

Effect of pre-treatments on the phytochemical composition of watermelon *(Citrullus lanatus)* rind

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<u>Abstract</u>

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Introduction

Processing of watermelon generates considerable amount of wastes in the form of peels, rind and seeds. Every part of the fruit has nutritional value, including the rind, peel and the seeds. These wastes are also rich in phytochemicals and can be exploited to investigate its nutraceutical properties. Thus, the present study was designed to determine the effects of blanching on the overall quality of the watermelon rind. The rind was steam and water blanched for 1, 2 and 3 minutes. The samples were then analyzed for their phytochemical contents. Results demonstrated that blanching had considerable effects on the phytochemical composition and anti-nutrient properties of the watermelon rind. There was drastic reduction in the anti-nutrients as a result of blanching. Steam blanching was more efficient in preserving the antioxidant activity and total phenol content in comparison to water blanching. While water blanching brought about a drastic reduction in the anti-nutritional factors.

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Significant amount of fruit and vegetable wastes are generated by the food processing industries. These large amounts of solids waste arising from the consumption of fruit and vegetable are known as agro wastes which pose problems in disposal and also lead to loss of valuable biomass and nutrients. These agro wastes can be converted into useful products for the production of functional foods from fruits and vegetables.

Enhancing food waste utilization in daily diets and also in drugs could enhance food supply, health and the environment. The increasing waste burden on the environment could be managed by preventing the accumulation of these solid food wastes by waste disposal or by increasing the dietary utilization of the wastes (Anthony, 2015).

Research by Peschel *et al.* (2006) shows that byproducts in general contain a variety of biologically active compounds that are mostly discarded as wastes. This not only causes a loss in the potentially valuable resource but also aggravates a problem of waste disposal. The use of these wastes can contribute to lower production costs in the food industry and create alternative functional foods for human consumption.

Fruits and vegetables are known to contain a variety of natural bioactive compounds (Pennington

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and Fisher, 2010) such as flavonoids, anthocyanins, vitamins C and E, phenolic compounds, dietary fibre, and carotenoids (Gonzalez-Aguilar *et al.*, 2008).

One such medicinal plant is *Citrullus lanatus*. Every aspect of the fruit has nutritional value, including the rind, peel and the seeds. The rind is usually discarded but they are edible, and sometimes used as a vegetable. Jayaprakasha *et al.* (2001) stated that the seed, peel and rind of some fruits have greater vitamins, fibres, minerals and other essential nutrients than the pulp. The most common way watermelon is eaten, is the consumption of the pink or yellow flesh. However, other ways of consumption include watermelon rind pickles, deep fried watermelon, watermelon flavoured cake and watermelon lemonade (Wind, 2008).

The therapeutic effect of watermelon has been attributed to antioxidant compounds (Leong and Shui, 2002; Lewinsohn *et al.*, 2005) such as citrulline which protects the body from free-radical damage. Also, citrulline is converted to arginine, an amino acid that is vital for the functioning of heart, circulatory system and immune system. Thus researchers speculate that watermelon rind might bring about a relaxation in the blood vessels (Rimando and Perkins-Veazie, 2005).

The knowledge of the nutritive and the antinutrient content of various parts of the fruits will encourage the consumption and re-utilization of seeds and peels discarded as waste for human food, animal feed and fertilizer. Nowak and Haslberger, (2000) proved that most of the anti-nutritional factors become ineffective by treatments like heating, soaking, blanching and germination.

Many food composition data bases do not take into consideration the fact that concentrations of nutrients may change through cooking practices such as blanching. This holds great importance as only a small amount of vegetables is consumed in the raw state (Amin *et al.*, 2006).

Consequently, the objective of this research work is to investigate the effect of steam and water blanching on the anti-nutritional properties and antioxidants in watermelon rind. An enhanced reduction in the anti- nutrients by these methods will improve the utilization of the rind.

Materials and Methods

Collection of raw materials

Fresh watermelon with dark and light green stripes was chosen. Watermelons were collected from a local market and were used fresh.

Preparation of sample

The whole fruit was washed, peeled and the rind was separated from the flesh and then the rind was grated to reduce its size.

Pre-treatments

Water blanching

Approximately 1000 mL of water was poured into a stainless steel vessel and heated at 100°C. Watermelon rind (200 g) was immersed in the boiling water at 100°C for 1, 2 and 3 mins. The samples were drained on a stainless steel sieve and then weighed.

Steam blanching

Steam blanching was conducted by suspending 200 g of watermelon rind above 1000 mL of boiling water for 1, 2 and 3 mins in a stainless steel steamer with a lid. The samples were drained on a stainless steel sieve and then weighed.

