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Studies on Fatty Acids Composition and Some Valuable Nutrients of *Piper nigrum* Linn. (Gol Morich)

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Abstract

This study deals with physical and chemical characteristics and some nutritional properties of the fatty oil extracted from *Piper nigrum* Linn. (Black Pepper). The seeds of Black Pepper contained fatty acids 12.02% of which the saturated and unsaturated fatty acids contributed 68.71% and 31.29%, respectively. The fatty acids identified by GLC are lauric acid (26.93%), myristic (8.26%), palmitoleic (13.47%), palmitic (12.24%), oleic (17.82%), stearic (17.28%) and lignoceric acid (4.00%). The specific gravity and refractive index were recorded as 0.924 at 30°C and 1.461 at 26°C, respectively. The saponification value, iodine value, peroxide value, acid value and percentage of unsaponifiable matter were found to be 177.92, 15.86, 472.97, 8.27 and 43.28, respectively. The moisture content (2.17%), dry matter (97.83%), ash (12.49%), nitrogen (1.85), protein (11.56%), carbohydrate (45.16%), crude fiber (16.66%) and food energy (335.06 g cal^{-1}) showed that *Black pepper* seeds are good source of dietary fiber.

Keywords: Black Pepper, fatty acid composition, saturated fatty acids, lauric acid, Gas Liquid Chromatography.

I. Introduction

Black pepper (*Piper nigrum* Linn.) is one of the oldest and best known spices in the world belonging to *Piperaceae* family. It is cultivated in damp nutrient rich soil of South India and also found in Indonesia, Malaysia, and Brazil. Dried unripe fruits usually known as Peppercorns[1]. In India it is called *Kali mirchi* and in Bangladesh it is called Gol Morich, which is a common household spice. The plant is originally a plant of forest and this climbing woody vine uses trees or other supports to grow to about twenty feet high, but is normally kept to about 12 feet for commercial purposes and has a life span of about twenty years. The plant is widely cultivated in the tropics as the source of black and white pepper. Black Pepper is obtained from the dried unripe fruit [1].

Black pepper can be used to help in the treatment of pain relief, rheumatism, chills, flu, colds, increase circulation, exhaustion, muscular aches, physical and emotional coldness, nerve tonic and fevers. It furthermore increases the flow of saliva, stimulates appetite, encourages peristalsis, tones the colon muscles and is a general digestive tonic. Sometimes it is used in place of cubebs for gonorrhea. Externally it is used for its rubefacient properties and as a local application for relaxed sore throat and some skin diseases. Its oleoresin has bacteriostatic and fungistatic properties [1-3].

Lipids are structural materials, reserve supply of fuels, vitamins, emulsifiers, flavours, aromatic compounds and barriers to the environment [4, 5]. Vegetables and plant seeds are the most promising sources of lipids, simple and complex carbohydrates, proteins, minerals and secondary metabolites. They also play a significant role in normal biological functions and in relation to major problems in health and agriculture[7-9]. These are used for better health of human also for industrial and as well as pharmaceutical purposes [6].

With the increasing demand of vegetable oils and spice in the country, the investigation of the seed oil of Black Pepper was undertaken to (a) estimate the chemical composition of the seed oil as well as that of the extracted oil from the remaining seed cake of Black Pepper, (b) compare the quality and quantity of fatty acids with other oils of commercial seed and explore the possibilities of proper utilization.

II. Materials and Methods

Plant materials

Black Pepper sample was collected from Dhaka New Market. The sample was cleaned to separate dirt, sun-dried and followed by the determination of the moisture content and then steam-distilled to make it free from essential oil and again sun-dried.

Extraction

The essential oil free dried samples were powdered by a warring blender. Crushed Black Pepper seed-cake (100.0 g) was then subjected to extraction with n-hexen (b.p. 68°C) in Soxhlet apparatus for 72 hours to isolate the oil with three replications. In each experiment, the mass filtered and filtrate was vacuum distilled to remove solvent completely. The yield of oils were calculated and stored in a refrigerator for further analyses using AOAC methods [10].

