Blanching Attenuates Antinutrient and Mineral Content of *Basella alba* and *Amaranthus hybridus* Leaves

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Abstract

Consumption of diets rich in vegetables and fruits protects the human body from acute and chronic diseases. This study determines the effect of blanching on some antinutritional factors and mineral content of Basella alba and Amaranthus hybridus using standard methods. The vegetables were blanched in deionized water for 5 minutes at 100^{o}C. The phytate and oxalate contents of unblanched Basella alba were significantly ($p<0.05$) lowered when compared with unblanched Amaranthus hybridus. Blanching caused a significant ($p<0.05$) decrease in the phytate content by 22.3% and 32.8%, respectively when blanched Amaranthus hybridus and blanched Basella alba BBA were compared with their corresponding unblanched leaves. Also blanching elicited a significant ($p<0.05$) reduction in the oxalate level by 44.4% and 18.8%, respectively when blanched Amaranthus hybridus and blanched Basella alba BBA were matched with their unblanched counterparts. The mineral (Fe, Ca, Mg, Cu, and Zn) content was significantly ($p<0.05$) higher in unblanched Basella alba than unblanched Amaranthus hybridus, which decreases significantly ($p<0.05$) to varying extents after blanching in both vegetables. Blanching reduces the mineral content of Basella alba and Amaranthus hybridus leaves but makes them more bioavailable by reducing their phytate and oxalate. Hence, other processing techniques should be developed which reduce the antinutrient content only.

Keywords: Blanching, *Basella alba*, *Amaranthus hybridus*, phytate, oxalate

1. Introduction

Vegetables are the edible part of plants that are rich sources of micronutrients and phytochemicals (Amao, 2018; Samtiya *et al.*, 2020). Ingestion of diets rich in vegetables and fruits is strongly associated with overall good health improvement, protect the human body from acute and chronic diseases (Nath *et al.*, 2009; Babalola *et al.*, 2010), and reduced risk for some forms of cancer, heart disease, stroke, diabetes, anaemia, gastric ulcer and rheumatoid arthritis (Naeem and Ugur, 2019). *Basella alba* is a highly succulent vegetable similar to waterleaf (Harry, 2000). Of the many spinach cultivars, *Basella alba* is one of the most cultivated in Africa, probably due to good environmental factors such as high temperature, high rainfall, and moist, fertile soils with high organic matter observed in the African continent (Procher, 1995). *Basella alba* is used largely as food in the southwestern part of Nigeria. It is commonly known as "Amunututu" in the Yoruba language. *Basella alba* has been an important source of some vitamins and is useful in treating many health-related diseases (Haskell *et al.,* 2004; Ramu *et al.,* 2011; Rahmatullah *et al.,* 2010). *Basella alba* leaves are consumed because they possess gastro-protective, anti-ulcer, anti-oxidative, anti-inflammatory activity, wound healing, testosterone improving, libido improving, laxative potentials (Deshmukh and Gaikwad, 2014). Amaranthus species are a diverse category of vegetables. It is known as 'inene' in southeastern Nigeria and 'alaihu' in northern Nigeria, and 'efo' in southwestern Nigeria. *Amaranthus hybridus,* commonly called smooth amaranth, smooth pigweed, red amaranth, or slim amaranth, it is a species of annual flowering plant that can grow up to 2m. *Amaranthus hybridus* is commonly planted in West Africa, Indonesia, and Malaysia (Hugue, 1989).

Amaranthus is highly nutritious; both the grain amaranth and leaves are utilized for humans and animal food (Tucker, 1986). The plant has been used in treating dysentery, ulcers, and hemorrhage of the bowel due to its astringent property (Krochmal *et al.*,1973). Leaves possess an antibacterial effect (Cyrus *et al.,* 2008), cleansing effect and also help to reduce tissue swelling (Chatterjee *et al.,* 2012). *Amaranthus hybridus* leaves have also been reported to inhibit and/or reverse jaundice, rheumatism, pain, ache, abscesses, burns, wound menorrhagia, inflammation, and eczema (Ganjare and Raut, 2019).

