THE EFFECT OF COWPEA (*Vigna unguiculata*) FLOUR AS AN EXTENDER ON THE PHYSICO-CHEMICAL PROPERTIES OF BEEF AND HAM BURGERS

Teye GA¹*, Teye M² and G Boamah¹

*Corresponding author email: teye.gabriel@yahoo.com

¹Department of Animal Science, University for Development Studies, P.O. Box TL 1882, Tamale, Ghana

²Department of Animal Science, School of Agriculture, University of Cape Coast, Cape Coast, Ghana
ABSTRACT

This study was conducted in search of meat extenders/filler, which would minimize excessive bulging (swelling at the centre) of beef and hamburgers so as to enhance their use in sandwiches. The potential of dehulled cowpea flour was evaluated in beef and hamburgers, to determine its effects on the sensory, physical and chemical characteristics of these products. The Black-eyed cowpea (Vigna unguiculata) variety was steam treated at 100°C, dehulled, sundried for 48 hours and ground into flour. Boneless beef and pork (6kg each) were minced and apportioned into four groups of 1.5kg each for the preparation of the burgers. The beef and hamburgers were formulated separately at four levels of cowpea flour inclusions; T1 (control; no cowpea flour), T2 (5%), T3 (7.5%) and T4 (10%) of minced meat, on weight basis. All other ingredients were added in equal amounts to the minced beef and mixed in a mechanical mixer, after which they were moulded manually using a cylindrical tube into uniform shapes and sizes, and stored in a chest freezer for 12 hours to harden, after which they were bagged and stored for analyses. The weights, thicknesses and diameters of the products were taken before, and after cooking to determine the physical changes in them. The crude protein, fat moisture and lipid peroxidation of the products were determined. The burgers were grilled in an oven to a core temperature of 70°C and served to a 15-member taste panel for evaluation. The results indicate that cowpea flour in burgers increases the crude protein content, reduces the crude fat content and has no negative effect on sensory and lipid peroxidation of these products. There were reductions in product bulging and shrinkage with an increase in cowpea flour inclusion. Cowpea flour is recommended for inclusion in beef and hamburgers up to 10% on weight basis.

Key words: burgers, cowpea, extender, sensory, bulging
INTRODUCTION

Processing of raw meat into products does not only add value and extend the shelf life of meat pieces, but also serves as a source of employment to the processors [1]. Meat products such as burgers require the use of mostly boneless meat; a practice which results in an increased production cost due to the expensive nature of boneless meat. The products consequently become very expensive [2], restricting their patronage to only the wealthy or higher income earners in the society [3]. One way of minimizing formulation cost in meat processing is by using meat extenders.

Extenders or fillers are usually protein additives used to increase water binding capacity and yield of meat products. Most extenders enhance protein content, improve processing yields and reduce formulation costs [1]. Some important meat extenders include soy proteins, milk proteins, starch and flours.

Currently, burgers are prepared with bread crumb as extenders, a practice that causes the products to bulge, making it unsuitable for use in sandwiches. A preliminary study with cowpea flour as an extender reported an increase in yield, but an insignificant margin of bulging in ham and beef burgers [4]. Recent studies involving cowpea flour as an extender up 10% inclusion in coarse smoked pork and beef sausages, meat balls and comminuted beef and pork frankfurter-style sausages have shown promising results in terms of yield and sensory qualities of these products [5, 6, 7, 8].

This study was, therefore, to determine the effect of dehulled cowpea flour as an extender on physical, chemical and sensory characteristics of beef and hamburgers.

MATERIALS AND METHODS

The research was conducted at the Meat Processing Unit and Laboratories of the University for Development Studies, (UDS) Tamale, Ghana.

Preparation of cowpea flour
The seeds of the erect Black-eyed Cowpea (Vigna unguiculata) variety were obtained from the local market and steam-treated at 100°C for an hour to remove the beany flavour that may affect the aroma and flavour of the burgers. The heat-treated beans were then rubbed in the palms to remove the testa, sun-dried for 48 hours and ground into flour using a conventional corn mill. The flour was stored in an airtight container for later use.

