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Improvement of the Biochemical Properties of Watermelon Rinds Subjected to *Saccharomyces cerevisae* Solid Media Fermentation

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Abstract: Improvement of the nutritional quality of watermelon rinds was attempted by *Saccharomyces cerevisae* solid media fermentation. The fermented rinds were analyzed with regard to proximate composition, antinutrient contents and phytonutrient properties and compared to unfermented watermelon rinds. Results revealed a significant increase ($p<0.05$) in the protein, lipid, ash and crude fibre contents. There were significant decreases ($p>0.05$) in the antinutrient contents of the fermented watermelon rinds. Significant increases ($p<0.05$) were observed in the phenolic and flavonoid contents of the fermented watermelon, as opposed to the significant decrease ($p>0.05$) of the alkaloid and saponin contents. These results indicate that *Saccharomyces cerevisae*, a cheap, non-pathogenic and saprophytic fungus, would efficiently improve the nutritional qualities of watermelon rinds and reduce the antinutrition levels.

Key words: Watermelon rinds, *S. cerevisae*, antinutrients and phytonutrients

INTRODUCTION

Watermelon, *Citrullus lanatus* (Thunb.) is a tropical fruit which grows in almost all part of Africa and South East Asia (Koocheki *et al*., 2007). It belongs to the family of cucumber (*Cucurbitacea*). It is large, oval, round or oblong in shape. The skin is smooth, with dark green rind or sometimes pale green stripes that turn yellowish green when ripe. Watermelon is a very rich source of vitamins and often used as an appetizer or snack, depending on how it is prepared (Kerje and Grum, 2003). It also serves as a good source of phytochemical and lycopene, a red carotenoid pigment which acts as antioxidant during normal metabolism and protects against cancer (Perkins-Veazie and Collins, 2004). Watermelon contains a significant amount of citrulline and after consumption of several kg an elevated concentration is measured in the blood plasma; this could be mistaken for citrullinaemia or other urea cycle disorder (Mandel *et al*., 2005). The rind is usually discarded, applied to feeds or fertilizer. But they are also edible and sometimes used as a vegetable. In China, they are stir-fried, stewed, or more often pickled. When stir-fried, the de-skinned and de-fruited rind is cooked with olive oil, garlic, chili peppers, scallions, sugar and rum. Pickled watermelon rind is also commonly consumed in the Southern US, Russia, Ukraine, Romania and Bulgaria (Southern U.S. Cuisine, 2010). Medicinally, watermelons are mildly diuretic. Watermelons contain large amounts of beta carotene and are a significant source of lycopene (Collins *et al*., 2005). In Nigeria, watermelon rinds are fermented, blended and consumed as juice.

Food fermentation can be described as a process of subjecting food to the action of micro-organisms or enzymes so that desirable biochemical changes cause significant modification to the food (Campbell-Platt, 1987). This study aims at investigating nutrient enrichment of watermelon rinds using cheap, non-pathogenic and saprophytic aerobic *Saccharomyces cerevisae*

MATERIALS AND METHODS

Sample preparation: Watermelon fruits were purchased from a local market at Lagos, Nigeria. The rinds were peeled-off and subjected to fermentation in plastic jars. A pure strain of *Saccharomyces cerevisae* was sub-cultured and inoculated into 500 g of the soaked rinds as the starter culture and 730 ml nutrient solution (urea (80 g), $MgSO_4$ $2H_2O$ (7 g), KH_2PO_4 (13 g) and citric acid (20 g)) and then allowed to ferment for 38 h. The micro-fungi fermented watermelon rinds were then blended. Another 1 kg of unfermented watermelon rind blend was also prepared. They were stored in laboratory freezers for further analysis.

Sample analysis

Proximate analysis: The proximate composition (protein, ash, lipid, crude fibre and carbohydrate) of the micro-fungi fermented and unfermented watermelon rinds was evaluated using the standard AOAC method (1997).

Anti-nutritional factors: Tannin was determined by the modified Vanillin - HCl method using 1.0 mg/ml of

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catechin in 1% HCl-MeOH as standard, the coloured substituted product was measured at 500 nm (Price *et al.*, 1978). Phytate was determined by the anion exchange method (Harland and Oberleas, 1986) using KH_2PO_4 as standard. Oxalate was determined by titrimetrically (Falade *et al.*, 2004) precipitation as calcium oxalate and titrated against standard potassium permanganate. The oxalate was calculated as sodium oxalate equivalent.

