Comparative studies of antioxidants and amino-acids concentrations in the fruits of two cultivars of watermelon (*Citrullus lanatus* L.)

Funmilayo M. OLOYEDE¹*, Aisha A. BAKARE¹, Gbenga G. DARAMOLA²

¹Osun-State University, College of Agriculture, Department of Agronomy, Osogbo, Nigeria; funmilayo.loyede@yahoo.co.uk (*corresponding author); mary.oyode@uniosun.edu.ng
²Redeemer’s University, Department of Biochemistry, Ede, Osun-State, Nigeria; daramola20032003@yahoo.com

Abstract

Importance of fruits and vegetables in diet and health cannot be overemphasized. Cultivation practices can however influence the nutritional compositions of fruits and vegetables. Hence, field and laboratory experiments were carried out to determine and compare the antioxidants and amino-acids concentrations of a hybrid and an indigenous cultivar of watermelon in Osun State University, Teaching and Research Farm, Nigeria. Influence of NPK 15:15:15 fertilizer at different rates (0, 100, 200 and 300 kg/ha) was evaluated on ‘Kaolack’ (hybrid) and ‘Rothmas’ (local) cultivars of watermelon. 200 kg/ha of the fertilizer was found to bring out the optimal yield of both cultivars. Hence, at 0 kg/ha and 200 kg/ha, mature fruits of the cultivars were analyzed for the antioxidants and amino-acids using standard analytical methods. Antioxidant properties of ‘Rothmas’ were found to be significantly (P≤0.05) higher than those of ‘Kaolack’ with the exception of the radical scavenging ability and lycopene content of ‘Kaolack’. Fruits that received no fertilizer across the cultivars exhibited more antioxidant potency compared to fruits under 200 kg. Results of amino acids concentration showed that, threonine, serine, glutamic-acid, alanine, valine, phenylalanine and arginine in ‘Rothmas’ were significantly higher compared to the contents in ‘Kaolack’. Except for tryptophan, alanine, lysine and phenylalanine, fruits that received no fertilizer had higher contents of amino acids. Where fertilizer is needed to boost soil fertility, 100 kg/ha of NPK is recommended, this can be augmented with the organic nutrient or green manure if necessary, depending on the available nutrients in the soil before cultivation. This is very critical so as not to compromise the health-giving substances in the fruits. Local landraces of fruits and vegetables obviously have to be brought into limelight through research.

Keywords: cultivar; diet; health substances; protein; vegetable; watermelon

Introduction

Watermelon (*Citrullus lanatus*) belongs to the family Cucurbitaceae. It is native to Africa and was then introduced to Asia, Europe, and the Americas. Even though watermelon is commonly classified as a vegetable, it is botanically considered as a fruit and used primarily as a dessert (Chafik *et al.*, 2020). Watermelon contains carotenoid, a precursor of vitamin A which is very important to the body. β-carotene has a protective role.
against cancer (Halter, 1989) and coronary heart disease (Guoyao et al., 2009). Watermelon also contains antioxidants, flavonoids, lycopene, pyrofluenue, lutein, neurosporene, zeaxanthin, phytoene and cryptoxanthin (De Lannoy, 2001). Potassium is also available in watermelon, which helps in the control of blood pressure and to prevent stroke (IITA, 2013).

Agronomic practices such as fertilizer application (type and amount), cultivar selection, planting season/date, etc. however influence not only crop yield but also the health substances contained in fruits and vegetables (Oloyede et al., 2012; Oloyede et al., 2014; Oloyede et al., 2015). Hence, this study determined the effect of NPK fertilizer rates and cultivar types on the antioxidants and amino acids of Citrullus lanatus.

**Materials and Methods**

**Field study**

Field experiment was carried out from September to December, 2016 at the Teaching and Research Farm of Osun State University, Nigeria. The experimental design used was a two factorial experiments, as a randomized complete block design (RCBD), in three replicates. At the experimental site, the soil had total N of 1.58%, K was 4.25 cmol/kg, while P value evaluated in ppm was 19. Influence of NPK 15:15:15 fertilizer at different rates (0, 100, 200 and 300kg/ha) was evaluated on ‘Kaoalack’ (hybrid) and ‘Rothmas’ (local) cultivars of watermelon. 200 kg/ha of the fertilizer was found to bring out the optimal yield of both cultivars. Hence, at 0 kg/ha and 200 kg/ha, mature fruits of the cultivars were analyzed for the antioxidants and essential amino-acids using standard analytical methods.

