Production and Evaluation of Biscuits Made From Wheat Flour and Toasted Watermelon Seed Meal as Fat Substitute

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Received: October 15, 2017    Accepted: November 3, 2017    Online Published: September 10, 2018
doi:10.5539/jfr.v7n5p112    URL: https://doi.org/10.5539/jfr.v7n5p112

Abstract

Production and evaluation of biscuits made from wheat flour and toasted watermelon seed meal as fat substitute was studied. Extracted watermelon seeds were toasted and grounded into seed meal, which were later used to formulate composite flour blends in the ratio of 10:90 (MW₁), 20:80 (MW₂), 30:70 (MW₃), 40:60 (MW₄) and 50:50 (MW₅) of watermelon seed meal: wheat flour respectively, while 100% wheat flour served as control (W₀). Each blend was used to produce biscuits adopting robbing process. The functional properties of flour blends, proximate composition and physical properties of the biscuit samples were determined. Sensory qualities of the biscuits were evaluated by 20 panelists. The accepted composite biscuit samples were stored on a shelf for 6 weeks and analyzed for moisture and total viable count. Data obtained were analyzed statistically (P≤0.05) for ANOVA, and means separated using Fisher’s LSD Test. The results of the functional properties showed variations in behaviour. There were no significant differences (P≤0.05) in pH, swelling index, viscosity and gelation temperature of the flour blends while significant differences (P≤0.05) existed in bulk density, water absorption capacity and wettability. There were significant differences (P≤0.05) in protein, fat, crude fibre, ash and carbohydrate for both flour blends and biscuit samples respectively. The addition of toasted watermelon seed meal increased protein, fat, crude fibre and ash contents while carbohydrate content decreased with higher substitution levels of wheat flour with the toasted watermelon seed meal. Moisture content ranged from 10-11 % (flour) and 6.25 -10% (biscuit). Significant differences (P≤0.05) were recorded in thickness, spread ratio and break strength within the samples. The result of sensory evaluation showed that W₁ was significantly different (P≤0.05) in taste, aroma and overall acceptance.

However, MW₁ and MW₅ were significantly different in texture, but in terms of crispiness, MW₁ and MW₃ were preferred. Moisture content of the preferred samples (MW₁ and MW₃) stored for 6 weeks increased from 6.25-11.75 % (MW₁) and 6.75 – 11.0% (MW₃) while the viable microbial counts ranged from 2.0 x 10²-6.8 x 10⁸ cfu/g for bacteria and 1.8 x 10⁵-7.4 x 10⁵ cfu/g for mould respectively.

Keywords: wheat flour, watermelon seeds, biscuits, acceptability, moisture

1. Introduction

The consumption of cereal snack foods such as biscuits (also known as cookies), wafers and short bread has become very popular especially among children. Biscuits possess several attractive features including wider consumption base, relatively long shelf life and good eating quality (Banureka and Mahendran, 2009).

Biscuit and biscuit-like products have been made and eaten by man for many hundreds if not thousands of years and their good eaten quality makes them attractive for protein fortification and nutritional improvements, particularly in children feeding programmes, for the elderly and low income groups (Banureka and Mahendran, 2009).

Wheat flour is the main ingredient for biscuit production, though imported; it is a carbohydrate based food raw material and also lack some nutrients.

Composite flour has been defined as a mixture of different ratios of non-wheat flour from cereals, legumes, roots and tubers with or without wheat flour. In many countries, biscuits are prepared with fortified or composite flour to increase their nutritive value (Hasmadi et al., 2014). Enrichment of cereal-based foods with other protein sources such as oilseeds and legumes has received considerable attention (Ayo and Olawale, 2003). This is
because oilseeds and legume proteins are high in lysine- an essential limiting amino acid in most cereals including wheat (Banureka and Mahendran, 2009). The usefulness of grain-oilseeds in developing high protein foods in meeting the needs of the vulnerable group of the population is now well recognized, and several high protein-energy foods have been developed industrially in different parts of the world (Mooriya, 2003).

Watermelon (Citrullus lanatus) is a typical fruit from the family of cucurbitaceae grown in the warmer part of the world whose seeds are underutilized. The seeds are rich source of dietary fibre, (5%) high in protein (35%) and fat (50%) ,it is a rich source of magnesium, calcium, potassium, iron, phosphorus and zinc (Odibo et al., 2012) and with excellent functional properties which have been found to be effective in baking (El-Adawy and Taha, 2001).

Watermelon seeds are known to be highly nutritional: they are rich sources of protein, B vitamins, Minerals (such as magnesium, potassium, phosphorus, sodium, iron, zinc, manganese and copper) and fat among others as well as phytochemicals (Braide et al., 2012). Watermelon seeds are rich for strong bones and teeth as well as hemoglobin formation. Fat in a biscuit formulation has a multifaceted function. It is the principal ingredient responsible for tenderness, eaten quality, keeping quality, grain and texture, and adding richness to biscuits (O’Brien, 2003). The food industry is primarily driven by consumer health trends. A present day dietary concern is the consumption of fat and sugar, which have been found to be associated with an increase in the incidence of obesity and related health problems, including coronary heart disease (CHD) (Boobier et al., 2006).

Biscuits have previously been highlighted in a report by Willet et al (1993) as being significantly associated with an increased risk of CHD. Nisbett et al (1986) noted that biscuit market is dominated by short dough biscuits having fat levels in excess of 20%. However, Mensink and Katan (1987) stated that the replacement of saturated or trans-unsaturated fat with unhydrogenated unsaturated fats is associated with larger reductions in risk factors associated with CHD.