Phytonutrient analysis: The qualitative and quantitative phytonutrient properties of the fermented and unfermented samples were determined using standard methods described by Harborne (1993); Boham and Kocipai (1994); Ebrahimzadeh *et al.* (2007) and Nabavi *et al.* (2008).

Statistical analysis: Statistical significance was established using One-Way Analysis of Variance (ANOVA) and data were reported as mean ± standard deviation. Statistical analyses were carried out using SPSS for Windows, version 14.0 (SPSS Inc. Chicago, IL.USA).

RESULTS AND DISCUSSION

The WHO food safety unit has set high priority to the research area of fermentation as a technique for food preparation/storage (Sahlin, 1999). Fermentation has been shown to improve protein quality and digestibility, vitamin B content and microbiological safety and keeping quality (Hotz and Gibson, 2007). Since watermelons are in abundance in almost all parts of Africa, methods for enhancing the nutrient content and reducing the antinutrients without adversely affecting the acceptability become very important as this fruit has potential to improve nutrition, boost food security, foster rural development and support sustainable land care. This study lays emphasis on the rinds.

The results of the proximate analysis (Table 1) revealed that the protein content of the *Saccharomyces cerevisae* fermented watermelon rinds was significantly higher ($p<0.05$) than the unfermented watermelon rinds. This high protein content could be attributed to the ability of the *Saccharomyces cerevisae* to secrete some extracellular enzymes (protein) into the watermelon rinds during their metabolic activities on the rinds during fermentation by the fungi (Oboh and Akindahunsi, 2003). The multiplication of the fungi in the watermelon rinds in the form of single cell proteins could also provide a reason for the increase in the protein content of fermented watermelon rinds (Akindahunsi *et al.*, 1999). The observed increase in lipid content of the fermented watermelon rinds could be as a result of possible transformation of carbohydrate to fat (Lehninger, 1987). Certain fungi have been reported to produce microbial oil during the course of fermentation (Akindumila and Glatz, 1998). There was a significant change in the ash and crude fibre contents of the fermented watermelon rinds. The increased ash content is an indication of increased mineral contents.

Table 1: Result of proximate analysis of fermented and unfermented watermelon rinds

Parameters (%)	UFWM	FWM
Moisture	91.22±0.65	87.06±0.29
Ash	0.92±0.04	1.61±0.07
Lipid	0.69±0.06	0.81±0.04
Protein	1.52±0.05	2.80±0.05
Crude fibre	0.97±0.04	1.78±0.06
Carbohydrate	4.68±0.10	5.94±0.10

Key: UFWM = Unfermented Watermelon Rinds; FWM = Fermented Watermelon Rinds. Data = mean±SD, n = 4

Results of the antinutrients analysis are shown in Table 2. Antinutrients are natural or synthetic compounds that interfere with the absorption of nutrients. A significant reduction ($p>0.05$) in the phytic acid content was observed in the *Saccharomyces cerevisae* fermented watermelon rinds compared to the unfermented rinds. Phytic acid has been shown to form insoluble complexes with calcium, zinc, iron and copper (Cheryan, 1980), interfering with their absorption. Fermentation can induce phytic acid hydrolysis via the action of microbial enzymes, which hydrolyze phytic acid to lower inositol phosphates. It can be said that *Saccharomyces cerevisae* brought about the hydrolysis.

Such hydrolysis is important because myoinositol phosphates with <5 phosphate groups (i.e., IP-1 to IP-4) do not have a negative effect on zinc absorption (Lonnerdal *et al.*, 1989) and those with <3 phosphate groups do not inhibit non-heme iron absorption (Sandberg *et al.*, 1999; Hurrell, 2004). Low molecular weight organic acids (e.g., citric, malic, lactic acid) produced during fermentation generates a low pH that optimizes the activity of endogenous phytase (Teucher *et al.*, 2004). The tannin content of *Saccharomyces cerevisae* fermented watermelon rinds was significantly lower ($p>0.05$) than the unfermented watermelon rinds. Tannins have been reported to affect nutritive value of food products by forming complex with protein thereby inhibiting digestion and absorption (Oboh and Akindahunsi, 2003). It also interferes with metal ion availability. Recent studies suggest that free or protein-complex condensed and hydrolysable tannins are more effective than small phenolics in antioxidant activities (Hagerman, 2002). Oxalate has a negative effect on mineral availability. Reduced oxalate content was observed in *Saccharomyces cerevisae* fermented watermelon rinds at a significant difference ($p<0.05$) compared to the unfermented rinds. Diets high in oxalate increases the risk of renal calcium absorption and has been implicated as a source of kidney stones (Chai and Liebman, 2004).