**Determination of selected antioxidant properties**

The radical scavenging ability of the fruit extract was determined using the stable radical DPPH (2, 2-diphenyl-1-picrylhydrazyl hydrate) as described by (Brand-Williams et al., 1995). The reaction of DPPH with an antioxidant compound which can donate hydrogen, leads to its reduction (Blois, 1958). The change in color from deep violet to light yellow was measured spectrophotometrically at 517nm. The proanthocyanidin content of the extract was determined spectrophotometrically. Extracts were diluted to provide a spectrophotometric reading between 0.1 and 0.8 absorbance units. Carotenoid was measured using the method of GODWIN et al. (2015). 500 µl of testing sample is taken in a test tube and added with 5 ml of the stock solution. BHT (90 µg/ml) is employed as a positive control agent. The absorbance of the mixture is noted at 470 nm. The reaction mixture is then incubated for 2 h at 50 °C. After incubation the absorbance was measured again at 470 nm (t = 120 min). Total lycopene was measured according to the method of FISH et al. (2002). The method of determining the total phenolic content was described by Singleton and Rossi (1965) using the Folin-Ciocalteu’s phenol reagent which is an oxidizing reagent.

**Amino acids determination**

Amino acid content was determined based on the method described by Moore et al. (1958) using Technicon Sequential Multi-sample (TSM) Amino Acid Analyzer, Model GFL 1083 (Technicon Instruments Corporation), Germany and with nor leucine as an internal standard. The hydrolysate was vacuum dried to remove buffer solution before loading into the TSM. Compressed nitrogen was passed into the TSM to serve as a segmented stream flow of the amino acid which helped the analyzer to detect any amino acid found without mixing up the amino acids.
Results and Discussion

Regarding the antioxidant properties of the two cultivars of watermelon fruits, the radical scavenging ability of ‘Kaolack’ was 92.3% while that of ‘Rothmas’ was 78.9% (Table 1). Lycopene content was higher in ‘Kaolack’ compared to ‘Rothmas’. However, the phenol and proanthocyanidin were higher in ‘Rothmas’ compared to ‘Kaolack’. Both cultivars had same carotenoid content (0.07 g/100 g). Moreover, both cultivars of watermelon evaluated have abundant antioxidant properties. Oxidation, a chemical reaction that transfers electrons from a substance to an oxidizing agent, produces free radicals (Muller et al., 2007) and initiates chain reactions that may damage cells. Antioxidants terminate these chain reactions by removing free radical intermediates, inhibit other oxidation reactions, or enhance the endogenous antioxidant defenses of the organism (Lu et al., 2010). Consumption of fruits and vegetables which contain these compounds have been scientifically validated to help slowdown the aging process and reduce the risk factors of many diseases including cancer, heart disease, stroke, high blood pressure, cataracts, osteoporosis, diabetes and urinary tract infections (Liu et al., 2004; Barjesteh et al., 2007).

<table>
<thead>
<tr>
<th>Cultivars</th>
<th>DPPH (%)</th>
<th>Proanthocyanidin (Mg/ml)</th>
<th>Carotenoid (%)</th>
<th>Lycopene (Mg/kg)</th>
<th>Phenol (Mg GAE/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>‘Rothmas’</td>
<td>78.79*</td>
<td>9.53*</td>
<td>0.07*</td>
<td>3.08*</td>
<td>2.86*</td>
</tr>
<tr>
<td>‘Kaolack’</td>
<td>92.31*</td>
<td>6.92*</td>
<td>0.07*</td>
<td>4.16*</td>
<td>1.60*</td>
</tr>
<tr>
<td>LSD (0.05)</td>
<td>8.29</td>
<td>0.88</td>
<td>NS</td>
<td>0.26</td>
<td>0.06</td>
</tr>
</tbody>
</table>