Physico-chemical study of the oil

The physico-chemical properties of extracted oil were determined with three replications using cited standard methods [10-13] and the results are given in the Table 1.

Nutritional analysis

The meal left after oil extraction was sun-dried and it was used for determination of the percentages of moisture, ash, nitrogen, protein, carbohydrate and crude fiber according to the standard methods [10-14] with three replications (presented in Table 3). The gross food energy was estimated using the equation[7, 15, 16]: FE = (% CP×4) + (% Lipids ×9) + (%CHO×4)

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Where: FE = Food energy (in $gcal^{-1}$), CP=Crude protein and CHO=Carbohydrate.

Identification and quantification of fatty acids

The fatty acid contents were determined by GLC as their methyl esters. The fatty acid methyl esters (FAMEs) were prepared by complete esterification (checked by TLC) of oil using BF_3-MeOH complex[7, 10, 17, 18].

Standard FAMEs (E. Merck) were used for the identification and quantification of the peaks the results are given in the Table 2.

Instrument and separation conditions

The FAMEs were analyzed by a PYE UNICUM PU: 4500U Gas Chromatograph (England) fitted with a flame ionization detector (FID) and an electronic integrator equipped with SE-54 quartz capillary column (30 m × 0.25 mm *i .d.* and 0.25µm film thickness). Carrier gas nitrogen (N_2) at a flow rate of 3 mL/min. The separation was affected at 100°C - 220°C. The following temperature program carried out during GC analysis: initial oven temperature 100°C, increases at a rate of 4°C min^{-1} to 220°C for 30 min. The oven, injection and detection temperatures were fixed at 100°, 220° and 230°C respectively. Splitting 80%, speed of the chromatogram was 0.5 mm/min. The fatty acids were identified by comparison of relative retention times and peak positions of the chromatogram with that for the standard fatty FAMEs. The amounts of fatty acids were calculated from the peak areas computed by LKB 2220 electronic recording integrator.

III. Results and Discussion

The Black Pepper collected from local sources were grinded to powder and the oil was extracted. The extract was found to contain 12.02% fatty oil. The oil was opaque, viscous almost semi-solid and dark green in colored with mild unpleasant but not pungent taste and have characteristics odor of pepper. It was freely miscible in chloroform, carbon tetrachloride, petroleum ether, n-hexane, diethyl ether and in alcohol upon warming but immiscible with water.

The refractive index of the oil was found to be 1.461 at 30°C and this is comparable with soybean oil (1.466-1.470) and rice bran oil (1.470-1.473) [6, 19, 20]. The specific gravity of the oil was found to be 0.924 at 30°C, which indicated that the oil contained high molecular weight of fatty acids. It is also closer to soybean oil (0.919-0.923), mustard oil (0.914-0.923) and sunflower oil (0.923-0.928) [6, 19, 20].

Saponification value (Table 1) was found to be 177.92 mg KOH g^{-1} which value is quite similar to the range of cod liver oil (171-190), white mustard oil (171-174) and rape seed oil (168-179) [6, 19, 20]. The extracted oil had the iodine value 15.86 (Table 1), the result of iodine value indicates the oil contained mostly saturated fatty acids.

Table 1. Chemical characteristics of Black pepper (*Piper nigrum* Linn.) fatty oil

Characteristics	Result
Acid value	8.27 ± 0.56
Iodine value	15.86 ± 0.82
Saponification value	177.92 ± 0.46
Unsaponified matter (%)	43.28 ± 0.79
Peroxide value	472.97 ± 0.23

Each value represents the average of three replicate analyses ± SD

The acid value 8.27 indicates that the proportion of free fatty acid content was higher than those in edible oils like soybean oil (0.38 - 0.54), mustard oil (3.65 - 4.56) and palm oil (0.17 - 1.06) [6, 19-22]. A higher percentage of free fatty acid (above 1.5%) is an indication of unsuitability of the oil for edible purpose. So the oil of Black Pepper might be not suitable for edible purposes. It may be used for consumption through refining or may be used for industrial purposes. The drying property of the oil was examined and it was found to be non-drying in nature.

