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Short Communication

Phytochemical screening and chemical composition of fixed oil from seed of *Abroma augusta*

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Abstract

Abroma augusta is one of the most important medicinal plant under Sterculiaceae family. All the parts of this plant used as folk medicine from ancient time for various treatments. The present study attempts to evaluate the phytochemicals and fatty acids composition from seeds of Abroma augusta. The presence of some phytochemicals like alkaloids, flavonoids and saponins strongly support the medicinal use of the plant. The compositions of fatty acids from the seed extract of Abroma augusta were identified by Gas Chromatography-Mass Spectrophotometer. Four fatty acids were found and they are methyl palmitate, methyl stearate, oleic acid and methyl linoleatte. Methyl linoleatte showed the highest concentration (72.61%) among all the fatty acids found in the seed of Abroma augusta.

Keywords: *Abroma augusta,* phytochemicals, fatty acids, Gas Chromatography-Mass Spectrophotometer.

Introduction

Nowadays, the researchers have been shown their interest in the use of medicinal plants as herbal medicines in developing countries. The herbal medicines are safe and free from any adverse side effects compared to synthetic drugs. Thus a search for new drugs with better and cheaper substitutes from plant origin is a natural choice. Some chemical constituents of these plants are accountable for curative values and produce a specific physiological action on the human body[1-2]. Our civilization generally differentiated plant by their food manufacturing ability, the presence of cell walls, and their unlimited type of growth. Generally, the green plants give the basic food to all organisms. Plants are continuously liberating the oxygen gas during food processing. This oxygen, which we obtained from air as we breathe in, is essential to life. Plant is the only source of food and oxygen, no animal or man alone can supply these[3]. Extraction and characterization of several phyto-chemicals from these plants have given some vital profile drugs[4]. Whereas, it is our big concern that currently many medicinally important plants loss their genetic diversity due to the increase market and public demand[5].

Abroma augusta commonly known as Ulatkombal in Bangali and Devil´s cotton in English. This plant is usually found in in tropical Asia, South and eastern Africa, and Australia. It has various uses in the traditional medicinal system such as Ayrveda and Unani[6-8]. Ulatkambal is described in unani literature for treating diabetes, stomache, dermatitis, leucorrhoea, scabies, gonorrhea, cough, leukoderma, jaundice, nerve stimulant, weakness, hypertension, uterine disorders, rheumatic pain of joints and headache with sinusitis. In ethanobotanical literature this plant is mentioned to be effective of dermatitis, anti-inflammatory and analgesics. All plant parts are used for medicinal purposes in Bangladesh, India or subcontinent for thousands of years[9].

Previously different parts of *A. augusta* have been studied for the isolation of pharmacologically active compounds[10] but no methodical study has been done for screening of phytochemicals and fatty acids present in the seeds of this plant. By considering the vast application of all the parts of *A. augusta* in ayurvedic preparation and traditional medicine, quantification of some phytoconstituents i.e. saponin, alkaloid and flavonoid content of seeds and fatty acid composition was analyzed by Gas Chromatography-Mass Spectrophotometer.

Materials and methods

Plant sample: Completely seasoned fresh seeds of *Abroma augusta* were collected on December, 2018 from the campus of Bangladesh council of scientific and industrial research, Dhaka. A taxonomist was identified the plant sample and a voucher specimen (No. 43071) has been deposited in Bangladesh National Herbarium, Dhaka.

Reagents: All reagents used were purchased from Sigma-Aldrich (Buchs, Switzerland) or Merck (Darmstadt, Germany). Analytical reagent grade pet-ether (b.p. 40-60^{o}C, Merck, Germany) was used for the extraction of plant compounds under normal atmospheric pressure.

Solvent was recovered from extract by distillation process and the dried extracts were stored in a sealed vial at 4°C until further analysis.

Preparation of methyl ester and extraction of fatty acids: The soil particles and other dust were removed from seeds of *Abroma augusta* by washing individually under running tap water. After that they were dried and grind to powder form by mortar grinder. The extraction of fatty acids was done from the seed (100gm) by using a soxhlet apparatus for 72 hours. Then the solvents were evaporated by using a rotary evaporator under reduced pressure. Filtration of extracts was carried out by using filter paper (Whatman no.1). Then the extact was vacuum distilled for completely removing the solvent. The weight of seed extracts was 6.38gm (6.38% w/w). The dried samples were kept in a refrigerator under nitrogenous atmosphere. At first the fatty acids were transferred to Fatty Acid Methyl Esters (FAMEs) for GC-MS analysis and analyzed according to the Griffin method[11].