Table 2 showed the effect of NPK fertilizer on the antioxidant properties of watermelon fruits. The radical scavenging abilities of the fruits that received 0 and 200 kg/ha were 88% and 83% respectively, though not statistically significantly different (P≤0.05). Proanthocyanidin and carotenoid contents did not differ statistically as well. Phenol and lycopene contents of the fruits that received no fertilizer were higher than those that received 200 kg/ha. Similar trends were observed in the antioxidant activities of pumpkin fruits. Application of 0 to 100 kg/ha of NPK 15:15:15 were recommended as opposed to higher rates (150-250 kg/ha) which were found to compromise the phenol, flavonoid, anthocyanin and proanthocyanidin concentrations of pumpkin fruits (Oloyede et al., 2012; Oloyede et al., 2014).

<table>
<thead>
<tr>
<th>NPK</th>
<th>DPPH (%)</th>
<th>Proanthocyanidin (Mg/ml)</th>
<th>Carotenoid (%)</th>
<th>Lycopene (Mg/kg)</th>
<th>Phenol (Mg GAE/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>83.04*</td>
<td>8.62*</td>
<td>0.09*</td>
<td>3.89*</td>
<td>2.69*</td>
</tr>
<tr>
<td>200</td>
<td>88.05*</td>
<td>7.82*</td>
<td>0.05*</td>
<td>3.34*</td>
<td>1.74*</td>
</tr>
<tr>
<td>LSD (0.05)</td>
<td>NS</td>
<td>0.88</td>
<td>NS</td>
<td>0.26</td>
<td>0.06</td>
</tr>
</tbody>
</table>

Table 3 revealed the effects of cultivars on the essential amino-acids concentrations (mg/100 g) in watermelon. Respective values of 2.07 and 2.19 of tryptophan in ‘Rothmas’ and ‘Kaolak’ cultivars were observed. ‘Kaolack’ cultivar had statistically higher concentrations of methionine (6.00), isoleucine (21.04), leucine (24.91) and lysine (58.54). Threonine and phenylalanine contents did not differ statistically in the two cultivars. Table 4 showed the effects of cultivars on the non-essential amino-acids concentrations (mg/100 g) in watermelon. ‘Rothmas’ had statistically higher glutamic acid content (75.08) while ‘Kaolack’ had statistically higher proline (29.03), tyrosine (2.48) and aspartic acid (49.22). Serine, glycine, alanine, cysteine, and arginine contents did not differ statistically in the two cultivars. Similar report was given on cultivar effects on snake tomato (Trichosanthes cucumerina) chemical compositions (Oloyede and Adebayo 2005).
Table 3. Cultivar effect on the essential amino acid concentration of watermelon on wet weight basis

<table>
<thead>
<tr>
<th>Cultivar</th>
<th>Tryptophan</th>
<th>Threonine</th>
<th>Valine</th>
<th>Methionine mg/100g</th>
<th>Isoleucine</th>
<th>Leucine</th>
<th>Phenylalanine</th>
<th>Lysine</th>
</tr>
</thead>
<tbody>
<tr>
<td>‘Rothmas’</td>
<td>2.07&lt;sup&gt;a&lt;/sup&gt;</td>
<td>34.64&lt;sup&gt;a&lt;/sup&gt;</td>
<td>21.14&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.54&lt;sup&gt;a&lt;/sup&gt;</td>
<td>19.60&lt;sup&gt;b&lt;/sup&gt;</td>
<td>23.36&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.17&lt;sup&gt;a&lt;/sup&gt;</td>
<td>57.41&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>‘Kaolack’</td>
<td>2.19&lt;sup&gt;c&lt;/sup&gt;</td>
<td>34.02&lt;sup&gt;c&lt;/sup&gt;</td>
<td>20.61&lt;sup&gt;c&lt;/sup&gt;</td>
<td>6.00&lt;sup&gt;c&lt;/sup&gt;</td>
<td>21.04&lt;sup&gt;c&lt;/sup&gt;</td>
<td>24.91&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.16&lt;sup&gt;c&lt;/sup&gt;</td>
<td>58.54&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>LSD (0.05)</td>
<td>0.01</td>
<td>2.06</td>
<td>0.21</td>
<td>0.14</td>
<td>0.66</td>
<td>0.99</td>
<td>0.02</td>
<td>0.93</td>
</tr>
</tbody>
</table>

