

Nutritional composition and oil fatty acids of Indian winter melon *Benincasa hispida* (Thunb.) seeds

¹*Rayees, B., ²Dorcus, M. and ²Chitra, S.

¹Department of LSTF, Central Food Technological Research Institute Mysore-570020, India.

²Department of Food Process Engineering, SHIATS Allahabad- 211007, India.

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Abstract

Winter melon (*Benincasa hispida*), locally known as Petha, is a vegetable crop with high functional properties (especially in medicinal treatment), were analyzed for nutritive parameters (dietary fiber, crude protein, crude fat, crude fiber, and ash) and oil fatty acids composition. Proximate analysis showed that the winter melon seeds have crude protein, crude fat, crude fiber and ash content of 16.57%, 34.12%, 21.53% and 2.7% respectively. The extracted winter melon seed oil mainly consisted of linoleic acid (C18:2 ω-6), accounting for 80.07% of the total fatty acids. Other important fatty acid detected were palmitic (C16:0), oleic (C18:1) and stearic (C18:0) acids with contribution of 11.71%, 4.10% and 3.74% respectively. The fatty acids profile of the tested winter melon seed oil was quite comparable to those of previously reported cucurbitaceae seed oils and some other wholesome oils, revealing that it can be used as a potential seed oil crop.

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Introduction

Cucurbitaceae (Cucurbit) is an important family comprising one of the most genetically diverse groups of food plants. Most of the plants belonging to this family are frost sensitive and drought-tolerant (Whitaker and Bohn, 1950). Some important Cucurbit family members include gourd, melon, cucumber, squash and pumpkin (Robinson and Decker-Walters, 1999). These are indigenous to both arid and temperate regions of the earth and require long periods of warm, dry weather for their optimal growth (Whitaker and Davis, 1962). The fruits from *Cucurbitaceae* species are valued for nutritional and medicinal purposes (Jeffrey, 1990). The remaining portion of the Cucurbit fruits, especially the seed (often discarded as agrowaste), can be utilized for other food applications such as preservative, and also in animal feed and oil extraction, contributing to less waste disposal and value-addition.

The uses of cucurbit seeds as potential sources of oils have been reviewed by (Jacks *et al.*, 1972) who reported that the dehulled seeds contain about 50% of oil. The high content of oil, showing useful characteristics such as odorlessness, good colour and appearance, make these seeds suitable for oil industrial applications (Al-Khalifa, 1996; Mariod *et al.*, 2009). Cucurbit seed oils mainly consist of

palmitic (16:0), stearic (18:0), oleic (18:1 n-9), and linoleic (18:2 n-6 or ω-6) acids. Due to high amounts of polyunsaturated fatty acids, these oils have favorable nutritional status and beneficial physiological effects towards prevention of coronary heart disease and cancer (Yehuda *et al.*, 2005).

Benincasa hispida (Thunb.) India. (synonym; *Benincasa cerifera*) is one of the most valuable plants in Cucurbit family. It is also known as *Petha* (Hindi), *Kundur* (Malay), ash gourd or winter melon (English), *Bhuru Kolu* or *Safed Kolu* (Gujarati), *Kushmanda* (Sanskrit), *Dōngguā* (Chinese) and *Beligo* (Indonesian). Previously, (Martin, 1984) claimed that Kundur fruit seed is the best of cucurbits as the source of seed oil for the hot and humid tropics.

Thus, preliminary consideration of winter melon fruit seeds as source of vegetable oil will not only add value to these agricultural crops, nevertheless it will also increase the number of sources of vegetable oils, available. However, little work has been reported on the nutritional composition of winter melon fruit seed, and especially on fatty acids profile of its oil. Therefore, the main objective of the present study was to evaluate the nutritional composition and oil fatty acids of winter melon seeds as potential source of valuable oil for commercial applications.

*Corresponding author.
Email: bakshirayees@gmail.com

Materials and Methods

Seeds

Winter melon (*Benincasa hispida*) fruits were purchased from the local market in Rambagh, Allahabad, India. Seeds were separated manually from fruits, and fibrous materials, then washed with tap water and oven-dried at 40°C for 24 hours.

Oil extraction

The seeds were ground using a coffee grinder. The material that passed through 72-mesh sieve was used for lipid extraction purposes. The ground seed materials were extracted with 200 milliliters (ml) of petroleum ether using Soxhlet apparatus for 6 hours on water bath according to Association of Official Analytical Chemists (AOAC) Method, 948.22 (Anon, 2000). After extraction, the excess of the solvent was removed under reduced pressure using a rotary vacuum evaporator (Buchi Type, "MSW91", MAC Pvt. Ltd., Delhi). The oil was capped in a dark brown sample vial and stored below 5°C until used for further analyses.