The aim of this work therefore is to produce biscuits from blends of wheat flour and toasted watermelon seeds for fat substitution.

2. Materials and Methods

2.1 Material Procurement

Watermelon fruits, wheat, sugar, margarine, milk powder, baking powder, salt, egg and vanilla powder were purchased from Owerri main market, Imo State, Nigeria.

2.2 Methods

2.2.1 Production of Watermelon Seed Flour

Toasted watermelon seed flour was prepared by deseeding watermelon pods, the brown seeds were washed (with potable water), drained and sun dried. The seeds were toasted in a shallow pan at 130°C for 20minutes, cooled, milled with harmer mill and sieved through 60 mesh sieve to obtain a fine flour. The watermelon seed flour was sealed in a cellophane bag and stored at room temperature (25°C) for further use. The modified method of Ubbor and Akobundu (2009) as shown in (figure 1) was used in the production of toasted watermelon seed flour.
2.2.2 Formulation of Flour Blend

Five blends were prepared by mixing toasted whole watermelon seed flour with wheat flour using an electric blender in the percentage ratio of 10:90, 20:80, 30:70, 40:60 and 50:50 respectively while hundred percent (100%) wheat flour served as the control.

Table 1. Formulation of flour blend

<table>
<thead>
<tr>
<th>Blends</th>
<th>Toasted watermelon seed flour (%) (M)</th>
<th>Wheat flour (%) (W)</th>
</tr>
</thead>
<tbody>
<tr>
<td>W₁</td>
<td>-</td>
<td>100</td>
</tr>
<tr>
<td>MW₁</td>
<td>10</td>
<td>90</td>
</tr>
<tr>
<td>MW₂</td>
<td>20</td>
<td>80</td>
</tr>
<tr>
<td>MW₃</td>
<td>30</td>
<td>70</td>
</tr>
<tr>
<td>MW₄</td>
<td>40</td>
<td>60</td>
</tr>
<tr>
<td>MW₅</td>
<td>50</td>
<td>50</td>
</tr>
</tbody>
</table>

2.2.3 Biscuit Preparation

Biscuits were prepared according to the method of AOAC (2000) with some modifications in the recipe. The dry ingredients (flour, sugar, salt, and baking powder) were thoroughly mixed in a bowl by hand for 3 min. Egg and water were then added and the mixture kneaded. The batter was rolled and cut with a biscuit cutter. The cut dough was placed on oil greased baking trays, leaving 25 mm spaces in between and was baked at 200°C for 25 min in the baking oven. Following baking, the biscuits were cooled at ambient temperature (28 ± 2°C) prior to subsequent analysis and sensory evaluation. Ingredients used for biscuit making were flour (200g), sugar (60g), milk powder (30g), egg (30g), baking powder (1.5g), salt (1.5g), vanilla powder (4 drops), water (15ml), while 80g of magarine was added to the control only. The procedure is shown in Figure 2.
2.2.4 Determination of Functional Properties
Bulk density, pH, emulsion capacity, wettability, viscosity, water absorption capacity, oil absorption capacity, swelling index and gelation temperature were analyzed as described by Onwuka (2005) and Ukpabi and Ndimele (1990).

2.2.5 Determination of Proximate Composition
Moisture content, ash content, crude fat, crude protein, crude fibre and carbohydrate content of the flour sample were carried out using AOAC(2000) methods.

2.2.6 Physical Analysis of Biscuits Produced
The weight and diameter of the baked biscuit were determined by weighing on a weighing balance and measuring with a calibrated ruler, respectively (Ayo et al, 2007). The thickness was measured according to AOAC method 10-50D (AOAC, 2000). The spread ratio was determined using the method of Gomez et., al. (1997). Three rows of five well-formed biscuit were made and the height measured. Also the same were arranged horizontally edge to edge and sum diameter measured. The spread ratio was calculated as diameter/ height. The break strength of the biscuit was determined according to Okaka and Isieh (1990) method. Biscuit of known thickness (0.4cm) was placed between two parallel wooden bars (3.0cm apart). Weights were added on the biscuit until the biscuit snapped. The least weight that caused the breaking of the biscuit was regarded as the break strength of the biscuit.

2.2.7 Sensory Evaluation
Sensory evaluations of the biscuits were determined using twenty-member panelist consisting of staff and students of the Department of Food Science and Technology, Federal University of Technology, Owerri, Nigeria. The panelists were either regular or occasionally consumers of biscuits and were not allergic to any food. Biscuits samples prepared from each flour blend were presented in coded white microwavable plastic container. The order of presentation of samples to the panelists was randomized. Sachet water was provided to rinse the mouth between evaluations. The panelists were instructed to evaluate the coded samples for taste, aroma, colour, texture, crispiness and overall acceptability. Each sensory attribute was rated on a 9-point Hedonic scale (for taste, aroma and overall acceptability, 1 = disliked extremely, 5 = neither like nor dislike, while 9 = liked extremely. For colour, 1 = extremely white, 5 = neither brown nor white, while 9 = extremely brown. For texture:
1 = extremely marshy, 5 = neither strong nor soft, while 9 = extremely strong and firm. And for crispiness, 1 = extremely soft and mealy, 5 = neither crispy nor soft, while 9 = extremely crispy and grainy (Ihekoronye and Ngoddy, 1985).