Preparation of burgers
Fresh boneless beef and pork (6kg each) were obtained and chopped into smaller pieces, and minced separately using a 5mm sieve in a tabletop mincer (Talleres Rammon, Spain). Each of the minced meat was apportioned into four groups of 1.5kg. The burgers were prepared at four inclusion levels: T1 (no extender), T2 (5% cowpea flour), T3 (7.5% cowpea flour) and T4 (10% cowpea flour). All other ingredients were added in equal amounts (g/kg) to the various minced meat: 13.0g curing salt,
0.5g red chillies, 1.0g black pepper, and 1.0g white pepper and 2g “adobo” (pre-formulated spices).

The minced meat, together with the spices were put in a mixer (Talleres Rammon, Spain) and mixed thoroughly for 5 minutes, moulded manually using a cylindrical tube into uniform shapes and sizes. They were then frozen at -5°C for 12 hours, bagged and labelled for analysis.

**Measurements of physical characteristics of the products**

**Cooking loss**
The burgers were weighed using an electronic scale before (W1) and after (W2) grilling. The difference in weight (cooking loss) was determined by subtracting W2 from W1.

**Thicknesses and Diameters**
The thicknesses and diameters of the products were measured using digital callipers before and after cooking, as shown in Plates 1 and 2 below:

![Plate 1: Measurement of Diameter of burgers](image-url)
The cooked thicknesses and diameters of the individual products were subtracted from the fresh ones to obtain the changes in thickness and diameters (in millimetre).

**Shrinkage**

Percentage (%) shrinkage was calculated using the formula [6]:

$$\text{Shrinkage (\%)} = \frac{(RT-CT) + (RD-CD)}{(RT+RD)} \times 100$$

Where RT = Raw thickness, CT = Cooked thickness, RD = Raw diameter and CD = Cooked diameter.

**Selection and training of taste panel**

A total of 15 panellists, comprising staff and students of the University were randomly selected and trained according to the British Standard Institution guidelines, to evaluate the products [9].

**Sensory Evaluations**

Sensory evaluations were carried out to determine the colour, firmness, cowpea flavour intensity, flavour liking and overall acceptability of the products by the taste panel. The burgers were grilled to a core temperature of approximately 70°C in an oven (Turbofan, Blue seal, UK). The products were sliced into uniform sizes (2cm²) and wrapped with coded aluminium foils and presented to the panellists. Each panellist was provided with water and pieces of bread to serve as neutralizers between the products.
A five-point category scale was used to evaluate the characteristics of the products as follows:

**Colour:** 1=Very light; 2= light; 3=Intermediate; 4=Dark; 5= Very dark.

**Firmness:** 1=Very weak; 2=Weak; 3=Intermediate; 4=Firm; 5=Very firm

**Juiciness:** 1=Very juicy; 2=Juicy; 3=Intermediate; 4=Dry; 5=Very dry

**Cowpea flavour intensity:** 1=Very weak; 2=Weak; 3=Intermediate; 4=Strong; 5=Very strong

**Flavour liking/Overall acceptability:** 1=Like very much; 2=Like; 3=Intermediate; 4=Dislike; 5=Dislike very much.

### Chemical characteristics

#### Proximate analysis of products

The burgers were analyzed for moisture, crude protein and fat contents according to the procedures of the Association of Official Analytical Chemists [10]. Analyses were conducted in duplicates; all reagents were of analytical grade.