Minerals are inorganic nutrients required by the body in a small amount. They are necessary for the maintenance of specific physicochemical processes that are essential to life. Although they yield no energy, they play essential roles in different metabolic processes (Soetan *et al*., 2010). Antinutritional factors such as phytate, tannin, and oxalate have been reported to be available in vegetables. Antinutritional factors are known to interfere with metabolic processes, such as the bioavailability of nutrients (Ram *et al*., 2020). Phytic acid has an affinity to chelate polyvalent cations, such as minerals, and trace elements and could, in this way, interfere with their absorption. Also, phytate inhibits the function of some digestive enzymes. Oxalate is known to interfere with the absorption of Ca^{2+} by reducing its availability due to the formation of calcium oxalate in the intestine of monogastric animals. However, it has little or no effect on the absorption of Zn (Ram *et al*., 2020).

Blanching is a technique performed before freezing, canning, or drying in which fruits or vegetables are heated for inactivating enzymes, modifying texture, preserving the colour, flavour, and nutritional value. Also, similar to other thermal processes, blanching has been shown to affect the content of some nutritional and bioactive components of vegetables such as vitamin C (Sablani, 2006). Therefore, the purpose of this study was to evaluate the effect of blanching on the antinutritional and mineral content of *Basella alba* and *Amaranthus hybridus* vegetables.

2. Materials and Methods

2.1 Collection of plant sample

Basella alba and *Amaranthus hybridus* vegetables were harvested from their natural habitat. *Basella alba* was harvested from Ikoro-Ekiti, Ekiti state, while *Amaranthus hybridus* was harvested from Samaru, Zaria, Kaduna state. The two vegetables were identified and authenticated in the Department of Biological Sciences, Ahmadu Bello University, Zaria, where vouchers numbers; 2225 and 20785 were deposited for *Basella alba* and *Amaranthus hybridus*, respectively.

2.2 Preparation of the vegetables

The harvested vegetables were washed in deionized water and divided into two parts. The first part was air-dried at room temperature (28°C), grounded, and stored for further use, while the second part was blanched.

2.3 Blanching of the vegetables

Basella alba and *Amaranthus hybridus* leaves were blanched separately in deionized water at 100°C for 5 minutes as described by De Corcuera *et al*. (2004). The blanched vegetables were dried at room temperature (28°C) and grounded. They were stored for further use.

2.4 Quantitative determination of some antinutrients

Phytic acid determination

Phytic acid content was determined using the method described by Lucas and Maskakas, (1975). Sample (2 g) was weighed into a 250 mL conical flask, 100 mL of 2% concentrated HCl was used to soak the sample for 3 hours. The mixture was filtered, 50 mL of the filtrate was placed in a 250 mL beaker, and 107 mL of distilled water was added to each solution; 10 mL of 0.003 M thiocyanate solution was added and then titrated with standard iron chloride solution, which contains 0.00195 iron per mL.

Calculations: % Phytic acid = Y × 1.19 × 100

Where Y = titre value × 0.00195

Oxalic acid determination

Oxalic acid content was determined by the titrimetric method as described by Day and Underwood, (1986). Sample (1 g) was weighed into a 100 mL conical flask with 75 mL of 3 M H_2SO_4 and stirred for 1 hour. It was then filtered using Whatman No 1 filter paper. From the filtrate, 25 mL was taken and titrated while hot (80-90°C) against 0.1 M $KMnO_4$ solution until a faint pink colour persists for at least 30 seconds.

Calculations: Oxalate (mg/100g) = $T \times V_{me} \times D.F. \times 100/M_e \times M_s$ (g)

Where T was titre value of $KMnO_4$ (mL), V_{me} was volume mass equivalent (1 cm^3 of 0.05 M $KMnO_4$ solution is equivalent to 0.00225 anhydrous oxalic acid), M_e was molar equivalent of $KMnO_4$ redox reaction, M_s was mass of sample used, D.F. was dilution factor (V_t/A =75/25 = 3), where V_t was total volume of titrate (filtrate 75 mL), and A was Aliquot used (25 mL).

2.5 Quantitative determination of mineral content and Sample digestion (Wet)

This was determined using the method of AOAC (2000). Powdered plant sample (0.2 g) was weighed into the digestion tube, and 5 mL of HNO_3-$HClO_4$ (2:1, v/v) mixture was added and mixed. The tube was placed into the digestion block inside a fume hood, and the temperature was set at 150℃ and digestion was done for 1 hour 30 minutes. The temperature was later increased to 230℃ and digested for another 30 min (white fuming stage). The temperature was reduced to 150℃, 1 mL of HCl was added and heated for 30 minutes. The digester was switched off and allows to cool to room temperature, and water was added to get to the 50 mL mark on the tube. The concentration of Fe, Mg, Zn, Cu, and Ca was read using the flame atomic absorption spectrophotometer (FAAS), AA 500 model.