2.2.8 Microbiological Analysis of Biscuits Produced

Total viable bacteria counts and fungi counts (yeasts and moulds) were carried out on biscuit samples to determine the microbial load of the samples as described by Roberts and Greenwood, 2003. One (1g) gram of each biscuit sample was prepared by mashing and mixing in 9ml of distilled water. Serial dilutions up to 10⁹ were carried out and 0.1ml each was poured in the nutrient agar (NA) media petri dish, and potato dextrose agar (PDA) petri dish for the enumeration of aerobic viable bacteria and fungi respectively. The NA petri dishes were incubated at the temperature of 37°C for 24 – 48 hours while PDA petri dishes were incubated at 30°C for up to 5 days. The colonies were then counted and expressed as colony forming units per gram (cfu/g) of samples. This was done on Weeks 1, 3 and 6 of storage periods.

3. Results and Discussion

3.1 Functional properties

The result of the functional properties of the flour blends are shown in Table (2). pH of the flour samples ranged from 6.528 to 6.670. MW1 had the highest value (6.670) while W1 had the least (6.528). There was no significant difference, however, pH increased with higher substitution with toasted watermelon seed meal. The pH values of the flour samples were > 6.5 being considered non acidic food product.

Table 2. Functional properties of the flour blends

<table>
<thead>
<tr>
<th>Sample</th>
<th>MW</th>
<th>pH</th>
<th>BD (g/ml)</th>
<th>WAC (g/g)</th>
<th>OAC (g/g)</th>
<th>SI (ml)</th>
<th>EC (%)</th>
<th>Wet (Sec)</th>
<th>Viscosity (%)</th>
<th>Gelation temp (°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>W1</td>
<td>6.528±0.05</td>
<td>0.796±0.00</td>
<td>1.217±0.00</td>
<td>1.454±0.02</td>
<td>1.056±0.08</td>
<td>1.987±0.02</td>
<td>51.00±6.48</td>
<td>5.80±0.98</td>
<td>67±2.45</td>
<td></td>
</tr>
<tr>
<td>MW1</td>
<td>6.619±0.04</td>
<td>0.771±0.07</td>
<td>1.213±0.00</td>
<td>1.626±0.03</td>
<td>0.875±0.09</td>
<td>2.000±0.00</td>
<td>34.00±3.27</td>
<td>5.95±0.20</td>
<td>63.67±2.87</td>
<td></td>
</tr>
<tr>
<td>MW2</td>
<td>6.646±0.05</td>
<td>0.798±0.07</td>
<td>1.287±0.00</td>
<td>1.812±0.03</td>
<td>0.849±0.26</td>
<td>2.000±0.00</td>
<td>30.33±7.04</td>
<td>4.95±1.27</td>
<td>64±1.63</td>
<td></td>
</tr>
<tr>
<td>MW3</td>
<td>6.690±0.04</td>
<td>0.789±0.02</td>
<td>1.280±0.01</td>
<td>1.889±0.08</td>
<td>0.786±0.01</td>
<td>2.033±0.00</td>
<td>28.67±1.89</td>
<td>6.35±1.10</td>
<td>63±0.82</td>
<td></td>
</tr>
<tr>
<td>MW4</td>
<td>6.667±0.04</td>
<td>0.717±0.07</td>
<td>1.303±0.00</td>
<td>1.626±0.03</td>
<td>0.774±0.06</td>
<td>1.973±0.04</td>
<td>16.00±1.63</td>
<td>5.85±1.43</td>
<td>61±2.45</td>
<td></td>
</tr>
<tr>
<td>MW5</td>
<td>6.679±0.05</td>
<td>0.771±0.00</td>
<td>1.357±0.00</td>
<td>1.531±0.04</td>
<td>0.758±0.06</td>
<td>2.000±0.00</td>
<td>12.00±1.41</td>
<td>6.15±0.61</td>
<td>62±1.63</td>
<td></td>
</tr>
<tr>
<td>LSD</td>
<td>-</td>
<td>0.019</td>
<td>0.012</td>
<td>0.094</td>
<td>-</td>
<td>0.142</td>
<td>9.349</td>
<td>-</td>
<td>-</td>
<td></td>
</tr>
</tbody>
</table>

Where pH = Hydrogen ion concentration, BD = Bulk density, WAC = Water absorption capacity, OAC = Oil absorption capacity, SI = Swelling index, EC = Emulsion capacity, Wet = wettability.

Mean ± Standard deviation of triplicate determination
Means with the same superscripts within a column are not significantly different (P < 0.05)

W1 = Wheat flour (100%)

MW1 = watermelon seed meal/Wheat flour (10:90)

MW2 = Watermelon seed meal/wheat flour (20:80)

MW3 = Watermelon seed meal/wheat flour (30:70)

MW4 = Watermelon seed meal/wheat flour (40:60)

MW5 = Watermelon seed meal/wheat flour (50:50)

W1 and MW2 were not significantly different. WAC is the ability of a product to associate with water under limiting condition (Singh, 2001). Water absorption capacity (WAC) was found to be higher in MW5 (1.357g/g) while MW1 had the least (1.213g/g). Significant difference (P≤ 0.05) existed within the samples. W1 and MW2 were not significantly different. Obasi et al (2012), reported that the water absorption capacity of wheat flour was 2.04g/g which was higher than the value obtain in this work (1.217-1.357g/g). This could be due to variation in cultivar, location of cultivation etc. It has been suggested that flours with high WAC will be very useful in bakery products as this could prevent staling by reducing moisture loss (Obatolu et al., 2007).