#### Measurement of peroxide value

Peroxide value (POV) was determined according to the method of the AOAC International [10], to ascertain lipid per-oxidation in the products. The samples (3g each) were weighed in 250-ml glass stoppered Erlenmeyer flasks and heated in a water bath at 60°C for 3 min to melt the fat, then thoroughly agitated for 3 min with 30 ml acetic acid–chloroform solution (3:2 v/v) to dissolve the fat. The samples were filtered through Whatman filter paper to remove meat particles. Saturated potassium iodide solution (0.5 ml) was added to the filtrates in each of the flasks. They were then titrated against standard solution of sodium thiosulfate (25 g/l) with 0.5ml 1% starch solution as an indicator. Titration continued with vigorous shaking to release all iodine from CHCl₃ layer until blue colour just disappeared.

The POV was calculated and expressed as milli-equivalent peroxide per kg of sample, as follows:

\[
POV = \frac{S \times N}{W}
\]

Where S = the titre value (ml), N= the normality of sodium thiosulfate solution (N=0.1), and W= the sample weight (kg).

#### Statistical analyses

The data obtained were analyzed using the General Linear Model (GLM) of Analysis of Variance (ANOVA) of Minitab, version 15[11]. Where significant differences occur, Tukey’s simultaneous test was used to separate the mean at P<0.05.

### RESULTS

#### Physical characteristics of the products

The results of cooking loss, changes in diameters, thicknesses and percentage shrinkage of the burgers after cooking, were as presented in Tables 1 and 2.
The hamburgers without extenders lost an average weight of 43.67g during cooking, whilst those with the extender lost weights ranging between 24.50g and 27.00g, about half (56%) of the weight loss in the control products (Table 1). The T1 products had significant reductions in weights (P< 0.01), relatively greater reductions in diameters and thicknesses compared with those containing the extenders. The products with the extenders (T2, T3 and T4) had a marginal increase in thickness; the highest being the T2 product (1.17mm) and the least being the T3 products (0.13mm).

**Sensory characteristics of burgers**
The use of cowpea flour as an extender resulted in products with similar colour, juiciness, flavour, flavour liking and overall acceptability of ham and beef burgers (Tables 3 and 4). The burgers without extenders however, were firmer than those with the extenders.

**Chemical characteristics**

**Proximate composition of the burgers**
The moisture, fat and crude protein contents of the burgers are presented in Tables 5 and 6.

The use of cowpea flour as an extender in beef burgers increased the crude protein content significantly (P< 0.01) from 17.89% in the control products to 23.67% in the T4 products (Table 5). The moisture content also increased whilst the fat level decreased, but the differences were not significant (P> 0.05).

The cowpea flour significantly reduced (P<0.01) the fat levels in the hamburgers. There was also an increase in moisture and crude protein contents but these differences were however, not significant (Table 6).

**Peroxide values of burgers**
The lipid peroxidation in the products was determined after 7 days in storage to determine the effect of cowpea flour on lipid peroxidation in the products. The results are presented in Figure 1.

![Figure 1: Peroxide value of beef and ham burgers](image-url)
The peroxide values in the hamburgers ranged between 12.00 milli-equivalent of active oxygen per kilogram product in the control (T1) products and 5.00 milli-equivalent of active oxygen per kilogram in the T4 products. Similarly, the POVs in the beef-burgers were between 10.66 and 5.66 milli-equivalent of active oxygen per kilogram product for T1 and T4 products, respectively.

DISCUSSION

Physical characteristics of the products
It can be realized that there were decreases in cooking loss, thicknesses and diameters of the products with an increase in cowpea flour content (Tables 1 and 2). The control (T1) products had a reduction in thickness while the cowpea resulted in a slight swelling in the other products. These can be explained by the following: Mincing destroys the structural integrity of the cell proteins lowering their water holding ability. This is aggravated when heat is applied since the high temperatures cause protein denaturation and a further reduction in the ability to bind added water [12]. This consequently causes the product to shrink and reduce in size (Table 1). The use of fillers/extenders in meat products improves the water retention capacity of these products, minimize the level of shrinkage and therefore result in an increased yield [1]. Investigations indicated that an increase in soy protein level lowered the shrinkage of patties [13]. Addition of corn flour increased cooking yields of meatballs, and this was attributed to the ability of the product to keep the moisture in the matrix [14]. These therefore explain the relatively lower shrinkage in the products with cowpea flour extenders.