The composition of fatty acids was identified by their methyl esters analysis. The FAMEs were analyzed by using BF_3-MeOH complex as stated in AOAC method[12]. 10mg of extract was mixed with 1ml of BF_3-MeOH complex in a screw capped glass tube and then heated in a water bath at 100°C for 1 hour. After cooling the mixture at room temperature, hexane (2ml) and deionized water (1ml) were added. The glass tube was violently shaken and centrifuged for two minutes at low RPM. We collect the top layer in a glass vial by using syringe and kept in refrigerator until further analysis.

Gas Chromatography-Mass Spectroscopy: GC-MS scanning of *A. augusta* seeds fatty acids were done with a mass spectrophotometer detector which was equipped on a Agilent 7890A system. The gas chromatograph connected with a HP-5MS capillary column (30m×0.25mm) of 0.25µm film thickness. The initial oven temperature was 70°C and then raised to 150°C at the rate of 10°C/min for 5 min. It was again increased to 200°C at 12°C/min for 15 min, finally 12°C/min to 220°C for 15 min and injector temperature was 260°C. Helium was used as carrier gas with flow rate 0.6ml/min at 17.69 psi pressure. 1µl sample was injected automatically after dissolving in methanol. Electron ionization was used. The mass spectrum was put in scan mode between 50-550 m/z range. Mass spectra of fatty acid compositions were confirmed by using NIST libraries.

Alkaloid Determination[13]: 5gms of powdered seed were taken into a 250ml conical flasks and 10% acetic acid (200ml) were added in ethanol, coated by aluminum foil and kept for two days before filter. The extract was concentrated after filtration to one fourth of actual weight by using water bath. Concentrate NH_4OH was mixed to the reduced volume of extract drop by drop until the precipitation was carried out. All the solution was allowed to settle and the precipitate was taken out by filtration. It was then dried and weighed.

Saponin Determination[14]: 5gms of powdered seed sample were mixed with 250ml of 25% ethanol into a 250ml conical flask. Then the suspension was heated with continuous stirring on a water bath at 60°C for four hours. After filtering the solution, the residue was again used with another 20% ethanol (200ml) and filtered again. All the filtrate were merged together and reduced the filtrate to 40ml using water bath at 90°C. The abated mixture was mixed with 20ml of diethyl ether into a 250 ml separating funnel and shaken strenuously. The ether layer was discarded while the aqueous layer was collected into 250ml conical flask. The purification steps were repeated three times and then mixed with n-butanol (60ml). Then the extracts were rinsed with 5% NaCl solution (10ml). The rest of the solution was kept for heating. The sample was dried by evaporation in the oven until a fixed weight. At last the percentage of saponin content was determined.

Flavonoid Determination[15]: 5gms of powdered seed sample with 150ml of 80% aqueous methanol were taken into 250ml conical flasks at room temperature. All the mixture was filtered by using Whatmann filter paper 42. Finally, the filtrate was dried and weighed.

Results and discussion

Fatty acid analysis: The constituents of the fatty acids of *Abroma augusta* seeds found from GC-MS analysis including their molecular weight, molecular formula and percent composition are presented in Table-1 and structural formula are presented in Figure-1. The compounds were present as methyl stearate (3.14%), methyl palmitate (14.67%), oleic acid (9.58), methyl linoleatte (72.61%).

Table-1: GC-MS analysis of fatty acids from pet-ether extract of *Abroma augusta* seeds.