Oil Absorption Capacity (OAC) values ranged from 1.389g/g to 1.812g/g. Significant difference (p≤0.05) occurred among the flour samples. MW2 was significantly different from the rest. Oil absorption capacity increased with increase in level of substitution from W1-MW2 (1.454g/g to 1.812g/g). The oil absorption capacity of the flour blends suggests that they may be useful in food preparation that involves mixing like bakery products where oil is an important ingredient. The oil absorption capacity of the flour blends increased with increase in protein content since the protein in foods influences fat absorption. OAC is the ability of flour to absorb oil, oil act as flavour retainer, and improves moth feel. This shows that sample MW2 could retain the biscuit flavour.

There was no significant difference (p≤0.05) among the flour samples in swelling index, however, the values
ranged from 0.758ml to 1.056ml. W<sub>i</sub> had the highest swelling index while MW<sub>5</sub> had the lowest. Observation indicated that swelling index decreased with higher substitution with toasted watermelon seed meal and could be due to higher protein content in watermelon seed meal. The presence of protein bodies around starch granules (due to increased protein content) may restrict granule swelling and hence, reduce the susceptibility to enzymatic attack.

Emulsion Capacity (EC) values ranged from 1.973 to 2.033. This is suggesting that the flour blends may be good emulsifying agent. Increase in protein content with increase in substitution level aid in formulation and stabilization of emulsion (Ababy et al., 1998). There was no significant difference among the flour samples. Emulsion capacity denotes the maximum amount of oil that can be emulsified by flour dispersion (Oluwalana and Oluwamukami, 2011). Brandt (1996), reported that emulsifiers help retard staling in baked goods, thereby prolonging shelf life. Wettability decreased with increased in substitution. W<sub>i</sub> had the highest (51sec) while MW<sub>3</sub> had the lowest (12sec). Significant difference (p≤0.05) occurred among the samples. W<sub>i</sub> was significantly different from the rest. According to Oti and Akobundu (2008), the lower the level of denatured protein in the starch the slower it takes to get wetted or imbibe water.

No significant difference occurred in viscosity among the flour samples, however, MW<sub>3</sub> had the highest value (6.35%) while MW<sub>2</sub> had the least (4.95%). Viscosity is an indication of viscous load (opposition to flow) likely to be encountered during mixing (Maziya-Dixon et al., 2007). Gelation temperature value was highest in W<sub>i</sub> (67°C) while MW<sub>4</sub> had the least (61°C) though there was no significant difference among the samples. Gels primarily enhance the body and texture of a food product.

3.2 Proximate Composition

The results for proximate composition of flour blends and biscuit samples are shown in Table (3) and (4) respectively. Moisture, protein, fat, ash and carbohydrate contents of the flour samples ranged from 10 – 11%, 14.15 – 23.25%, 1.25 – 9.25%, 1.3 – 4.18%, 1.6 – 3.55% and 49.77 – 70.70% respectively. The moisture content of W<sub>i</sub> (11%) was highest while the rest samples were below 11%. No significant difference was observed among the flour samples even though addition of toasted watermelon seed meal decreased the moisture content. The moisture contents of the composite flours were slightly above moisture level (below 10%) recommended for safe keeping of flour samples. Moisture content of biscuit samples range from 6.25 – 10.00%. MW<sub>3</sub> had the highest value, followed by MW<sub>2</sub> even though there was no significant difference among the samples. The highest value obtained in MW<sub>5</sub> could be due to variation in the oven heat distribution. Smith (1972), established that total moisture content for biscuits should not exceed 14% and that 5% is the best. Therefore, the moisture contents of the biscuits were within the acceptable level.

MW<sub>3</sub> had the highest value in protein, fat, crude fibre, and ash contents for the flour samples, while W<sub>i</sub> had the least. There were significant differences (p≤0.05) among the flour samples in the parameters mention above. W<sub>i</sub> and MW<sub>2</sub> were each significantly different from the rest of the samples respectively. Protein, fat, crude fibre, and ash contents increased with higher substitution with toasted watermelon seed meal. This is expected as watermelon seed is high in protein, fat (oil), crude fibre and ash (El-Adawy and Taha, 2001).

Table 3. Proximate Composition of the flour blends from toasted watermelon seed meal (M) and wheat flour (W)

<table>
<thead>
<tr>
<th>Flour blends</th>
<th>Moisture(%)</th>
<th>Protein(%)</th>
<th>Fat(%)</th>
<th>Crude fibre(%)</th>
<th>Ash(%)</th>
<th>Carbohydrate(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>W&lt;sub&gt;i&lt;/sub&gt;</td>
<td>11.00 ± 0.00</td>
<td>14.15 ± 0.35</td>
<td>1.25 ± 0.25</td>
<td>1.30 ± 0.100</td>
<td>1.60 ± 000</td>
<td>70.70 ± 0.00</td>
</tr>
<tr>
<td>MW&lt;sub&gt;1&lt;/sub&gt;</td>
<td>10.50 ± 0.00</td>
<td>18.11 ± 0.10</td>
<td>2.75 ± 0.25</td>
<td>1.77 ± 0.015</td>
<td>2.35 ± 0.15</td>
<td>64.52 ± 0.01</td>
</tr>
<tr>
<td>MW&lt;sub&gt;2&lt;/sub&gt;</td>
<td>10.75 ± 0.25</td>
<td>18.70 ± 0.20</td>
<td>4.60 ± 0.00</td>
<td>2.35 ± 0.050</td>
<td>2.60 ± 0.10</td>
<td>61.00 ± 0.00</td>
</tr>
<tr>
<td>MW&lt;sub&gt;3&lt;/sub&gt;</td>
<td>10.25 ± 8.25</td>
<td>19.33 ± 0.08</td>
<td>7.00 ± 0.00</td>
<td>3.15 ± 0.050</td>
<td>3.00 ± 0.00</td>
<td>57.27 ± 0.01</td>
</tr>
<tr>
<td>MW&lt;sub&gt;4&lt;/sub&gt;</td>
<td>10.00 ± 0.50</td>
<td>20.19 ± 0.14</td>
<td>8.00 ± 0.00</td>
<td>3.60 ± 0.100</td>
<td>3.25 ± 0.05</td>
<td>54.96 ± 0.01</td>
</tr>
<tr>
<td>MW&lt;sub&gt;5&lt;/sub&gt;</td>
<td>10.00 ± 0.50</td>
<td>23.25 ± 0.10</td>
<td>9.25 ± 0.25</td>
<td>4.18 ± 0.025</td>
<td>3.55 ± 0.05</td>
<td>49.77 ± 0.01</td>
</tr>
<tr>
<td>LSD</td>
<td>-0.633</td>
<td>0.612</td>
<td>0.227</td>
<td>0.274</td>
<td>0.014</td>
<td></td>
</tr>
</tbody>
</table>

