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Studies on fatty acid profile, physico-chemical properties and phyto-esterol contents of bitter gourd (Momordica charantia) seed oil

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Abstract

Present work have been undertaken to know the physico-chemical qualities of dried bitter gourd (*Momordica charantia*) seed oil. Bitter gourd seed oil possessed antibacterial, anti-diabetic, anti-fungal, anti-tumour etc properties, for these reasons bitter gourd seed oil gained great importance in pharmaceuticals and ayurvedic medicine. In present investigation the fresh bitter gourd seed oil was analyzed for physico-chemical properties, fatty acid composition and phyto-esterol contents. It was observed that bitter gourd seed oil possessed 64.27 \pm 3.77 percent of poly unsaturated fatty acid. The physico-chemical properties of bitter gourd seed oil showed that, bitter gourd seed oil contain 145.22 \pm 2.20 mg KOH/g value for saponification value, 111.00 \pm 1.20 gI₂ for iodine value and 4.02 \pm 0.12 mg KOH/g for acid value. The phytoesterol contents of bitter gourd seed oil contained campesterol, stigmasterol and beta sitosterol in good proportion. From the research it was concluded that bitter gourd seed oil possessed good fatty acid profile and phytoesterol contents.

Keywords: bitter gourd seed oil, Fatty acid profile, phytoesterol and Poly unsaturated fatty acid

1. Introduction

Momordica charantia Linn. (Bitter gourd) commonly known as Bitter melon or Bitter gourd is tropical and subtropical climber of the family Cucurbitaceae. The Bitter gourd is believed to be originated in the tropics of the old world. It is widely grown in India and other parts of the Indian subcontinent, Southeast Asia, China, Africa, and the Caribbean and South America as a food and medicine (Kumar 2010)^[7]. Bitter gourd is an annual or perennial climber found throughout India and also cultivated up to an altitude of 1500 m. It is cultivated during warm season i.e. during April to July by sowing seeds in a pit. Seeds are sown at a distance of half a meter and provided with manures. Only one plant is retained and plant seedlings are watered once or twice a week. Plants begin to flower 30-35 days after sowing and fruits are ready for harvesting after flowering 15-20 days.

The fruits of bitter melon are utilized as vegetable where as the whole plant parts like, fruits, leaves, roots and seeds of bitter melon as medicine. *Momordica charantia* Linn. (Bitter gourd), a vegetable/ medicinal plant is used in the Ayurvedic system of medicine for treating various diseases including diabetes mellitus, measles, fever, hepatitis, itch etc.

Bitter gourd has been used in various Asian traditional medicine systems for a long time, as useful for preventing and treating various diseases. Fruits of *Momordica charantia* (bitter gourd) used in asthma, burning sensation, colic, constipation, cough, diabetes, fever (malaria), gout, helminthiases, inflammation, leprosy, skin diseases, ulcer and wound. It has also been shown to have hypoglycaemic properties (antidiabetic) in animal as well as human studies. Juice of the Bitter gourd leaves used to treat piles completely. Bitter gourd is used as a blood purifier due to its bitter tonic properties. Boils and other blood related problems that show up on the skin. Juice of bitter gourd is also beneficial in treating and preventing the liver damage (Garau *et al.*, 2003) ^[6].

Bitter gourd seed and its oil are used in the treatment of ulcers, liver and spleen problems, diabetes, intestinal parasites, high cholesterol, and intestinal gas, heal wounds and stomachache etc. Roots are used in the treatment of syphilis, rheumatism, boils, ulcer, septic swellings, opthelmia, and in Prolapsus vagenae. Bitter gourd juice helps to reduce the problem of Pyorrhea (bleeding from the gums). Bitter gourd capsules and tinctures are widely available in the United States for the treatment of diabetes, viruses, colds flu, cancer, tumors, high cholesterol and psoriasis.

In India, *Momordica charantia* Linn. (Bitter gourd) used by tribal people for abortions, birth control, increasing milk flow, menstrual disorders, vaginal discharge, constipation, food, diabetes, hyperglycemia, jaundice, stones, kidney, liver, fever (malaria), gout, eczema, fat loss, hemorrhoids, hydrophobia, intestinal parasites, skin, leprosy, pneumonia, psoriasis, rheumatism, scabies, snakebite, vegetables, piles, tonic, anthelmintic, purgative. By considering all the facts related to nutritional quality of bitter gourd and its seed oil, the present investigation was formulated to characterize the fatty acid profile and phyto-esterol contents of bitter gourd seed oil.

