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Anti-Nutrient, Phytochemical and Free Fatty Acid Composition of Dehulled and Undehulled Sweet Princess Watermelon (*Citrullus lanatus*) Seed Flour

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Abstract: Dietary alterations have strong effects (both positive and negative) on health throughout life. Nutrition transition is implicated with the development of chronic non communicable diseases (NCD) and these are risk factors of disabilities and premature death in developing countries. Legumes have been shown to play important role in the prevention of NCD because of their constituents. This study assessed anti-nutrient, phytochemical and free fatty acid composition of dehulled and undehulled sweet princess watermelon (*Citrullus lanatus)* seed flour. Seeds were extracted manually from fresh ready-to-eat sweet princess watermelon fruit purchased from a local market in Enugu State, Nigeria. Water was used to wash off pulp on seeds before sun-drying for 72 h. The dried seeds were divided into two portions, one dehulled manually and milled and the other milled undehulled. Anti-nutrient, phytochemical and free fatty acids composition of the samples was determined in triplicate using standard procedures. Means were statistically separated and compared. The undehulled sample contained significantly ($p<0.05$) more hemagglutinin (16.80 mg/100 g) than the dehulled (10.19 mg/100 g). There was no significant differences in oxalate and phytate values of undehulled (0.09 and 0.24 mg/100 g, respectively) and dehulled (0.06 and 0.17 mg/100 g, respectively) samples. Differences in flavonoid and alkaloid contents of the samples were significant. The samples had varied palmitic, oleic and lauric acid values (0.12 g/100 g, 0.13 mg/100 g and 0.09 g/100 g, respectively in undehulled and 0.18 g/100 g, 0.21 mg/100 g and 0.14 g/100 g, respectively in dehulled). Dehulled sample had low levels of anti-nutrients except for tannins and appreciable amounts of phytochemicals and free fatty acids, its consumption should be popularized.

Key words: Anti-nutrient, phytochemical, free fatty acid, *Citrullus lanatus*, seed flour

INTRODUCTION

Nutrition is a major modifiable determinant of non-communicable diseases (NCDs) and general wellbeing. Urbanization precipitates changes in lifestyle and dietary pattern which lead to nutrition transition - a shift from consumption of traditional diets rich in complex carbohydrates and dietary fibre to high consumption of energy dense modified and processed foods. Changes in dietary and lifestyle patterns are contributory factors to the development of NCDs and premature death in developing countries (WHO, 2003). The underlining principle for healthy dietary pattern according to WHO (2002) is consumption of fruits and vegetables, legumes, oily fish, whole grains and nuts, limited intake of dairy products, high-fat meat, snacks, sweetened drinks and other foods with low or no nutritional values. Consumption of fruits is important in the maintenance of good health and protection against NCDs (USDA, 2003; Scalzo *et al.*, 2005). This is because of their nutritional constituents.

Many edible tropical fruits have seeds which are often discarded as waste despite their nutrient potentials. A specie of watermelon (*Citrullus lanatus*) known as the sweet princess is one of such fruits whose seeds are not routinely eaten either alone or with the pulp. The seed of watermelon belongs to a class of minor legumes that is under exploited and utilized in Nigeria despite the nutritive values of legumes. There is consistent evidence from epidemiological studies that consumption of legumes play a role in the prevention of chronic diseases like diabetes, cardiovascular diseases and overweight (Mann *et al.*, 2007) as well as improving gut health (Setiawan *et al.*, 2001). Consumption of legumes is also associated with lower serum cholesterol (Leterme, 2002).

Legumes are rich in vitamins, minerals, fatty acids and phytochemicals. They also contain anti-nutrients. Anti-nutrients are natural compounds in plants which affect bioavailability of nutrients and they include among others phytate, tannins, saponins oxalate, cyanogenic glycosides and hemagglutinin (Kumar *et al.*, 2010). At low levels some antinutrients are beneficial to the body and are classified as phytochemical. Phytochemicals are biologically active compounds found in plants in small amounts which are not established nutrients but contribute significantly to protection against degenerative diseases (Ajayi *et al.*, 2011). Free fatty acids (FFA) are fatty acids that are not bound. Its value is used as one of

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the determinants of the quality of plant oil. Low FFA value is indicative of good quality of the oil. Some legumes are high in mono and poly-unsaturated fatty acids (Garjani *et al*., 2009) and these have positive health implications for humans.