Mean ± SD of duplicate determination
Means with the same superscript in column are not significantly different (P≤0.05)
W<sub>i</sub>: Wheat flour (100%)
MW<sub>i</sub>: Watermelon seed meal/wheat flour (20:80)
MW<sub>2</sub>: Watermelon seed meal/wheat flour (30:70)
MW<sub>3</sub>: Watermelon seed meal/wheat flour (40:60)
MW<sub>5</sub>: Watermelon seed meal/wheat flour (50:50)
Table 4. Proximate Composition of the Biscuits Produced

<table>
<thead>
<tr>
<th>Biscuits</th>
<th>Moisture(%)</th>
<th>Protein(%)</th>
<th>Fat(%)</th>
<th>Crude fibre(%)</th>
<th>Ash(%)</th>
<th>Carbohydrate(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>W₁</td>
<td>6.50 ± 0.50</td>
<td>12.00 ± 0.50</td>
<td>23.25 ± 0.25</td>
<td>1.45 ± 0.05</td>
<td>1.79 ± 0.02</td>
<td>55.01 ± 0.01</td>
</tr>
<tr>
<td>MW₁</td>
<td>6.25 ± 0.75</td>
<td>15.25 ± 0.25</td>
<td>4.00 ± 0.00</td>
<td>2.30 ± 0.10</td>
<td>2.49 ± 0.02</td>
<td>69.71 ± 0.01</td>
</tr>
<tr>
<td>MW₂</td>
<td>7.67</td>
<td>17.25 ± 0.25</td>
<td>5.25 ± 0.25</td>
<td>3.55 ± 0.25</td>
<td>3.10 ± 0.10</td>
<td>60.85 ± 0.01</td>
</tr>
<tr>
<td>MW₃</td>
<td>6.75 ± 1.25</td>
<td>17.68 ± 0.18</td>
<td>8.25 ± 0.25</td>
<td>3.60 ± 0.10</td>
<td>3.50 ± 0.10</td>
<td>60.22 ± 0.01</td>
</tr>
<tr>
<td>MW₄</td>
<td>6.50 ± 1.00</td>
<td>19.25 ± 0.25</td>
<td>9.25 ± 0.25</td>
<td>4.20 ± 0.10</td>
<td>4.00 ± 0.00</td>
<td>56.80 ± 0.00</td>
</tr>
<tr>
<td>MW₅</td>
<td>7.75 ± 0.25</td>
<td>21.25 ± 0.25</td>
<td>11.25 ± 0.25</td>
<td>4.4 ± 0.2</td>
<td>4.60 ± 0.05</td>
<td>50.75 ± 0.01</td>
</tr>
<tr>
<td>LSD</td>
<td>1.029</td>
<td>0.790</td>
<td>0.519</td>
<td>0.213</td>
<td>0.016</td>
<td></td>
</tr>
</tbody>
</table>

Mean ± SD of duplicate determination
Means with the same superscript in column are not significantly different (P ≥ 0.05)

W₁: Wheat flour (100%)
MW₁: Watermelon seed meal/wheat flour (20:80)
MW₂: Watermelon seed meal/wheat flour (30:70)
MW₃: Watermelon seed meal/wheat flour (40:60)
MW₄: Watermelon seed meal/wheat flour (50:50)

Proteins are the body’s building blocks and foods that are rich in protein are known to reduce protein energy malnutrition. MW₅ (21.25%) was highest in protein for the biscuit samples while W₁ (12%) had the least. Significant difference (p≤0.05) occurred within the samples however, protein increased with increase in substitution.

Fat content of biscuit samples was found to be highest in W₁ (23.25%) while MW₁ (4%) was the least. Significant difference (p≤0.05) existed within the biscuits samples. However, fat increased with increase in substitution. Fat gives flavour and soft texture to foods; nevertheless, high levels of fat are undesirable in food products because they could lead to rancidity in foods (Ihekonye and Ngoddy, 1985). Furthermore, eating foods rich in trans fats present in bakery fats creates inflammation, which is linked to heart disease, stroke, diabetes, and other chronic health conditions (http://www.health.harvard.edu).

Crude fibre of biscuit samples was found to be highest in MW₃ (4.4%) while W₁ (1.45%) had the least. Significant difference (p≤0.05) occurred among the samples. The fibre contents of the biscuits were within the recommended range of not more than 5g dietary fibre / 100g of dry matter (FAO/WHO, 1994). Dietary fibre has a protective action against various disorders such as diabetes mellitus, cardiovascular diseases, constipation, appendicitis, hemorrhoids, and colon cancer (Mervat, 2011; Bhawna et al., 2013).

