyl esters, or FAMEs.

Afterwards 10 g of the extract were examined using the Griffin (Winterkorn and Fang 1991) technique and the AOAC method (Herborne *et al.,* 1984) using the BF_3-MeOH complex. Using TLC, the complete conversion of esterification process was verified. Afterwards fatty acid esters of *M. koenigii* were subjected to GC-MS analysis using an Agilent 7890A system fitted with a split-less injection mechanism and a mass spectrophotometer (MS) detector. A HP-5MS capillary column (30 m X 0.25mm; film thickness: 0.25µm) was installed in the GC. During the temperature program's operation, the injector temperature was kept at 260˚C. The oven's starting temperature was 70˚C. After that, the gradual increment of temperature was 10ºC/min to 150 ˚C for 5 minutes, 12˚C/min to 200˚C for 15 minutes, and lastly 12˚C/min to 220˚C for 15 minutes. The carrier gas, helium, was employed at a pressure of 17.69 psi with a flow rate of 0.6 ml/min. After the samples were dissolved in methanol, an automated injection of 1µl aliquot was made. Mass spectra (MS) were in the scanning mode. The mass range was established between 50 and 550 m/z. For fatty acid compositions, NIST libraries were used to identify the MS spectra of the separated components. The peaks were identified using standard FAMEs (E. Merck).

RESULTS AND DISCUSSION

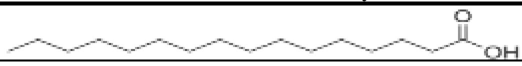
In the GC-MS study of *M. koenigii*, the fatty acid profile comprises five saturated and three unsaturated fatty acids obtained from the petroleum ether extract of aerial parts. The qualitative and quantitative profiling of fatty acids determined by GC-MS is expressed in form of concentration (%), the molecular weight and molecular formula. Table 1 shows four notable concentrations (%) of saturated and unsaturated fatty acids. Among all fatty acids, palmitic acid with retention time 22.33 has the highest concentration (35.07%). In addition, oleic acid at retention time 31.61 refers the second highest concentration (17.31%) while the content of α-Linolenic acid with retention time 36.11 decreased by 4.01% compared to oleic acid.

Moreover, arachidic acid showed the significant concentration 21.7% at retention time 37.85 whereas linoleic acid revealed its presence at retention time 33.62 with concentration 7.85%. Finally, capric, myristic and stearic were obtained at very low concentration below 2% with retention time 1.81 1.76 and 1.20 respectively. The chemical structures of the most predominant compounds from petroleum ether extract of *M. koenigii* have been shown in the Table 2.

Table 1. GC-MS results of fatty acids from petroleum ether (PE) extract of the aerial part of *M. koenigii*

Sl	Compounds name	Retention time (min.)	Molecular weight	Molecular formula	Concentration (%)
1.	Capric acid (10:0)	4.07	172.26	$C_{10}H_{20}O_2$	1.81
2.	Myristic acid (14:0)	14.11	228.37	$C_{14}H_{28}O_2$	1.76
3.	Palmitic Acid (16:0)	22.33	256.42	$C_{16}H_{32}O_2$	35.07
4.	Stearic acid (18:0)	30.86	284.47	$C_{18}H_{36}O_2$	1.2
5.	Oleic acid (18:1)	31.61	282.46	$C_{18}H_{34}O_2$	17.31
6.	Linoleic acid (18:2)	33.62	280.44	$C_{18}H_{32}O_2$	7.85
7.	α-Linolenic acid (18:3)	36.11	278.43	$C_{18}H_{30}O_2$	13.3
8.	Arachidic acid (20:0)	37.85	312.53	$C_{20}H_{40}O_2$	21.7

Table 2. Chemical structures of the most predominant fatty acids obtained from PE extract of *Murraya koengii* Linn

Name of fatty acids	Chemical structure of fatty acids
Palmitic acid	
Oleic acid	
A-Linolenic acid	
Arachidic acid	

Previous study showed that cardiovascular health is significantly influenced by saturated fats (Abdelwahab *et al.,* 2023). They are necessary for the proper insertion of calcium into the bone.