Table 4. Cultivar effect on the non-essential amino acid concentration of watermelon on wet weight basis

<table>
<thead>
<tr>
<th>Cultivars</th>
<th>Serine</th>
<th>Glutamic acid</th>
<th>Proline</th>
<th>Glycine</th>
<th>Alanine mg/100g</th>
<th>Cysteine</th>
<th>Tyrosine</th>
<th>Arginine</th>
<th>Aspartic acid</th>
</tr>
</thead>
<tbody>
<tr>
<td>‘Rothmas’</td>
<td>20.97&lt;sup&gt;a&lt;/sup&gt;</td>
<td>75.08&lt;sup&gt;a&lt;/sup&gt;</td>
<td>27.91&lt;sup&gt;b&lt;/sup&gt;</td>
<td>13.07&lt;sup&gt;a&lt;/sup&gt;</td>
<td>22.38&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>2.22&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.36&lt;sup&gt;a&lt;/sup&gt;</td>
<td>62.99&lt;sup&gt;a&lt;/sup&gt;</td>
<td>46.99&lt;sup&gt;ab&lt;/sup&gt;</td>
</tr>
<tr>
<td>‘Kaolack’</td>
<td>20.50&lt;sup&gt;b&lt;/sup&gt;</td>
<td>73.52&lt;sup&gt;b&lt;/sup&gt;</td>
<td>29.03&lt;sup&gt;c&lt;/sup&gt;</td>
<td>13.67&lt;sup&gt;c&lt;/sup&gt;</td>
<td>22.10&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.22&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.48&lt;sup&gt;c&lt;/sup&gt;</td>
<td>63.12&lt;sup&gt;c&lt;/sup&gt;</td>
<td>49.22&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>LSD (0.05)</td>
<td>0.64</td>
<td>1.24</td>
<td>0.93</td>
<td>0.97</td>
<td>0.59</td>
<td>0.04</td>
<td>0.02</td>
<td>0.93</td>
<td></td>
</tr>
</tbody>
</table>

Table 5 revealed the effects of NPK 15:15:15 fertilizer on the essential amino-acids concentrations (mg/100 g) in watermelon. Respective values of 55.64 and 60.31 of lysine in fruits under 0 and 200 kg/ha of NPK were observed. Fruits under 0 kg/ha of fertilizer had statistically higher concentrations of methionine (6.10), isoleucine (20.77), leucine (24.89) and valine (21.14), while tryptophan, phenylalanine and lysine with respective values of 2.15, 2.21 and 60.31 were statistically higher in fruits under 200 kg/ha. Threonine content was indifferent to fertilizer application.

Table 5. NPK effect on the essential amino acid concentration of watermelon on wet weight basis

<table>
<thead>
<tr>
<th>NPK</th>
<th>Tryptophan</th>
<th>Threonine</th>
<th>Valine</th>
<th>Methionine mg/100g</th>
<th>Isoleucine</th>
<th>Leucine</th>
<th>Phenylalanine</th>
<th>Lysine</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>2.11&lt;sup&gt;b&lt;/sup&gt;</td>
<td>33.77&lt;sup&gt;b&lt;/sup&gt;</td>
<td>21.14&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6.10&lt;sup&gt;a&lt;/sup&gt;</td>
<td>20.77&lt;sup&gt;b&lt;/sup&gt;</td>
<td>24.89&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.11&lt;sup&gt;b&lt;/sup&gt;</td>
<td>55.64&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>200</td>
<td>2.15&lt;sup&gt;a&lt;/sup&gt;</td>
<td>34.89&lt;sup&gt;a&lt;/sup&gt;</td>
<td>20.61&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.44&lt;sup&gt;b&lt;/sup&gt;</td>
<td>19.88&lt;sup&gt;b&lt;/sup&gt;</td>
<td>23.34&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.21&lt;sup&gt;c&lt;/sup&gt;</td>
<td>60.31&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
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<td>LSD (0.05)</td>
<td>0.01</td>
<td>2.06</td>
<td>0.21</td>
<td>0.14</td>
<td>0.66</td>
<td>0.99</td>
<td>0.02</td>
<td>0.93</td>
</tr>
</tbody>
</table>