2.6 Statistical analysis

All the data were expressed as mean ± standard error of the mean (S.E.M). Differences between the experimental groups were assessed by one-way ANOVA followed by Duncan's test. Values were considered statistically significant when $p< 0.05$.

3. Results and Discussion

Vegetables play an essential role in the human diet as they are essential sources of vitamins, nutrients, and antinutrients (Oboh, 2005). They are widely recommended because of their health-promoting properties, and they have a high concentration of minerals, especially electrolytes (Slavin and Lloyd, 2012). The effect of blanching on the antinutrient content of *Basella alba* and *Amaranthus hybridus* leaves was quantified (Fig. 1). The phytate and oxalate contents of unblanched *Basella alba* were significantly ($p<0.05$) lowered when compared with unblanched *Amaranthus hybridus*. Blanching-induced a significant ($p<0.05$) decrease in the phytate content (Fig. 1a) by 22.3% and 32.8%, respectively when blanched *Amaranthus hybridus* and blanched *Basella alba* BBA were compared with their unblanched counterparts. Also blanching caused a significant ($p<0.05$) reduction in the oxalate level (Fig. 1b) by 44.4% and 18.8%, respectively when blanched *Amaranthus hybridus* and blanched *Basella alba* BBA were compared with their unblanched counterparts. The reduction in phytate and oxalate in all the vegetables disagrees with the earlier report by Adegunwa *et al.* (Adegunwa *et al*., 2011), that blanching increases the phytate content of vegetables. On the other hand, it agrees with the finding of Udei *et al.* (2004), who stated that since most of the antinutritional factors are heat-labile, blanching could reduce or inactivate them. In the research carried out by Udei et al. (2004), they examined the effect of blanching for a long period of time (90 minutes). But in our research, we used a very short time (5 minutes) and we observed that the difference is significant. The reduction in these antinutritional factors can also result from rupture of the superficial layer of the vegetables where the concentration of the antinutritional factors is highest due to blanching (Virginia *et al*., 2012).

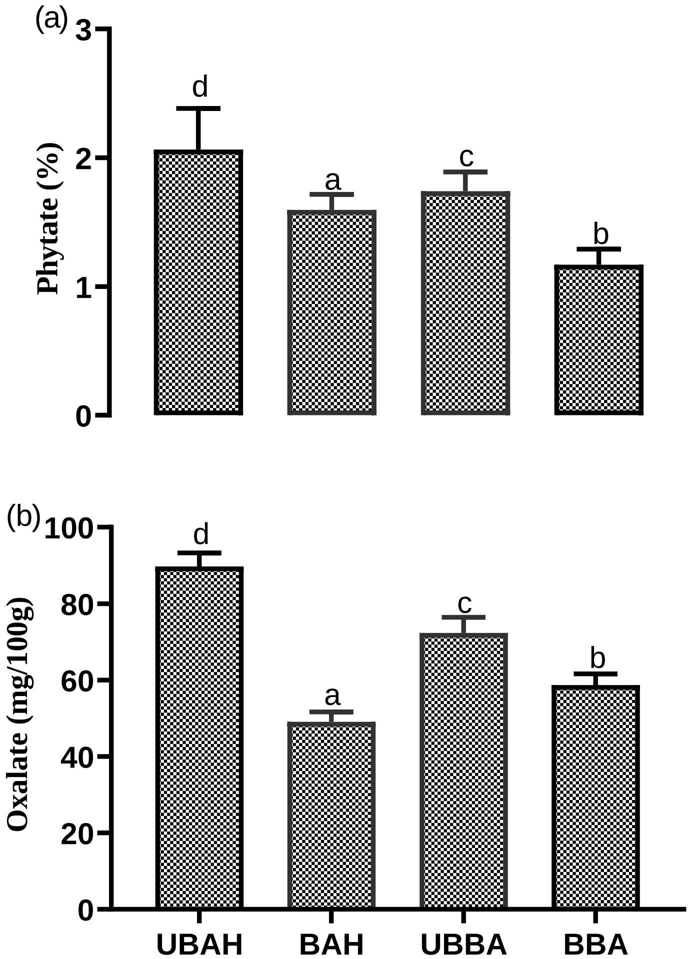

Figure 1: (a-b) Influence of blanching on antinutrient content of *Basella alba* and *Amaranthus hybridus* leaves (a) phytate (b) oxalate. UBAH, unblanched *Amaranthus hybridus*; BAH, blanched *Amaranthus hybridus*; UBBA, unblanched *Basella alba*; blanched *Basella alba* BBA. Bars represent mean ± standard deviation. Bars with different letters are significantly different at $p < 0.05$.