Proximate analysis

The seeds were analyzed for crude protein, ash, crude fat and crude fiber according to the Association of Official Analytical Chemists (AOAC) method (Anon, 2000), 920.152, 930.05, 948.22, and 935.53, respectively. All analyses were carried out in triplicate. About 5 grams of the sample was taken for crude fiber and ash analyses. Percent (%) of crude fiber was equal to the weight of residue without ash divided by the weight of sample, then multiplied by 100. The ash content was determined by incinerating the sample in muffle furnace at 500°C for 6 hours. For determination of protein content, the tested sample was digested using copper catalyst and sulphuric acid, distilled and then titrated by 0.05 Normal Sulphuric acid (H₂SO₄) for nitrogen determination. The amount of protein was calculated by multiplying percentage (%) of nitrogen determined by the conversion factor 6.25 according to Kjeldhal method (Anon, 2000).

Fatty acid methyl esters (FAMES) preparation and gas chromatographic analysis

Fatty acid methyl esters (FAMES) were prepared according to International Union Of Pure and Applied Chemistry (IUPAC, 1992) standard method 2.301. winter melon seed oil were put into a screw capped test tube. 0.25 milliliters (ml) of 2 Normal sodium hydroxide (NaOH) was then added. The test tube was heated in a water bath at 60°C for 10 minutes. After heating it was cooled in a fridge. 1 milliliter of 5% methyl HCl solution was then added into the test tube

and the test tube was shaken. The test tube was again heated over water bath at 80°C for 10 minutes and then cooled again in a fridge. one milliliter of petroleum ether was added into the test tube. A pinch of sodium sulphate was also added into the test tube to remove the moisture. The upper layer from the test tube was taken and was injected into the Gas Chromatographic instrument. Record the reading from the integrator.

The analysis of FAMES was performed by Gas Chromatographic instrument (Hewlett-Packard 6890; Agilent, Wilmington, DE, USA) fitted with a flame ionization detector (FID). A fused silica capillary BPX-70 column (60 m x 0.32 mm; 0.25 µm film thickness) was used for separation. The initial column oven temperature was set at 115°C and raised to 180°C @ 8°C/min. The temperature was further raised to 240°C @ 8°C/minute and held for 10 minutes. The sample volume injected on to the capillary column was one micro-liter (µL) using splitless injection mode. Helium was used as a carrier gas at a flow rate of 1.6 milliliters /minute. The unknown Fatty acid methyl esters (FAMES) were identified by comparing their retention times with those of pure standards of Fatty acid methyl esters (FAME). The fatty acids composition was expressed as relative percentage of the total peak area.

Statistical analysis

All experiments were conducted in triplicate and the statistical significance differences of mean were calculated using Stastical Analysis System (SAS, 8.1), with the help of one-way Analysis of variance (ANOVA); results are expressed as means ± Standard Deviation (SD). A probability value at p < 0.05 was considered to denote the statistically significant differences.

Results and Discussion

Proximate composition of seeds

In the present study physicochemical properties of seeds and the extracted fatty acid composition of seed oil from fruits of Indian winter melon were analyzed and compared. The oil, protein, fiber and ash contents of tested winter melon seeds were found to be 34.12%, 16.57%, 21.53%, 2.7% respectively. The present work showed some what similar data for protein and fat content to those reported by (Anwar *et al.*, 2011). The obtained results for ash content were similar to the results given by (Sew *et al.*, 2010). Interestingly, like other cucurbit seeds, winter melon seeds in the present analysis were found to be a good source of food protein and oil. Winter melon seeds in the present study show a comparable range of oil, protein, fiber and ash content among the

other species of cucurbits. Previously, Mariod *et al.* (2009) and Achu *et al.* (2005) investigated nutritional composition of various cucurbit seeds from Sudan and Cameroon respectively and found values for protein, fiber, ash were in the range of 14.50-40.49, 3.44-36.04, and 3.41-8.33%, respectively as shown in Table 1. Cucurbits seeds from different species have been reported as an impressive source of oil with yields varying between 26.9% and 56.67%. However, the seeds of cucurbit species: *Citrullus lanatus*, *Cucumis prophetarum*, *Cucumis sativus*, *Luffa echinata*, and *Cucumis melo* from Sudan contained oil in the range of 10.9-27.10% (Mariod *et al.*, 2009) as shown in Table 1. In another study, the seeds from *Cucumeropsis mannii*, *Cucurbit maxima*, *Cucurbita moschata*, *Lagenaria siceraria* and *Cucumis sativus*, cultivated in Cameroon, showed a much higher oil content ranging from 44.85% to 53.76% (Achu *et al.*, 2005) as shown in Table 1. Furthermore, water melon, wild melon, hybrid melon, bitter melon, egusi melon, gourd, pumpkin, cucumber, loofah seeds cultivated in Turkey, Bangladesh, Ivory Coast, Egypt, Nigeria, Iran, Serbia and Malaysia exhibited 52.00% (Paksoy *et al.*, 2010), 33.93-36.21% (Ali *et al.*, 2008), 42.67-56.67% (Loukou *et al.*, 2007), 50.10-51.01% (El-Adawy and Taha, 2001), 13.15-56.5% (Badifu, 1991; Dawodu, 2009; Anhwange *et al.*, 2010), 50.00% (Baboli and Safe Kordi, 2010), 22.1% (Milovanovic and Picuric-Jovanovic, 2005), 29.8-33.55 (Ismail *et al.*, 2010) oil yields, respectively.