Table 6 showed the effects NPK 15:15:15 fertilizer on the non-essential amino-acids concentrations (mg/100 g) in watermelon. Fruits under 0 kg/ha of fertilizer had statistically higher concentrations of serine (21.44), glutamic acid (75.37), proline (29.62), cysteine (2.28) and tyrosine (2.51). Alanine (23.08) and aspartic acid (48.30) were however statistically higher in fruits under 200 kg/ha. Arginine and glycine contents were not influenced statistically fertilizer application. Amino acids are organic compounds composed of nitrogen, carbon, hydrogen and oxygen, along with a variable side chain group. The body needs different amino acids to grow and function properly.

Table 6. NPK effect on the non-essential amino acid concentration of watermelon on wet weight basis

<table>
<thead>
<tr>
<th>NPK</th>
<th>Serine</th>
<th>Glutamic acid</th>
<th>Proline</th>
<th>Glycine mg/100g</th>
<th>Alanine</th>
<th>Cysteine</th>
<th>Tyrosine</th>
<th>Arginine</th>
<th>Aspartic acid</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>21.44&lt;sup&gt;a&lt;/sup&gt;</td>
<td>75.37&lt;sup&gt;a&lt;/sup&gt;</td>
<td>29.62&lt;sup&gt;b&lt;/sup&gt;</td>
<td>13.38&lt;sup&gt;a&lt;/sup&gt;</td>
<td>21.40&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.28&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.51&lt;sup&gt;a&lt;/sup&gt;</td>
<td>64.75&lt;sup&gt;c&lt;/sup&gt;</td>
<td>47.91&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>200</td>
<td>20.02&lt;sup&gt;b&lt;/sup&gt;</td>
<td>73.23&lt;sup&gt;b&lt;/sup&gt;</td>
<td>27.32&lt;sup&gt;c&lt;/sup&gt;</td>
<td>13.36&lt;sup&gt;c&lt;/sup&gt;</td>
<td>23.08&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.16&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.33&lt;sup&gt;c&lt;/sup&gt;</td>
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<td>48.30&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>LSD (0.05)</td>
<td>0.64</td>
<td>1.24</td>
<td>0.93</td>
<td>0.97</td>
<td>0.59</td>
<td>0.04</td>
<td>0.02</td>
<td>0.93</td>
<td>0.7</td>
</tr>
</tbody>
</table>

Note: Means on the same column with different letters are significantly different (P<0.05).
NS: Not significantly different at (P<0.05).
Unlike nonessential amino acids, essential amino acids can’t be made by the body and must be obtained through diets. When protein is eaten, it is broken down into amino acids, which are then used to help the body with various processes such as building muscle and regulating immune function (Health line, 2021). According to Oloyede et al. (2015) agronomic input especially fertilizer types and rates influence the health substances in fruits and vegetables and there is a need to apply moderate dose to boost the yield while the health substances in the plants remain uncompromised.

Conclusions

Both ‘Rothmas’ and ‘Kaolack’ cultivars of watermelon evaluated have abundant antioxidant properties. There is a need to bring the local landraces of fruits and vegetables obviously into limelight through research. 200 kg/ha of NPK 15:15:15 limited some of the concentrations of antioxidants and amino-acids parameters evaluated at the location where this study was carried out. Where fertilizer is needed to boost soil fertility, 100 kg/ha of NPK is recommended, this can be augmented with the organic nutrient or green manure if necessary, depending on the available nutrients in the soil before cultivation. This is very critical so as not to compromise any of the health-giving substances in the fruits.

Authors’ Contributions

Conceptualization: F.M.; Data curation; Formal analysis; Investigation; Methodology: F.M and A.B.; Project administration: F.M.; Laboratory analyses: G.G.; Resources: The 3 authors; Supervision F.M.; Writing: F.M. and A.B.; Writing review and editing: The 3 authors. All authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

References


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