The influence of blanching on mineral levels of *Basella alba* and *Amaranthus hybridus* leaves was analysed (Figures 2 and 3). All the minerals (Ca, Mg, Fe, Cu, and Zn) contents of unblanched *Basella alba* were significantly ($p<0.05$) higher when compared with unblanched *Amaranthus hybridus*. Blanching caused significant ($p<0.05$) decreases in the Ca (Fig. 2a) by 15.8% and 21.6%, Mg (Fig. 2b) by 7.0% and 44.2%, respectively when blanched *Amaranthus hybridus* and blanched *Basella alba* BBA were compared with their unblanched counterparts. Also blanching elicited a significant ($p<0.05$) reduction in the transition metals Fe (Fig. 3a) by 80.7% and 41.4%, Cu (Fig. 3b) by 50.6% and 60.0%, and Zn (Fig. 3c) by 34.8% and 73.1%, respectively when blanched *Amaranthus hybridus* and blanched *Basella alba* BBA were compared with their unblanched counterparts.

The higher mineral content of *Basella alba* compared to *Amaranthus hybridus* can be due to the difference in soil composition they were planted on and the rate of mineral intake by the individual plant (Asaolu and Asaolu, 2010; Kawashima and Valente-Soares, 2005). The decrease in the mineral content of both the vegetables after blanching agrees with the finding of De Corcuera (2004), who suggested that depending on the type and duration of blanching, it can affect the colour, texture, and nutritional content of vegetables. Minerals and vitamins leach out of the vegetables during blanching, especially water blanching (Asaolu and Asaolu, 2010).

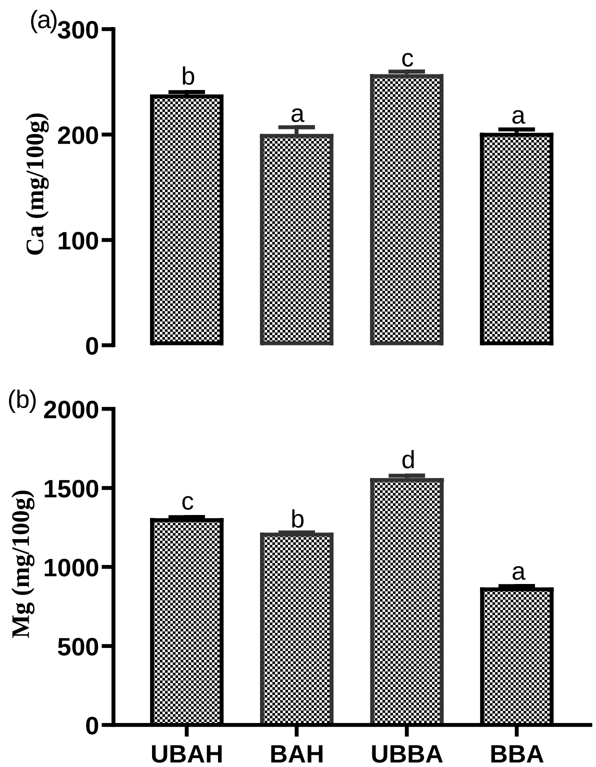

Figure 2: (a-b) Influence of blanching on the mineral content of *Basella alba* and *Amaranthus hybridus* leaves (a) Calcium (b) Magnesium. UBAH, unblanched *Amaranthus hybridus*; BAH, blanched *Amaranthus hybridus*; UBBA, unblanched *Basella alba*; blanched *Basella alba* BBA. Bars represent mean ± standard deviation. Bars with different letters are significantly different at $p < 0.05$.