Sweet princess watermelon seed flour have been shown to contain appreciable amount of ascorbic acid, minerals (magnesium, potassium and zinc) and dietary fibre (Ibeanu *et al*., 2012). The thrust of this study therefore was to assess the anti-nutrient, phytochemical and free fatty acid composition of sun-dried dehulled and undehulled sweet princess watermelon (*Citrullus lanatus*) seed flour.

MATERIALS AND METHODS

Sweet princess watermelon (*Citrullus lanatus)* was the specie of watermelon used in this study. It has round shape, light green skin with soft sweet deep red flesh or pulp and small tan coloured seeds. Fresh ready-to-eat whole watermelon fruits were bought from a local market in Enugu state, Nigeria. The fruits were washed, cut and the seeds extracted manually from the pulp. Water was used to wash off pulp on the seeds before sun-drying for 72 h. The dried seeds were divided into two portions, one portion was dehulled manually and milled with a laboratory hammer mill into fine powder and the second portion was milled undehulled.

Anti-nutrient, phytochemical and free fatty acids composition of the samples was determined in triplicate. Phytate was determined by the method described by of Griffiths and Thomas (1981). Hemagglutinin was determined by Onwuka (2005) method and Saponin and oxalate were determined by the methods of the Association of Official Analytical Chemist (2005). Flavonoids and alkaloids were determined by the gravimetric procedure of Harborne (1973). Tannins were determined using the Follin-Dennis spectrophotometric method of Pearson (1976). Glycosides determination was carried out by the AOAC (2005). Fatty acid composition was determined by Metcalf *et al*. (1966) method. Means and standard error of the mean were determined using Statistical Package for the Social Sciences version 18. ANOVA and Duncan multiple range test were used to separate/compare the means at 5% significance level.

RESULTS

The anti-nutrient composition of dehulled and undehulled sweet princess watermelon seed flour is shown in Table 1. Hemaglutinin, oxalate and phytate were higher in undehulled sample (16.80, 0.09 and 0.24 mg, respectively) than in dehulled sample (10.19, 0.06 and 0.17 mg, respectively). These differences were significant ($p<0.05$) except for hemaglutinin. The dehulled sample had significantly ($p<0.05$) less tannin than the undehulled sample (273.56 and 389.61 mg, respectively).

Table 1: Anti-nutrient composition of dehulled and undehulled sweet princess watermelon seed flour per 100 g

Antinutrient (mg)	Dehulled	Undehulled	p-value
Hemaglutinin	10.19±0.04	16.80±0.03	0.005
Oxalate	0.06±0.00	0.09±0.00	0.108
Phytate	0.17±0.00	0.24±0.00	0.050
Tannin	273.56±12.51	389.61±4.31	0.082

Mean±SD of 3 determinations
Significance level is $p<0.05$

Table 2: Phytochemical composition of dehulled and undehulled sweet princess watermelon seed flour per 100 g

Phytochemicals (mg)	Dehulled	Undehulled	p-value
Saponin	3.27±0.09	2.42±0.06	0.078
Flavonoid	5.87±0.09	2.42±0.04	0.016
Alkaloid	2.17±0.04	3.28±0.04	0.003
Glycoside	0.23±0.00	0.27±0.01	0.210

Mean±SD of 3 determinations
Significance level is $p<0.05$

Table 3: Free fatty acid composition of dehulled and undehulled sweet princess watermelon seed flour 100 g

Free fatty acids (g)	Dehulled	Undehulled	p-value
Palmitic acid	0.18±0.00	0.12±0.00	0.00
Oleic acid	0.21±0.01	0.13±0.00	0.05
Lauric acid	0.14±0.14	0.09±0.00	0.00

Mean±SD of 3 determinations
Significance level is $p<0.05$

Table 2 shows the phytochemical composition of dehulled and undehulled sweet princess watermelon. Saponin (3.27 mg) and flavonoid (5.87 mg) were higher ($p<0.05$) in the dehulled than in the undehulled sample (2.42 mg each). The undehulled sample had more alkaloid (3.28 mg) and glycoside (0.27 mg) than the dehulled sample (2.17 and 0.23 mg, respectively.