Table 2: Results of antinutrient analysis of fermented and unfermented watermelon rinds

Antinutrients	FWM (mg/100 g)	UFWM (mg/100 g)
Tannin	12.15±0.09	13.23±0.42
Phytic acid	394.88±4.43	620.50±6.57
Oxalate	90.00±3.35	158.00±1.65

FWM = Fermented Watermelon Rinds; UFWM = Unfermented Watermelon Rinds. Data = mean ± SD, n = 4

Table 3: Results of phytonutrient screening of fermented and unfermented watermelon rinds

Phytonutrients	FWM	UFWM
Saponin	+	+
Phlobatanin	-	-
Terpenoid	-	-
Flavonoid	+	+
Cardiac glycoside	+	+
Phenol	+	+
Alkaloids	+	+

Key: + = Present; - = Absent. FWM = Fermented Watermelon Rinds; UFWM= Unfermented Watermelon Rinds

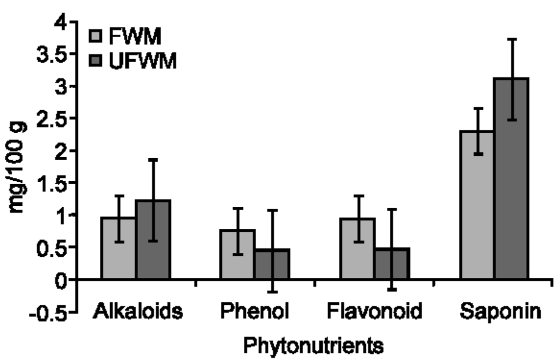

Fig. 1: Qualitative analysis of phytonutrients of fermented and unfermented watermelon rinds. Data = means ± SD. FWM = Fermented Watermelon Rinds; UFWM = Unfermented Watermelon Rinds

Phytonutrient analysis (Table 3 and Fig. 1) revealed the presence of saponin, flavonoid, cardiac-glycoside, alkaloids, phenols and saponins. Phytonutrients are natural bioactive compounds from plants with general benefits to human health. Saponin, flavonoids, phenol and alkaloids contents were quantified (Fig. 1). There was a significant decrease in the alkaloid and saponin contents of *Saccharomyces cerevisae* fermented watermelon rinds. Alkaloids, comprises of a large group of nitrogenous compounds and widely used as cancer chemotherapeutic agents. Alkaloids have also been reported to interfere with cell division (Valero and Salmeron, 2003). Saponins have been reported to possess haemolytic activity and cholesterol binding properties (Nijveldt *et al*., 2001). They serve as natural antibiotics, helping the body to fight infections and microbial invasions (Nijveldt *et al*., 2001). There was a significant ($p<0.05$) increase in the total phenolic and flavonoid contents of *Saccharomyces cerevisae* fermented watermelon rinds compared to that of the unfermented rinds. The best-described property of almost every group of flavonoids is their capacity to act as antioxidants. The flavones and catechins seem to be the most powerful flavonoids for protecting the body against ROS (Sodipo *et al*., 2000). Studies have revealed that consumption of flavonoids can be used in the management of coronary heart disease (Knekt *et al*., 1996). The relationship between total phenol content and antioxidant activity has been widely studied in different foodstuffs (Jayaprakasha *et al*., 2008). Antioxidant activity of foodstuff significantly increases with the presence of high concentration of total phenol and flavonoid contents. Therefore the increased phenolic and flavonoid contents of the fermented watermelon rinds indicate high potential antioxidant activities.

Conclusion: Results from this study indicate that *Saccharomyces cerevisae*, a cheap, non-pathogenic and saprophytic fungus, would efficiently increase the nutritional qualities of watermelon rinds and reduce the antinutrition levels.

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