Unsaponifiable matter (43.28) was higher than those in any edible or non-edible oils like soybean oil (0.015%), rapeseed oil (0.02%), sesame seed oil (0.02%) and palm oil (0.012%) [6, 21, 22]. The oil is supposed to be contaminated with mineral oil, higher aliphatic alcohols, sterols, pigments, hydrocarbons and sufficient amount of tocopherols [6, 21, 23, 24]. Tocopherols play important role as antioxidant [23, 24].

Peroxide value was found 472.97 which showed that the oil had much free active oxygen enabling autoxidation of the oil [6, 21]. Gas liquid chromatographic analysis (presented in Table 2) of the fatty acid showed that the unsaturated fatty acids present in the oil sample was mainly oleic acid (17.82%) and palmitoleic (13.47%) which contained 31.29% of the total oil.

Table 2. Fatty acid composition (in wt %) of Black Pepper (*Piper nigrum* Linn.) seed determined by GLC

Name of the fatty acids	Retention time of the standard acids	Retention time of the experimental acids	Peak Area	Relative percentage of fatty acids (%)
Lauric acid $_{12.0}$	5.98	5.72	4176	26.93 ± 0.23
Myristic$_{14.0}$	9.11	8.58	1253	8.26 ± 0.56
palmitoleic $_{16.1}$	11.92	10.53	2044	13.47 ± 0.24
Palmitic acid$_{16.0}$	12.32	12.24	1857	12.24 ± 0.28
Oleic acid$_{18.1}$	14.95	14.91	2856	17.82 ± 0.42
Stearic acid$_{18.0}$	15.30	15.22	2773	17.28 ± 0.33
Lignoceric acid $_{24.0}$	24.21	24.83	1213	4.00 ± 0.52

Each value represents the average of three replicate analyses ± SD

The saturated fatty acids present in the oil sample was mainly myristic acid (8.26%), lauric acid (26.93%), palmitic acid (12.24%), stearic acid (17.28%) and lignoceric acid (4.00%) which altogether accounted for about 68.71% saturated FA of the total fatty acid. The proportion of palmitic acid and oleic acid are comparable to the same in soya bean oil.

From Table 3, it appears that the moisture, dry matter, crude fiber, protein, ash and total carbohydrate are 2.17%, 97.83%, 16.6%, 11.56%, 12.49% and 45.16%, respectively. The Black Pepper has fairly high food energy value 335.06 (g cal^{-1}) which might be due to the high lipid content (12.02%) and high carbohydrate content (45.16%). The high proportion of carbohydrate, protein as well as crude fiber is noticeable characteristics of the seed. The high fiber content (16.6%) of Black Pepper seed may possess positive effect in human diet. Besides, it may increase feacal bulk and lowers gastric cholesterol [16, 25].

Table 3. Nutritive and proximate composition of the Black Pepper (*Piper nigrum* Linn.) seed

Parameters	Result
Moisture content	2.17 ± 0.56%
Dry matter content	97.83 ± 0.53%
Ash content	12.49 ± 0.23%
Nitrogen content	1.85 ± 0.49
Protein content	11.56 ± 0.56%
Carbohydrate content	45.16 ± 0.89%
Crude fiber content	16.66 ± 0.83%
Food energy	335.06 ± 0.89 ($gcal^{-1}$)

Each value represents the average of three replicate analyses ± SD

IV. Conclusion

From the foregoing findings it appears that the Black pepper is a good source of protein, carbohydrate and crude fiber. The fatty oil of this species is rich in saturated acid, and it is not suitable for edible purposes but it is a good source of lauric acid.

So in fine it may be concluded that although the oil of Black pepper is not suitable for human health[26, 27] but its nutritive value may have positive effect in human diet.

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