Table 1. Nutritional composition (%) of winter melon seed oil in comparison with other cucurbit seed oil

Cucurbit seed	Crude protein (%)	Crude Fat (%)	Crude fiber (%)	Ash (%)	References
<i>Benincasa hispida</i>	16.57 ± 0.05	34.12±0.40	21.53±0.03	2.7±.0.01	
<i>Citrullus lanatus</i> var. <i>colocynthisoides</i>	15.75	27.1	31.34	4.62	Mariod <i>et al.</i> , 2009
<i>Cucumis prophetarum</i>	14.5	10.9	31.58	8.33	Mariod <i>et al.</i> , 2009
<i>Cucumis sativus</i>	17.5	25.85	25.86	4.01	Mariod <i>et al.</i> , 2009
<i>Luffa echinata</i>	15.75	23.8	31.81	3.41	Mariod <i>et al.</i> , 2009
<i>Cucumis melo</i> var. <i>flexuosus</i>	15.75	22.23	36.04	5.69	Mariod <i>et al.</i> , 2009
<i>Cucumis melo</i> Var. <i>agrestis</i>	16.62	23.33	34.36	5.3	Mariod <i>et al.</i> , 2009
<i>Cucumeropsis mannii</i>	40.49	44.85	3.81	3.74	Achu <i>et al.</i> , 2005
<i>Cucurbita maxima</i>	34.39	49.05	3.44	3.95	Achu <i>et al.</i> , 2005
<i>Cucurbita moschata</i>	32.03	50.81	3.54	4.75	Achu <i>et al.</i> , 2005
<i>Lagenaria siceraria</i>	34.19	50.08	4.04	3.68	Achu <i>et al.</i> , 2005
<i>Cucumis sativus</i>	28.68	53.76	4.15	3.47	Achu <i>et al.</i> , 2005

Values are mean ± SD of 3 replications

Oil fatty acids composition

The Gas Chromatograph chromatogram (Figure 1) showed the identification of palmitic, stearic, oleic, linoleic and linoleliadic acid in winter melon seed oil. Table 2. shows the fatty acids (FA) composition of winter melon seed oil in comparison with other cucurbitaceae seed oils. The most abundant fatty acid detected in the tested seed oils was linoleic acid, the

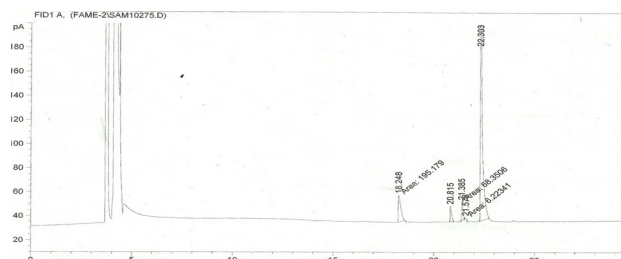


Figure 1. GC Chromatogram for fatty acid profile of winter melon (*Benincasa hispida*) seed oil

predominant fatty acid in the oil, accounted for 80.07% of the total fatty acids. The contents of palmitic, oleic, stearic and linoleliadic acids were found to be 11.71%, 4.10%, 3.74% and 0.37% respectively. Previously, Anwar *et al.* (2011) also found that winter melon seed oil had the highest amount of linoleic acid (70.64%) as the principal component, followed by palmitic acid (12.45-17.59%), oleic (8.46-12.87%) and stearic acid (5.13-7.48%) respectively. These results were also in comparison with the results given by Sew *et al.* (2010). The results of Sew *et al.* (2010) showed that the winter melon seed oil contains high amount of linoleic acid (67.37%), followed by palmitic acid (12.45-17.59%), oleic (8.46-12.87%) and stearic acid (4.83%). The present fatty acid composition revealed winter melon seed oil to be a rich source of a valuable essential fatty acid (linoleic acid C18:2- ω-6).

The fatty acid composition of the winter melon oil in the present analysis was in agreement to those reported in the literature for other Cucurbitaceae seed oils (Table 2). The winter melon seed oil generally containing low amounts of saturated fatty acids and high contents of polyunsaturated fatty acids (PUFA), especially C18:2. It is now widely accepted that diet with low saturated fatty acids and high in polyunsaturated fatty acids (PUFA) is beneficial for health.