Biscuit could enhance the intake of minerals, significant amount of vitamin C, minerals, fats, starch, and riboflavin have been obtained from watermelon seed (Erhierhe and Ekene, 2013); as ash is an index to determine mineral constituents in food. Highest ash content of biscuit samples was found in MW₅ (4.6%) while W₁ (1.79) had the least. There was significant difference (p≤0.05) among the samples. This result showed increase in ash with higher substitution and could be attributed to addition of watermelon seed. Similar result was obtained by Ubbor and Akobundu (2009) from cookies produced from composite flour blends of watermelon seed flour/wheat flour. Carbohydrate contents was highest (70.70%) in W₁, while MW₅ (49.77%) had the least. Significant difference (p≤0.05) existed within the flour samples. The carbohydrate contents of composite flours decreased with increase in substitution and are similar to that observed from composite flour of unripe plantain/wheat/watermelon seed. The carbohydrate contents for biscuit samples ranged from 50.75 – 69.71%. Significant different (p≤0.05) occurred within the samples. MW₁ (69.71%) had the highest while MW₅ (50.75%) had the least. However, carbohydrate contents of composite biscuit samples decreased according to higher substitution level.

Physical Properties of biscuits:

The results of the physical properties of the biscuit samples are shown in Table (5). Thickness, diameter, weight, spread ratio and break strength values ranged from 0.61 – 0.70cm, 3.0 – 3.4cm, 5.22 – 6.24cm, 5.05 – 5.26, and 1400 – 2995g respectively. Thickness and spread ratio have inverse relation. MW₁ (0.70) had the highest value in thickness while MW₂ (0.61) had the least. Significant difference (p≤0.05) occurred among biscuit samples. MW₂ and MW₅ were each significantly different from the rest of the samples respectively.

Spread ratio was found to be highest in MW₂ (5.26) while MW₃ (5.05) had the least. Significant difference (p≤0.05) occurred within the samples. W₁ and MW₄ are not significantly different likewise MW₁, MW₃ and
MW₃. MW₄ was significantly different from the rest of the samples. This result could be due to the dough viscosity as evidenced in viscosity of the flour blends (Table 5). According to Miller et al (1997), spread ratio is restricted by dough viscosity as dough with lower viscosity causes cookies to spread faster. Low spread ratio suggests that the starches in the cookies are very hydrophilic (Yahya, 2004; cited in Okpala and Egwu, 2015).

Table 5. Physical properties of Biscuits

<table>
<thead>
<tr>
<th>Sample</th>
<th>Thickness (CM)</th>
<th>Diameter (CM)</th>
<th>Weight (g)</th>
<th>Spread ratio (SR)</th>
<th>Break strength (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>W₁</td>
<td>0.66 ± 0.01</td>
<td>3.3 ± 0.08</td>
<td>6.24 ± 0.81</td>
<td>5.18 ± 0.02</td>
<td>1400 ± 0.00</td>
</tr>
<tr>
<td>W₂</td>
<td>0.69 ± 0.01</td>
<td>3.2 ± 0.05</td>
<td>5.74 ± 0.38</td>
<td>5.07 ± 0.02</td>
<td>1975 ± 20.41</td>
</tr>
<tr>
<td>W₃</td>
<td>0.61 ± 0.01</td>
<td>3.4 ± 0.00</td>
<td>6.01 ± 0.45</td>
<td>5.26 ± 0.02</td>
<td>2395 ± 4.08</td>
</tr>
<tr>
<td>W₄</td>
<td>0.70 ± 0.00</td>
<td>3.0 ± 0.05</td>
<td>6.05 ± 0.36</td>
<td>5.05 ± 0.04</td>
<td>2645 ± 4.08</td>
</tr>
<tr>
<td>W₅</td>
<td>0.65 ± 0.02</td>
<td>3.3 ± 0.05</td>
<td>5.42 ± 0.42</td>
<td>5.15 ± 0.04</td>
<td>2885 ± 4.08</td>
</tr>
<tr>
<td>MW₁</td>
<td>0.63 ± 0.01</td>
<td>3.1 ± 0.08</td>
<td>5.22 ± 0.15</td>
<td>5.07 ± 0.02</td>
<td>2995 ± 4.08</td>
</tr>
<tr>
<td>LSD</td>
<td>0.0199</td>
<td>-</td>
<td>-</td>
<td>0.0663</td>
<td>19.56</td>
</tr>
</tbody>
</table>

Mean ± standard deviation of triplicate determination. Means with the same superscripts in a column are not significantly different (P<0.05). W₁ = Wheat flour (100%)  
MW₁ = Watermelon seed meal/wheat flour (10:90)  
MW₂ = Watermelon seed meal/wheat flour (20:80)  
MW₃ = Watermelon seed meal/wheat flour (30:70)  
MW₄ = Watermelon seed meal/wheat flour (40:60)  
MW₅ = Watermelon seed/wheat flour (50:50)

Chinma and Gernah (2001) observed a similar trend in cookies produced from cassava/soya/mango composite flour and attributed it to the hydrophilic nature of the flour used in the products which led to decrease in spread ratio and increase in thickness. It was observed that MW₃, which had the least spread ratio, also had the highest thickness.