The free fatty acid composition of the dehulled and undehulled samples is presented in Table 3. There was significantly ($p<0.05$) higher palmitic acid (0.18 g) and lauric acid (0.14 g) in the dehulled than undehulled (0.12 and 0.09 g, respectively) sample. Oleic acid (0.21 g) was also higher in dehulled sample than the undehulled sample (0.13 g, but the difference was not significant.

DISCUSSION

Hemaglutinin, oxalate and phytate were more in the undehulled sweet princess watermelon than the dehulled seeds. It is possible that these anti-nutrients are concentrated on the hard coat of the seeds. Dehulling is therefore necessary before consumption. Hemagglutinin contents of the samples were low relative to the toxic level (<25 mg/100 g) reported by Munro and Basir (1973). Heating should also be employed in processing of watermelon seeds to further reduce the hemagglutinin contents since this anti-nutrient is not heat stable (Gernah *et al*., 2012). The values of oxalate in both dehulled (0.06 mg) and undehulled (0.09 mg) samples were comparable to that reported by Johnson *et al*. (2012). These values are below the toxic level of 2-5 mg/100 g in humans reported by Hassan and Umar (2004) and the WHO (2003) tolerable oxalate limit of 105 mg/100 g. The phytate contents of the samples were within 0.63 mg/100 g reported by Johnson *et al.* (2012) and the standard safe level of 22.10 mg/100 g (WHO, 2003). The low values of phytate in both study samples, was beneficial for the absorption of minerals because phytate is known to bind with calcium, iron and zinc and thereby reduce their bioavailability.

Tannins found in the study samples were relatively higher than the safe level (150-200 mg/100 g) observed by Schiavone *et al.* (2008) in broiler chicks. This was not surprising because the seed of watermelon are legume and legumes such as beans, chickpea, cowpea and lentil are known to be high in tannins (Lila, 2007; Serrano *et al.*, 2009). Tannins are soluble in water (Chung *et al.*, 1998) therefore soaking the seeds of watermelon in water for some time before consumption might reduce the content of tannins. Tannins also act as a phytochemical at low levels and plays important role in wound healing, treating diarrhea and preventing the onset of hyperlipidemia. Saponins are known to have both beneficial and deleterious properties depending on its concentration (Seigler, 1998). The saponin contents of the dehulled and undehulled watermelon seed flour were low but slightly more than the values (0.72 mg/100 g) reported by Braide *et al.* (2012). Low levels of saponin have health promoting effects. They reduce the uptake of some nutrients such as glucose and cholesterol in the gut through intra-lumenal physicochemical interaction (Ali, 2012). According to Seigler (1998) saponins have anti-carcinogenic, immune modulation and regulation of cell proliferation properties. They form strong insoluble complexes with cholesterol and bile making them unavailable for absorption (Oakenfull and Sidhu, 1990). The flavonoid content of the undehulled watermelon seeds flour (2.42 mg/100 g) reported was not in line with the values (0.97 mg/100 g) reported by Braide *et al.* (2012). This could be attributed to geographical variations. The values of study samples were however, below the range (20-70 mg/100 g) reported by Beecher, (2003) and the tolerable limit (52.02 mg/100 g) of the WHO (2003). This is a positive result because flavonoids have been implicated with the anti-viral, anti-inflammatory, anti-tumor, anti-oxidant properties (Dreosti, 2000) and reduced risk of lung cancer (Willet and Speizr, 2000). Alkaloid and glycoside values reported in this study were higher than Braide *et al.* (2012) values. This again could be due to geographical variations. However the values for alkaloids in this study were lower than the standard limit of 61 mg/100 g (WHO, 2003). The quantities of glycoside in the study samples were lower than 1 mg/kg body weight recommended (FSANZ, 2004). This meant that the consumption of the seeds would not be harmful to the body. The amounts of oleic acid in the two samples were lower than the values reported by Raziq *et al.* (2012) and Duduyemi *et al.* (2013). Low values of oleic, palmitic and lauric acids in the study samples suggested that the seeds of sweet princess watermelon were not oil seeds. However, the low values of oleic meant that the oil of sweet princess watermelon was of good quality.

Conclusion: Saponin, alkaloid, flavonoid and glycoside in sweet princess watermelon were within acceptable ranges to act as phytochemicals. The anti-nutrients present in the seeds were below toxic levels except for tannins which were higher than the standard. Dehulling, soaking and heating were recommended to further reduce the anti-nutrients in the seeds in general and tannins in particular.

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