Material and Methods

Methodology for preparation of bitter gourd seed oil

Fresh, disease free and sound quality bitter gourd were selected. The fresh bitter gourd were washed with clean water and subjected to the treatments like cutting followed by separation of seeds. The dried bitter gourds seeds were grinded to make powder and from seed powder seed oil was extracted.

Fatty acid profile of bitter gourd seed oil

Fatty acids in bitter gourd seed oil were converted into methyl esters using NaOCH3 in methanol as described in Christie and Sédédio. Samples were then injected into a Shimadzu GC-2010 chromatograph equipped with a flame ionization detector and a fused silica column (SP-2560, 100×0.25 mm). Samples were run under chromatographic conditions described in Baublits *et al* (2007) ^[3]. Fatty acids were identified by comparing their retention times with those of commercial standards (C4-C24 methyl esters, Sigma 18919). Conjugated fatty acids were identified by comparing their retention times with those reported in the literature. Results were expressed as percentages of total fatty acids.

Physico-chemical analysis of bitter gourd seed oil Refractive Index

The Abb's refractometer was used for the determination of refractive index. The refractive index is denoted by nD 25" where n is the refractive index at 25° C taken with sodium light (D-line). First, the refractometer was standardized with distilled water which has refractive index of nD 29.5 = 1.3315. Then it was cleaned with acetone and dried with cotton. After this, a drop of bitter gourd seed oil was placed between the prisms of refractometer. The telescope was rotated to bring the border line of total refraction to the junction of cross-wire in the telescope. The refractive index was recorded at room temperature (Njoku and Evbuomwan, 2014) ^[9].

Specific gravity

Density bottle was used for determining the density of the oil. A clean and dry bottle of 25 ml capacity was weighed (W_0) and then filled with the bitter gourd oil, stopper inserted and reweighed to give (W_1). The oil was substituted with water after washing and drying the bottle and weighed to give (W_2). The specific gravity is expressed in terms of ratio of oil to water (Njoku and Evbuomwan, 2014) ^[9].

Saponification value

Bitter gourd seed oil 5.3716 g was weighed into a conical flask separately. 25 ml of 0.1N ethanolic potassium hydroxide was added to the conical flask and content constantly stirred for 1 hr followed by reflux. Phenolphthalein indicator was

then added to the conical flask and titrated with 0.5M HCl till the solution changes to colorless. The same procedure was repeated for the blank (Njoku and Evbuomwan, 2014)^[9].

Iodine value

Iodine value of bitter gourd seed oil was measured with slight modification as per method Njoku and Evbuomwan, (2014)^[9]. 0.2 g of oil was taken in 500 ml conical flask. This 20 ml of chloroform was added and oil was completely dissolved. This solution was kept in dark for 30 min. To this, 20 ml of KI solution was added and was mixed well. Titration was done against 0.1N Na₂S₂O₃ solution using starch as an indicator with vigorous shaking to extract iodine from the chloroform layer. Blank was conducted similarly in absence of oil.

Iodine number = $\frac{A \times N \times 0.1269 \times 100}{Weight of oil}$

Where, A = ml of $Na_2S_2O_3$ N =Normality of $Na_2S_2O_3$

Acid value

Acid value of bitter gourd seed oil was measured as per method given by AOAC, (2000) ^[1]. 100 ml of neutral ethyl alcohol was heated with 10 g of oil sample in a 250 ml beaker until the mixture began to boil. The heat was removed and was titrated with N/10 KOH solution, using two drops of phenolphthalein as indicator with consistent shaking for which a permanent pink color was obtained at the end point. The acid value was calculated using the expression (mg KOH/g);

Acid Value = $0.56 \times No.$ of ml. N/10 KOH used

Determination of phytosterols content of bitter gourd seed oil

Determination of phytosterols was carried out in three steps, as recommended by Almeida (2009)^[2].

(1) Heat saponification using 3 % KOH in water bath at 50 °C under stirring for 3 hrs. (2) Extraction of the unsaponifiable fraction in hexane (10 ml) under vortex mixing for 1 min.

(3) Quantification of phytosterols by gas chromatography (GC).