Experimental design

Steam blanching and water blanching was carried out for 1, 2 and 3 minutes respectively. Phytochemical analysis of all the samples obtained was determined. All analysis was carried out in triplicates and the values were reported as mean \pm SD. Statistical analysis of the data was carried out using SPSS version19.

Determination of saponins

Saponins were quantified in accordance to the method described by Famurewa et al. (2014). 5 g of the blanched sample was put into a conical flask with 100 ml of 20% aqueous ethanol. The samples were heated over a hot water bath at 55°C for 4 hours with continuous stirring. The mixture was then filtered and the residue was re-extracted with 200 ml of 20% ethanol. The combined extracts were then reduced to 40 ml by heating over a water bath at 90°C.The concentrate was transferred into a separating funnel with 20 ml of diethyl ether and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. The combined extracts were then washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath and after evaporation, the samples were dried to a constant weight and the saponin content was calculated as percentage.

Determination of alkaloids

The alkaloid content of the samples was determined as per the method described by Oseni and Okoye (2013). 5 g of the sample was weighed into a beaker with 200 ml of 10% acetic acid in ethanol and allowed to stand for 4 hours. This was filtered and the extract was concentrated by heating on a water bath to one-quarter of the original volume. Concentrated ammonium hydroxide was added dropwise until the precipitation was complete. The solution was then allowed to settle down and the precipitate was separated and washed with dilute ammonium hydroxide and filtered. The residual alkaloid was dried and weighed.

Determination of tannins

The total tannin content was assessed by the standard protocol of Keerthana *et al.* (2013) with little modifications as follows: 0.5 mL of the sample extract was diluted with 80% ethanol. From the diluted sample, 0.1 mL was added to 2 mL of Folin-Ciocalteu reagent. After 8 mins, 7.5 mL of 7% sodium carbonate solution was added and incubated for 2 hours. The absorbance was measured at 760 nm and the tannin content was estimated using tannic acid curve as the standard.

Determination of total phenolic content

Total phenolics were analysed spectrophotometrically using the modified Folin– Ciocalteu colorimetric method as described by Chang *et al.* (2005). 0.5 g of sample was extracted with 50 mL ethanol for 1 hour and diluted with distilled water. 125 μ l of the diluted extract was mixed with 0.5 mL of distilled water in a test tube followed by addition of 125 μ l of FC reagent and allowed to incubate for 6 mins. Then, 1.25 mL of 7% sodium carbonate solution was added and the volume was made up with distilled water. The samples were incubated for 90 mins at room temperature and measured using an UV/Vis spectrophotometer at 760 nm. The total phenolic content was determined from the standard curve of gallic acid.

Determination of antioxidant activity

Antioxidant activity was determined by DPPH assay as described by Sarker *et al.* (2006). 0.5gm of sample was extracted with 10ml of 80% ethanol in a water bath for 3 hr at 45°C. The samples are then centrifuged at 5000 rpm for 5 mins. The supernatant is separated and varying concentrations of sample are taken in tube. 0.004% of DPPH solution is made and 6 ml of solution is added in each tube. The tubes are incubated in dark for 30 mins and OD at 517 nm is taken.

Results and Discussion

The results for anti-nutritional factors as affected by blanching methods are represented in table1 and 2. Anti-nutritional factors are mainly responsible for hindering with the uptake and utilisation of nutrients from plants. The alkaloid, tannin and saponin content present in raw watermelon rind were in close agreement to Anthony (2015). Blanching brought about a clear reduction in the tannins, alkaloids and saponin as these factors are sensitive to thermal treatments. The loss in steam blanching was less compared to water blanching as the area of contact between the watermelon rind and the blanching medium is comparatively less in steam blanching. Similar results were obtained by Nkafamiya et al. (2010) and Yusufu and Obiegbuna (2015). With an increase in the blanching time there was subsequent reduction in the anti-nutritional factors. Similar results were obtained by Nwosu (2010) in effect of blanching on anti- nutritional properties of Asparagus bean.

Phenolic compounds account for the antioxidant activity in fruits and vegetables. High intensity heat treatment brings about maximum loss of phenolic content which may be due to several reasons like thermal degradation or diffusion of various components into water (Gonçalves *et al.*, 2010). Jaiswal *et al.* (2012) reported a similar trend during blanching of cabbage.