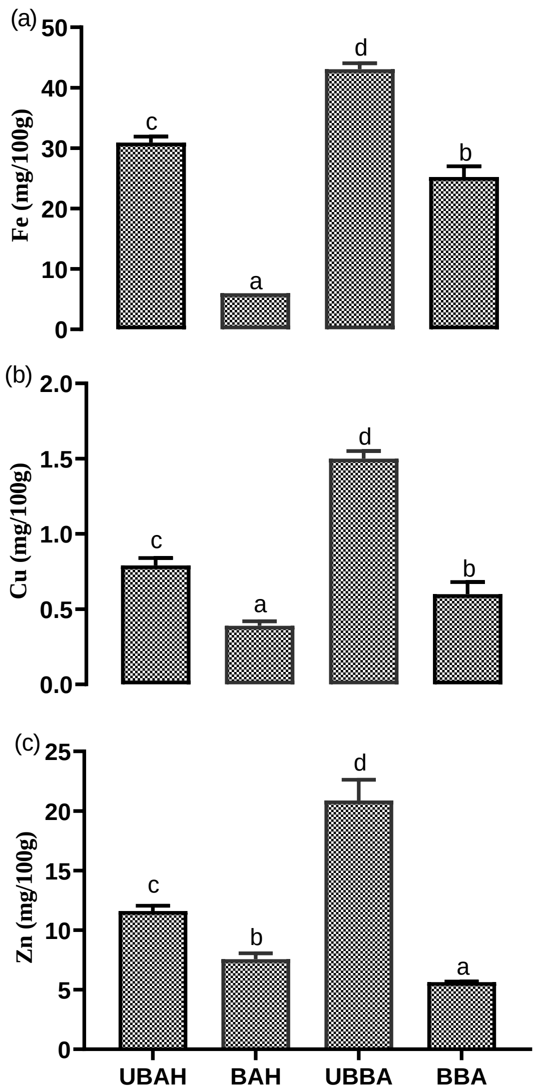

Figure 3: (a-c) Influence of blanching on transition element of *Basella alba* and *Amaranthus hybridus* leaves (a) Iron (b) Copper (c) Zinc. UBAH, unblanched *Amaranthus hybridus*; BAH, blanched *Amaranthus hybridus*; UBBA, unblanched *Basella alba*; blanched *Basella alba* BBA. Bars represent mean ± standard deviation. Bars with different letters are significantly different at $p < 0.05$.

An alteration of the concentrations of these minerals could have severe physiological penalties for the organism consuming the vegetables. This could be seen from the diverse functions of the minerals. The body cannot biosynthesize these minerals and must acquire them from diets. Calcium is a major structural component of bones and teeth, and essential in blood clotting, maintenance of blood pressure and acid-base balance in the blood, nerve regulation, muscle contraction, cellular metabolism, milk, and egg production, reduces colorectal adenomas and cholesterol (Cormick and Belizán, 2019).

Magnesium participates in hundreds of biochemical reactions in the body. It attenuates depression, type 2 diabetes, blood pressure, inflammation, migraines, insulin resistance, and premenstrual syndrome (Barbagallo *et al*., 2021). Iron is an essential element for most life on earth, including human beings. It is responsible for the formation of haemoglobin, myoglobin, and cytochrome, regulates body temperature, muscle activity, catecholamine metabolism, thyroid function, immune system, brain development, and function (Moustarah and Mohiuddin, 2020). Copper serves as a cofactor and/or a fundamental structure of many metalloenzymes (superoxide dismutase, ceruloplasmin, lysyl oxidase, cytochrome oxidase, and tyrosinase), reduces oxidative stress and cholesterol level, improves wound healing, maintain the immune system, participate in immune cells, collagen, mitochondria, and bone formation, and sustains the elasticity of blood vessels, which allows maintenance of blood pressure. Copper is crucial to the normal formation of the brain and nervous system throughout life. The human brain has over 100 billion neurons, copper connects them to 10 thousand other neurons (Hefnawy and Khaiat, 2015). Zn plays important roles in cell-mediated immunity, bone formation, tissue growth, brain function, healing, and repair, DNA synthesis, growth and development of tissues (skin, muscles, joints), antioxidant systems SOD and embryogenesis processes such as proliferation, differentiation, apoptosis, migration, maturation, myelination, synaptogenesis, and pruning (Roohani *et al*., 2013).

4. Conclusion

In conclusion, short time blanching significantly reduces the mineral content of *Basella alba* and *Amaranthus hybridus* leaves while also lowering phytate and oxalate, making them more accessible. As a result, additional processing strategies that merely lower the antinutrient content should be developed.

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