The system consisted of a Shimadzu GC- 2010 chromatograph equipped with a DB-5 poly (methylphenyl) siloxane column (5 % phenyl, 60 m, 0.25 mm) and a flame ionization detector. The temperature gradient was as follows: 150 °C for 0.1 min, 150–300 °C at 10 °C/min, and 300 °C for 10 min. The temperatures of the injector and detector were 250 and 300 °C, respectively. Helium (1 ml/min) was used as the carrier gas, and the split ratio (the amount of sample entering the column) was 1:50. Identification of the peaks was performed by comparing the retention times with those of Sigma standards (campesterol-C5157, stigmasterol-S6126, and β -sitosterol-S9889), and quantification was done by internal standardization using 5 α -cholestane (Sigma C8003). Results were expressed as milligrams of phytosterols per 100 g of oil.

Results and Discussion

Fatty acid profile of bitter gourd seed oil

The bitter gourd seed contained good proportion of acid which are essential for offering various health promoting and diseases preventing properties. The some essential fatty acid present in bitter gourd seed oil had cancer preventing properties. The fatty acid profile of bitter gourd seed had been

analyzed by gas chromatography. The data related to fatty acid profile of bitter gourd seed is tabulated in table 1.

| Fatty acid profile | Bitter gourd seed oil (%) | | |
|------------------------------------|---------------------------|--|--|
| Palmitic acid | 2.73±0.06 | | |
| Stearic acid | 30.11±1.12 | | |
| Oleic acid | 4.29±0.10 | | |
| Linoleic acid | 4.60±0.13 | | |
| Linolenic acid | | | |
| Elostearic acid | 55.80±3.62 | | |
| Catalpic acid | 0.69±0.01 | | |
| Saturated fatty acid | 32.15±1.22 | | |
| Mono unsaturated fatty acid | 3.58±0.17 | | |
| Poly unsaturated fatty acid (PUFA) | 64.27±3.77 | | |

Table 1: Fatty acid profile of bitter gourd seed oil

The data presented in table 1 showed the fatty acid profile of bitter gourd seed oil. The palmitic acid, stearic acid comes under category of saturated fatty acid. The palmitic acid contents of bitter gourd seed oil was 2.73 percent and stearic acid contents of bitter gourd seed oil was 30.11 percent. The similar result were obtained by Bialek *et al.* (2014) ^[4]. The oleic acid, lenoliec and lenolenic acid are categorized under unsaturated fatty acid. The oleic acid contents of bitter gourd seed oil was 4.29 percent. The bitter gourd seed oil was free from linolenic acid.

The elostrearic acid and catalpic acid contributes anticancer properties to seed oil and hence it is important in preventing the development of cancer cell. The eleostearic acid and catalpic acid content of bitter gourd seed oil were 55.80 and 0.69 percent respectively. The saturated fatty acid, mono saturated fatty acid and poly unsaturated fatty acid (PUFA) contents of bitter gourd seed oil were 32.15, 3.58 and 64.27 percent respectively. The similar results were obtained by Yoshime *et al.* (2016) ^[11].

Physico-chemical properties of bitter gourd seed oil

The bitter gourd seed oil was obtained by soxlet method. The physical properties of bitter gourd seed oil were determined and results were presented in Table 2.

| Characteristics | Observations |
|--------------------------------------|-------------------|
| L^* | 88.76±0.20 |
| <i>a</i> * | -1.97±0.005 |
| b^* | 5.24±0.10 |
| Refractive Index (20 ⁰ C) | 1.44 ± 0.001 |
| Specific gravity | 0.85 ± 0.005 |
| Saponification value (mg KOH/g) | 145.22 ± 2.20 |
| Iodine value (gI ₂ /100g) | 111.00 ± 1.20 |
| Acid value (mg KOH/g) | 4.02 ± 0.12 |

 Table 2: Physico-chemical properties of bitter gourd seed oil

The data presented in table 2 showed the physico-chemical properties of bitter gourd seed oil. The oil from bitter gourd seed was extracted by soxlet method has color values L^* (88.76±0.20), a^* (-1.97±0.005) and b^* (5.24±0.10). The color of oil was light yellow to light green. The refractive index and specific gravity of lemongrass oil was found to be 1.44 ± 0.001 and 0.85 ± 0.005, respectively. The results are in agreement with the findings of Vázquez-Briones *et al.* (2015)

^[10]. The saponification value, acid and iodine value of oil were $145.22 \pm 2.20 \text{ mg KOH/g}$, $4.02 \pm 0.12 \text{ mg KOH/g}$ and $111.00 \pm 1.20 \text{ gI}_2/100\text{g}$, respectively which are similar with Dutta *et al.* (2014) ^[5].