No significant difference occurred within the samples. Ade et al. (2012), noted that when a dough or batter becomes less viscous, it tends to spread more thereby increasing in diameter and consequently the spread factor. Again the biscuit sample with the highest spread ratio (MW₂) also had the highest diameter.

W₁ (6.24) was found to be highest in weight while MW₁ (5.22) had the least. No significant difference occurred among the samples, however, the weight decreased with higher percentage substitution with watermelon seed meal. Helen and Ndidiamaaka (2014) observed similar trend in cookies produced from wheat/beniseed/unripe plantain flours and attributed it to higher contents of starch and low crude fibre in wheat than unripe plantain and beniseed. It was also observed that the trend in weight was similar to the result of bulk density (Table 2) and crude fibre content (Table 4).

The result obtained from break strength showed that MW₅ (2995g) had the highest value while W₁ (1400g) had the least. Biscuit samples from composite flours were very hard and lacked the characteristic quality of biscuit due to the absence of fat in their recipe; as fat plays a major role in determining texture as well as rheology and overall quality of product (Ubbor and Akobundu, 2009). Though watermelon seed meal substituted fat in the biscuits, however, from the observation, small amount of fat should be added to aid that from the watermelon seed to be able to get the characteristic quality needed.

3.3 Sensory Qualities

The sensory evaluation scores of biscuit samples are shown in Table 6. There were significant differences (p<0.05) among the biscuit samples in the parameters evaluated. Biscuits made from wheat flour (100%) were significantly different from the biscuits made from composite flour samples except in colour and crispiness. The non-inclusion of fat in the recipe of composite flour samples affected biscuits made from them in taste, aroma, texture and overall acceptability as fat is an important factor which helps in improving flavour and mouthfeel and contributes to the appearance, palatability and texture of biscuits (Sushma et al., 2016).

No significant difference occurred among the composite biscuit samples, however, biscuits from MW₁ (6.05) received a higher score in aroma and overall acceptability.

Colour is an important sensory attribute of any food because of its influence on acceptability. The brown colour resulting from Maillard reaction is always associated with baked goods (Ubbor and Akobundu, 2009). Biscuit samples from MW₁ was not significantly different from that of W₁, likewise MW₂ and MW₃ while MW₄ and MW₅ were each significantly different (p<0.05) from the rest of the samples respectively.
Table 6. Sensory Evaluation Scores of Biscuits

<table>
<thead>
<tr>
<th>Sample code</th>
<th>Parameter</th>
<th>Taste</th>
<th>Aroma</th>
<th>Colour</th>
<th>Texture</th>
<th>Crispiness</th>
<th>Overall acceptance</th>
</tr>
</thead>
<tbody>
<tr>
<td>W&lt;sub&gt;1&lt;/sub&gt;</td>
<td></td>
<td>7.70 ± 1.01</td>
<td>7.20 ± 1.03</td>
<td>6.30 ± 1.58</td>
<td>6.45d ± 1.02</td>
<td>6.85ab ± 1.24</td>
<td>7.75 ± 0.94</td>
</tr>
<tr>
<td>MW&lt;sub&gt;1&lt;/sub&gt;</td>
<td></td>
<td>5.50 ± 1.29</td>
<td>6.05 ± 0.97</td>
<td>6.50 ± 0.50</td>
<td>6.90c ± 1.09</td>
<td>7.35a ± 1.28</td>
<td>6.05 ± 1.07</td>
</tr>
<tr>
<td>MW&lt;sub&gt;2&lt;/sub&gt;</td>
<td></td>
<td>5.85 ± 1.42</td>
<td>5.40 ± 1.28</td>
<td>7.20c ± 0.91</td>
<td>7.60b ± 0.92</td>
<td>5.0d ± 0.95</td>
<td>5.80b ± 1.72</td>
</tr>
<tr>
<td>MW&lt;sub&gt;3&lt;/sub&gt;</td>
<td></td>
<td>5.70 ± 1.82</td>
<td>5.50 ± 1.53</td>
<td>7.40c ± 0.80</td>
<td>7.85b ± 0.73</td>
<td>7.45a ± 1.28</td>
<td>6.00 ± 1.30</td>
</tr>
<tr>
<td>MW&lt;sub&gt;4&lt;/sub&gt;</td>
<td></td>
<td>5.65 ± 1.59</td>
<td>5.25 ± 1.70</td>
<td>8.10b ± 0.62</td>
<td>8.00ab ± 1.05</td>
<td>6.05bc ± 1.75</td>
<td>5.60b ± 1.28</td>
</tr>
<tr>
<td>MW&lt;sub&gt;5&lt;/sub&gt;</td>
<td></td>
<td>5.90 ± 1.58</td>
<td>5.20 ± 1.75</td>
<td>8.70a ± 0.48</td>
<td>8.35a ± 0.73</td>
<td>5.7c ± 1.38</td>
<td>5.7b ± 1.34</td>
</tr>
<tr>
<td>LSD</td>
<td></td>
<td>0.946</td>
<td></td>
<td>0.906</td>
<td>0.577</td>
<td>0.420</td>
<td>0.856</td>
</tr>
</tbody>
</table>

Mean ± Standard deviation of sensory evaluation score.
Means with the same superscripts within a column are not significantly different (P ≤ 0.05)

W<sub>1</sub> = Wheat flour (100%)
WM<sub>1</sub> = Watermelon seed meal/wheat flour (10:90)
WM<sub>2</sub> = Watermelon seed meal/wheat flour (20:80)
WM<sub>3</sub> = Watermelon seed meal/wheat flour (30:70)
MW<sub>1</sub> = Watermelon seed meal/wheat flour (40:60)
MW<sub>1</sub> = Watermelon seed meal/wheat flour (50:50)

This result showed increase in brown colour with higher substitution with toasted watermelon seed meal and could obviously be attributed to brown colour effect of watermelon seed meal.