Also, long blanching times is likely to bring

Table 1. Phytochemical composition in Steam Blanching

| Sample | Tannins | Alkaloids | Saponins | Total phenolic |
|-----------|------------------------|-----------------------|------------------------|--------------------------|
| | (mg/100g) | (mg/100g) | (mg/100g) | content (mg/100g) |
| Fresh | 1.12±0.02 ^a | 1.6±0.04 ^ª | 2.11±0.05 ^a | 0.4017±0.02 ^a |
| 1 minute | 1.03±0.01 ^b | 1.2±0.01 ^b | 2.0±0.00 ^b | 0.3714±0.01b |
| 2 minutes | 0.96±0.01 ^c | 0.9±0.00 ^c | 1.77±0.01° | 0.3125±0.00 ^c |
| 3 minutes | 0.81±0.03 ^d | 0.6±0.01 ^d | 1.52±0.02 ^d | 0.2875±0.04 ^d |

Data bearing different letters in the same column are significantly different at 5% significance level (p<0.05).

Table 2. Phytochemical composition in Water Blanching

| Sample | Tannins | Alkaloids | Saponins | Total phenolic |
|-----------|------------------------|------------------------|------------------------|--------------------------|
| (mg/100g) | | (mg/100g) | (mg/100g) | content (mg/100g) |
| Fresh | 1.12±0.02ª | 1.6±0.04 ^a | 2.11±0.05 ^a | 0.4017±0.02ª |
| 1minute | 0.91±0.00 ^b | 1.08±0.03 ^b | 1.88±0.06 ^b | 0.3418±0.01b |
| 2minutes | 0.74±0.01 ^c | 0.55±0.02 ^c | 1.29±0.04 ^c | 0.2574±0.02 ^c |
| 3minutes | 0.47±0.00 ^d | 0.31±0.02 ^d | 0.87±0.05 ^d | 0.1412±0.00 ^d |

²Data bearing different letters in the same column are significantly different at 5% significance level (p<0.05).

about disruption of cell walls and thus result in the breakdown of phenolic compounds (Francisco *et al.*, 2010). Various enzymes like polyphenol oxidase play a vital role in the synthesis of phenolic compounds in fruits and vegetables. Thus when blanching is performed for prolonged periods it may bring about in activation of these enzymes which accounts for the reduction in the total phenolic content of watermelon rind. Similar results were obtained by Badwaik *et al.*, (2015).

Phenolic compounds in vegetables are present in 2 forms-soluble as well as in combination with cell-wall complexes. The highest loss was observed in water-blanched water melon rind, 0.402% to 0.141%, respectively which could be due to the breakdown of phenolic compounds or losses due to leaching out of compounds during blanching as most of bioactive compounds are unstable to heat and are easily solubilized. Ahmed and Rehab (2013) reported a similar trend in the blanching of cauliflower.

Anti-oxidant activity of the samples as affected by blanching is represented in figure 1 and 2. Kenny and O'Beirne (2009) reported that the loss in antioxidant activity was proportional to the area of contact between vegetables and the blanching media as well as the blanching time. The contact area in steam blanching was lesser compared to water blanching and thus the loss in antioxidant activity was comparatively lower in steam blanching as

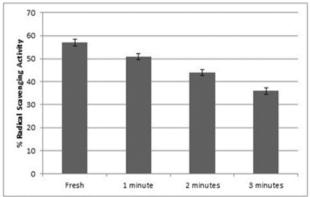


Figure 1. Anti-oxidant activity of the Steam blanched samples

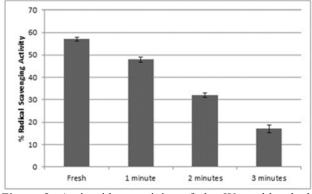


Figure 2. Anti-oxidant activity of the Water blanched samples

compared to water blanching. Similar results were obtained by Ahmed and Rehab (2013). Thus steam blanching for 1 minute was found to be favourable for the preservation of the antioxidant activity of watermelon rind. Similar results were obtained by Nurhuda *et al.* (2013).

Conclusion

As shown in this study, both water and steam blanching were found to have distinct effects on the phytochemical composition of the watermelon rind. Between the two pre-treatment methods used steam blanching caused lesser losses in the anti-oxidant content as well as in the anti-nutritional factors and thus preserved the quality to greater extent in comparison to water blanching. Blanching time also significantly affected the retention of phytochemicals as longer blanching times brought about greater losses. 1 minute blanching was found to be favorable for the preservation of phytochemicals in watermelon rind. Thus steam blanching for 1 minute is a preferred method to retain the phytochemicals in watermelon rind and also enhance its utilization for the production of beverages and pickles.

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