It is obvious from Table 2. that all Cucurbitaceae seed oils, except *Telfairia pedata*, expressed the presence of more than 50% of polyunsaturated fatty acids (C18:2). However, the levels of C18:2 and other fatty acids varied among the species referred. These differences might be due to varying features such as harvesting time, seed drying conditions, seasonal variation and fruit seed maturity. Winter melon seed exhibited the highest level of linoleic acid among the other oils from the same plant family. The high proportion of polyunsaturated fatty acids (PUFA) and monounsaturated fatty acids (MUFA) and low amounts of saturated fatty acids (SFA) indicate the possible higher oxidation rate of Cucurbitaceae seed oils due to high degree of unsaturation. Although none of these oils are presently utilized on industrial scale, but some are used as cooking oil in several countries of Africa and the Middle East (Al-Khalifa, 1996).

Table 2. Fatty acid composition in percentage (%) of Indian winter melon seed oil in comparison with other cucurbit seed oil

Cucurbit seed oil	Common name	C16:0 (s)	C16:1 (m)	C18:0(s)	C18:1(m)	C18:2(p)	C18:2 (t)	C18:3	SFA	MUFA	PUFA	References
<i>Benincasa hispida</i>	Wax gourd	11.71±0.14	N.A	3.74±0.01	4.10±0.02	80.07±0.40	0.37±0.00	N.A.	15.45	4.10	80.37	
<i>Citrullus colocynthis</i>	Wild gourd	10.1	N.A	6.7	13.1	70.1	N.A.	N.A.	16.8	13.1	70.1	Yamiv et al., 1999
<i>Cucumis dipsaceus</i>	Wild cucumber	10	N.A	5	6	78	N.A.	N.A.	15	6	78	Chisholm and Hopkins, 1964
<i>Cucumis melo</i>	Honey dew melon	11	N.A	4	15	70	N.A.	N.A.	15	15	70	Chisholm and Hopkins, 1964
<i>Cucumis sativus</i>	Cucumber seed	16	N.A	5	7	71	N.A.	N.A.	21	7	71	Mattson and Volpenhein, 1963
<i>Cucurbita ficifolia</i>	Asian pumpkin	12	N.A	4	26	57	N.A.	N.A.	16	26	57	Chisholm and Hopkins, 1965
<i>Cucurbita foetidissima</i>	Buffalo gourd	9.7	N.A	4.5	27	58.5	N.A.	N.A.	14.2	27	58.5	Weber et al., 1980
<i>Cucurbita lanatus (Chinese)</i>	Water melon	10.4	0.21	7.6	12.5	68.5	N.A.	0.13	18	12.71	68.63	Al-khalifa, 1996
<i>Cucurbita lanatus (Egyptian)</i>	Water melon	11.91	0.21	8.2	15.4	62.4	N.A.	0.1	20.11	15.61	62.5	Al-khalifa, 1996
<i>Cucurbita lanatus (Iranian)</i>	Water melon	11	N.A	8	17.8	63.7	N.A.	0.11	19	17.8	63.81	Al-khalifa, 1996
<i>Cucurbita maschata</i>	Winter squash	13.1	0.3	6	26.2	53.2	N.A.	0.12	19.1	26.5	53.32	Al-khalifa, 1996
<i>Cucurbita pepo</i>	Squash seed	17	N.A	7	16	60	N.A.	N.A.	24	16	60	Mattson and Volpenhein, 1963
<i>Lagenaria siceraria</i>	Bottle gourd	14	N.A	3	7	76	N.A.	N.A.	17	7	76	Chisholm and Hopkins, 1964
<i>Luffa Cylindrical</i>	Dishcloth gourd	13	N.A	6	15	66	N.A.	N.A.	19	15	66	Chisholm and Hopkins, 1964
<i>Telfairia pedata</i>	Oyster nut	35	N.A	14	7	44	N.A.	N.A.	49	7	44	Chisholm and Hopkins, 1964

N.A.: Data not available

Values are mean ±SD of 3 replications

C 18:2 (t) is a trans isomer of linoleic acid

SFA= saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: poly unsaturated fatty acids

Conclusion

The results of the present study showed that winter melon seeds contained appreciable amounts of dietary fiber, protein and crude fat and thus can be consumed for dietary purposes. The oil, extracted from the winter melon seeds, purchased from Allahabad, India, revealed the presence of high amount of linoleic acid (80.07%) as well as it exhibited low levels of saturated fatty acids, suggesting that winter melon oil can be used as a diluent with other edible oils to enhance their essential fatty acid content i.e. linoleic acid and make its potential food uses for health benefits. Additional research on the detailed physicochemical and bioactive properties of Indian winter melon seed and seed oil is crucial to explore their commercial and functional foods applications.

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