MW<sub>3</sub> was significantly different (p≤0.05) in texture with values ranged from 6.45 to 8.35. MW<sub>3</sub> and MW<sub>4</sub> were not significantly different, likewise MW<sub>2</sub>, MW<sub>3</sub> and MW<sub>4</sub>. The texture was strong and firm in composite biscuit samples with increase in substitution. This observation could be due to the non-inclusion of fat in the composite biscuit recipe as fats help to lubricate, weaken, or shorten the structure of food components to provide a food product with desirable textural properties (Hasmadi and Sandra, 2014).

Crispiness is a desired quality of biscuits. Significant difference (p≤0.05) existed among the biscuit samples. With respect to overall acceptability, W<sub>1</sub> (100% wheat flour) was most preferred. Significant difference (p≤0.05) occurred between 100% wheat flour biscuit samples and composite flour biscuit samples. W<sub>1</sub> was significantly different from the other samples. No significant difference among biscuit samples from composite flour blends even though MW<sub>1</sub> (6.05) and MW<sub>3</sub> (6.0) received higher scores respectively.’

3.4 Storage Study (Microbial Count And Moisture Absorption)

Total viable count reflects the conditions in which the food was produced, stored, or abused during handling and can be used to predict the shelf life or keeping quality of the product. The spoilage of many foods may be imminent when the total viable count reaches from 10 – 100 million per gram of the product (Nagi et al., 2012). The total microbial load on nutrient agar for bacteria and Potato-dextrose agar for fungi were 2.0×10<sup>3</sup>cfu/g and 1.8×10<sup>3</sup>cfu/g for MW<sub>1</sub> and 2.8×10<sup>3</sup>cfu/g and 2.7×10<sup>3</sup>cfu/g for MW<sub>3</sub> respectively in week one.

Almost all microorganisms were destroyed during baking due to high temperature. But during packaging, cross contamination could occur due to handling of biscuits. Bacteria usually require at least 0.91 water activity (a<sub>w</sub>), and fungi require at least 0.7 a<sub>w</sub>, while below 0.6 a<sub>w</sub>, microorganisms cannot survive (Pupulawaththa et al., 2014). Total microbial load increased in week 3. The result in Table 4.6 also indicated that biscuits absorbed moisture during the period of storage.

Table 7. Total Viable Counts of Bacteria and Mould from the Accepted Blends of Biscuits Stored For 6 Weeks and the %Moisture Content

<table>
<thead>
<tr>
<th>Sample code</th>
<th>Types of organism</th>
<th>Storage time (weeks)</th>
<th>% moisture absorbed in 6 weeks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>MW&lt;sub&gt;1&lt;/sub&gt;</td>
<td>Bacteria</td>
<td>2.0x10&lt;sup&gt;3&lt;/sup&gt;</td>
<td>5.2x10&lt;sup&gt;3&lt;/sup&gt;</td>
</tr>
<tr>
<td>MW&lt;sub&gt;3&lt;/sub&gt;</td>
<td></td>
<td>2.8x10&lt;sup&gt;3&lt;/sup&gt;</td>
<td>6.8x10&lt;sup&gt;3&lt;/sup&gt;</td>
</tr>
<tr>
<td>MW&lt;sub&gt;1&lt;/sub&gt;</td>
<td>Mould</td>
<td>1.8x10&lt;sup&gt;3&lt;/sup&gt;</td>
<td>6.1x10&lt;sup&gt;3&lt;/sup&gt;</td>
</tr>
<tr>
<td>MW&lt;sub&gt;3&lt;/sub&gt;</td>
<td></td>
<td>2.7x10&lt;sup&gt;3&lt;/sup&gt;</td>
<td>7.4x10&lt;sup&gt;3&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

WM<sub>1</sub>=Watermelon seed meal/wheat flour (10:90)
WM<sub>3</sub>= Watermelon seed meal/wheat flour (30:70)

The gain in moisture might be due to hydrosopic nature of dried products, storage environment (temperature and relative humidity) as well as the nature of packaging material (Nagi et al., 2012). The increase in microbial
load as the storage period lengthened might have been due to a corresponding increase in moisture contents during storage and availability of nutrient in the biscuits. Also, there was no addition of preservative to inhibit microbial growth, low acid nature of biscuit and low level of antimicrobial activities of watermelon seed extract (Braide et al., 2012) may have contributed an increase of the microbial load. The percentage moisture absorbed ranged from 6.25% (zero) to 11.75%. MW1 absorbed the highest moisture and was also the most crispest biscuit amongst all.

4. Conclusion

The results of the functional properties of the flour blends showed that the composite flour blends could be suitable for biscuit production and other food formulations.

The research has also revealed that the proximate composition of biscuits produced from the composite flour blends are better, compared to that made solely from wheat flour and hence toasted watermelon seed meal can be incorporated up to 30% substitution level based on the sensory score. Biscuits prepared from such composite flour could help in combating protein-energy malnutrition; have the potential as a functional food especially for coeliac, diabetic, obese and heart disease patients in view of their high protein, crude fibre, and ash contents, with low fat and carbohydrate contents than 100% wheat flour biscuits.

References


Helen, O. A., & Ndidiamaaka, A. O. (2014). Physico-chemical, sensory, and microbiological assessments of


https://doi.org/10.3923/pjn.2009.1097.1102


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