Name of fatty acids	Molecular weight	Molecular formula	Conc. (%)
Methyl Stearate	298.504	$C_{19}H_{38}O_2$	3.14
Methyl Palmitate	270.45	$C_{17}H_{34}O_2$	14.67
Oleic Acid	282.468	$C_{18}H_{34}O_2$	9.58
Methyl Linoleatte	294.479	$C_{19}H_{34}O_2$	72.61

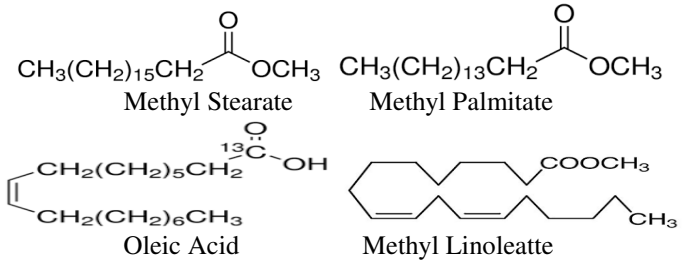

Figure-1: Structure of identified fatty acids from pet-ether extract of *Abroma augusta* seeds.

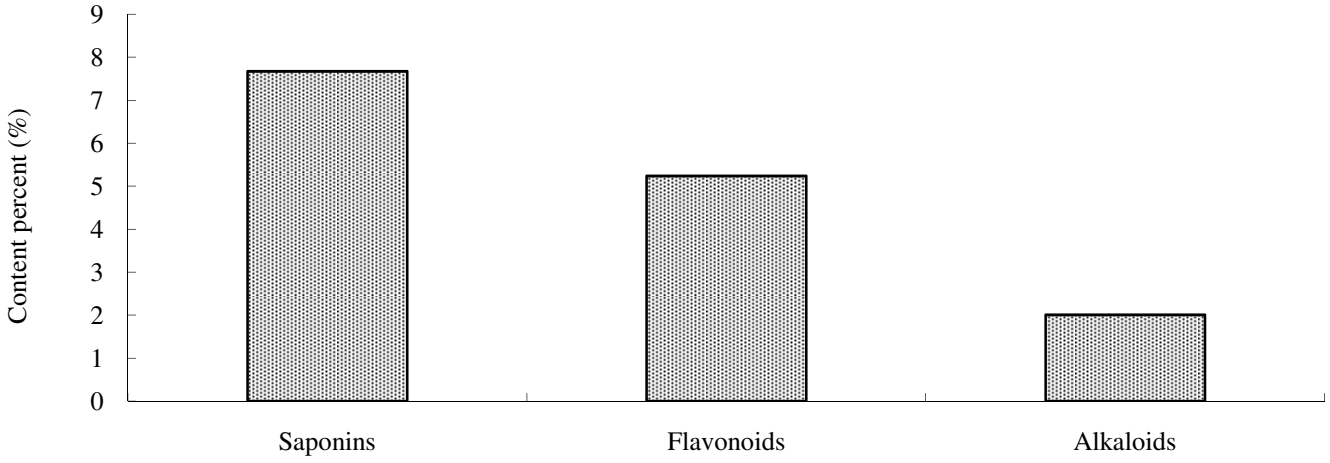

Figure-2: Phytochemical contents of *Abroma augusta.*

Phytochemical Content: The present study showed that seeds of *Abroma augusta* contain phytochemical such as flavanoids, alkaloids and saponins in appreciable quantities (Figure-2).

It was found that seeds of this medicinal plant showed higher saponin content (7.67%), flavanoid content is (5.24%) and alkaloid content is (2.01%). Saponins with detergent like properties are producing by various plant parts can help to resist microbial pathogens[16]. Saponins and alkaloids protect excessive intestinal absorption of cholesterol and decrease the risk of cardiovascular diseases such as hypertension[17]. In medicine, saponins are also used as anti-cancer, anti-inflammatory and weight loss etc[18]. Flavonoid has been used as nature's biological response modifiers because of strong authentication of their inherent capacity to convert the body's reaction to allergen, virus and carcinogens. Flavonoids contain polyphenolic compounds with identical properties including inhibition of hydrolytic and oxidative enzymes, anti-inflammatory and free radical scavenging activity[19]. Some flavonoids have also been reported to act like coumarins in the retardation of giant-cell tumor formation in HIV-infected cell cultures[20]. The existences of these secondary metabolites in the plant part indicate that it might have various medicinal and industrial importances.

Conclusion

In this study, we found four constituents from seeds of Abroma augusta by GC-MS analysis. The phytochemicals present in seeds of this plant justified the extensive applications to treat various diseases from ancient time by conventional practitioner. However, further research is necessary to evaluate its toxic profile and bioactivity.

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