Acetaminophen and other regularly used pain and arthritis medications, as well as alcohol, demonstrated liver damage that can be prevented by eating saturated fatty acids. Some saturated fatty acids, especially those present in plants, act as direct signaling messengers that impact metabolism and affect vital processes such as the proper release of insulin. The capacity of white blood cell to identify and eliminate foreign invaders like viruses, bacteria, and fungi is reduced when required quantity of saturated fatty acids are not present (Peichev *et al.,* 2000). Conversely unsaturated fatty acids decrease the probability of developing diabetes. Vitamins A, D, E, and K, which are fat-soluble, exhibit enhanced absorption in the intestines when consumed with unsaturated fat. They are the primary sources of energy derived from food. Consumption of a substantial amount of protein in conjunction with unsaturated fat is advantageous for the overall well-being of the brain (Lunn *et al.,* 2006). In connection to the above literature review, the aerial section of *M. koenigii* provides a scientific basis for the traditional usage of this essential medicinal plant in treating numerous ailments since the current investigation clearly demonstrates that the aerial section of *M. koenigii* contains significant quantities of both saturated and unsaturated fatty acids. The levels of saturation and unsaturation in the fatty acid derived from the petroleum ether extract of the aerial section of *M. koenigii* are shown in Fig. 1.

Fig. 1. *Murraya koenigii* L.

Fig. 2. Depicted that the saturated fatty acid content is significantly high compared to unsaturated fatty acids. The essential fatty acids, namely linoleic acid and α-linolenic acid, are not biochemically synthesized in the human body due to the absence of necessary enzymes. Linoleic acid is regarded as the most crucial of the omega-6 fatty acids due to its ability to be acquired with other acids in this category,

such as α-linolenic acid or γ-linoleic acid (Zielińska *et al.,* 2014). Linoleic acid has both anti-inflammatory and anti-asthmatic properties (Reifen *et al.,* 2015, Wendell *et al.,* 2014). Arachidic acid and its metabolites have significant involvement in several biological processes, such as transmission of chemical or physical signal through cell as a series of molecular events, contraction, guided movement of a cell in response to a chemical stimulus, cell proliferation and mortality (Zielińska *et al.,* 2014). Linoleic and α-linolenic are two principal polyunsaturated fatty acids (PUFAs). The ratio of PUFAs in the phospholipids found in both serum and erythrocytes is a crucial factor that affects both health and illness. This ratio is influenced by genetic variations and food intake which regulate the endogenous metabolism of PUFAs. PUFAs may provide cardiovascular protection due to being reducer of inflammation, inhibitor or preventer of cardiac arrhythmia, lipid-lowering agents and able to lessen high blood pressure level (Ristić-Medić *et al.,* 2013).

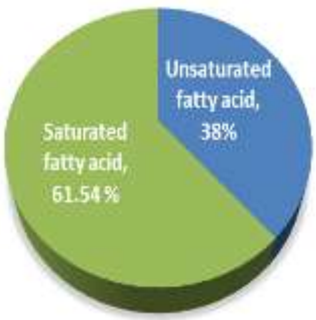


Fig. 2. Saturation and unsaturation portion of fatty acid from PE extract of the aerial part of *M. koenigii*

Therefore, it may be inferred that the plant researched here has a significant amount of fatty acids in its aerial part. Consequently it may be considered a promising source of valuable medications. The substantial abundance of these fatty acids may indicate the prospective pharmacological significance of *M. koenigii* in disease control. The healing efficacy of plants resides in specific chemical compounds that have a distinct physiological effect on the human body. Conclusively we propose that more research may intensely be focused to separate, refine, and analyze the biologically active substances found in *M. koenigii* L. to acquire valuable chemotherapeutic drugs.

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