Phytosterol contents of bitter gourd seed oil

The data related to phytoseterol contents of bitter gourd seed oil is analyzed and tabulated in table 3.

| Table 3: Phytoesterol cont | ents of bitter | gourd s | eed oil | |
|----------------------------|----------------|---------|---------|--|
| | - | - | / | |

| Phytoesterol | Bitter gourd seed oil (mg/100g) |
|-----------------|---------------------------------|
| Campesterol | 56.34 |
| Stigmasterol | 53.80 |
| Beta sitosterol | 271.73 |

*Each value is average of three determinations

The data presented in table 3 showed the phytosterol contents of bitter gourd seed oil. It was observed that campesterol content of bitter gourd seed oil was 56.34 mg/100g. Phytosterols are present in all vegetable oils and account for the largest group of compounds in their unsaponifiable fraction. The phytosterol profile is characteristic of each type of oil (Mancini-filho *et al.*, 2015) ^[8]. The stigmasterol

contents present in bitter gourd seed oil was 53.80 mg/100g whereas beta sitosterol contents of bitter gourd seed oil was 271.73 mg/100g. The similar results were obtained by research outcome of Yoshime *et al.* (2016) ^[11].

Conclusion

From the present investigation it is concluded that bitter gourd

seed oil possessed good nutritional as well as medicinal value. The bitter gourd seed oil is good source of poly unsaturated fatty acid as well as contained good quantity of phytoesterol.

References

- 1. AOAC. Approved Methods of the American Association of Cereal Chemists, 10th Edition 2000.
- Almeida CAS. Avaliação dos principais fitosteróis em óleos vegetais eazeite. (Dissertation) Universidade Estadual de Campinas 2009, 89p.
- 3. Baublits RT, Pohlman FW, Brown AH, Johnson ZB, Proctor A, Sawyer J. Injection of conjugated linoleic acid into beef strip loins. Meat Science 2007;75(1):84-93.
- 4. Bialek A, Jelinska M, Bamburowicz M, Tokarz A. Effect of bitter melon aqueous extract and pomegranate oil on glucose concentration and lipid profile in blood of ratspreliminary study effect of bitter melon aqueous extract and pomegranate oil on glucose concentration and lipid profile in blood of rats. International Journal of Cardiology and lipidology Research 2014;1(1):1-7.
- 5. Dutta D, Kumar P, Nath A, Verma N, Gangwar B. Qualities of lemongrass (*Cymbopogan citratus*) essential oil at different drying conditions. International Journal of Agriculture, Environment and Biotechnology 2014b;7(4):903-909.
- 6. Garau C, Cummings E, Phoenix DA, Singh J. Beneficial effect and mechanism of action of *Momordica charantia* in the treatment of diabetes mellitus a mini review. International Journal of Diabetic Metabolism 2003;11(1):46-55.
- Kumar DS, Sharathnath KV, Yogeswaran P, Harani A, Sudhakar K, Sudha P *et al.* A medicinal potency of *Momordica charantia*. International Journal of Pharmaceutical Science and Review Research 2010;1(2):95-99.
- 8. Mancini-filho J, Takemoto E, Aued-Pimentel S, Parametros de, Identidade E, Qualidade de *et al.* Almeida-Muradian LB, Penteado MDVC, editors 2015;6:63–82.
- 9. Njoku VI, Evbuomwan BO. Quantitative and qualitative analysis, and comparative study of essential oil extracted from Nigerian orange, lemon and lime peels. International Journal of Applied Science and Engineering Research 2014;3(2):520-531.
- 10. Vázquez-Briones MC, Hernández LR, Guerrero-Beltrán JA. Physicochemical and antioxidant properties of *cymbopogon citratus* essential oil. Journal of Food Research 2015;4(3):36-45.
- Yoshime L, Melo I, Gasparotto J, Bonificia E, Carvalho T, Jorge M. Bitter gourd seed oil as a naturally rich source of bioactive compounds for nutraceutical purpose. Nutrite 2016;41(12):01-07.