This is an indication that the use of cowpea flour in burgers would minimize shrinkage, and at the same time prevent excessive bulging of the products as compare to products with bread crumbs.

Sensory characteristics of burgers
The products with extenders did not have an intense cowpea flavour compared to the control, suggesting that the cowpea flour did not have significant influence on the flavour intensity in the products (Tables 3 and 4). This observation may be attributed to the heat treatment given the cowpea, which removed the beany flavour [15]. The firmer structure of the control products may be due to the greater loss of moisture, which resulted in meat particles holding on tight to each other and therefore making the product dry and firm. Higher moisture content in a product improves tenderness [6], and that could have resulted in the products with extenders being less dried compared to the control products.

Proximate composition of the burgers
The cowpea flour increased the crude protein contents of the burgers (Table 5). Basic requirements of extenders in meat products include improving the nutritional quality and minimizing fat content without having an adverse effect on sensory characteristics of the product [1]. Proteins are required in higher levels in growing children and also for productive functions such as pregnancy and lactation because of increased output of proteins in the products of conception and in milk [17]. Therefore,
with a higher crude protein level in a product, a small quantity of it will be required by consumers to meet their nutrient requirements. This will reduce expenditure on meat and meat products, as well as satisfy health concerns over excessive meat intake. Since the cowpea flour increased the crude protein levels of beef-burgers, its use will improve the nutritional quality of the product.

The fat contents of the products were reduced with an increase in cowpea flour inclusion (Table 6). Excessive intake of dietary saturated fats has been associated with the development of hypertension, cardio-vascular diseases, obesity, cancers of the colon, breast and prostate [18, 19]. A number of health organizations including the World Health Organization, have made recommendations to reduce daily fat intake for improved health [20]. However, dietary fat plays a major role in the texture, juiciness and flavour of comminuted meat products [21]. Research indicates that the sensory properties of fat make a diet flavourful [22]. Reduction in dietary fat is therefore likely to reduce the sensory characteristics of food products [23]. Since the use of cowpea flour caused a reduction in fat content without an adverse effect on product acceptability, it could be used in making healthy burgers.

**Lipid peroxidation in the burgers**

In both the beef and hamburgers, the lipid per-oxidation reduced with an increase in cowpea flour inclusion. Lipid per-oxidation progresses at faster rates in foods with high fat content, especially those with higher levels of unsaturated fatty acids [24]. The unsaturated fatty acids present in the products react with oxygen to form fatty acid hydro-peroxides. Hydro-peroxides are unstable, and breakdown into various compounds which produce off-flavours, leading to formation of a stale, rancid flavour in food products [25]. However, in all the products, the POVs were lower than the 25millequivalent of active O₂/kg, which is the acceptable limit in fatty foods [26, 27]. The use of cowpea flour therefore, has the potential of extending the shelf-life of the products.

**CONCLUSION**

The use of cowpea flour as an extender in burgers increased the crude protein content, reduced the fat content and had no adverse effect on the sensory characteristics of these products. It also minimized excessive bulging and shrinkage in the products. Cowpea flour is, therefore, recommended for use as an extender in beef and ham burgers.
Table 1: Cooking loss, changes in Diameters and Thicknesses of hamburgers after cooking

<table>
<thead>
<tr>
<th>Parameters</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>Sed</th>
<th>Sig.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cooking loss (g)</td>
<td>43.67&lt;sup&gt;a&lt;/sup&gt;</td>
<td>27.00&lt;sup&gt;b&lt;/sup&gt;</td>
<td>24.50&lt;sup&gt;b&lt;/sup&gt;</td>
<td>24.50&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.63</td>
<td>**</td>
</tr>
<tr>
<td>Change in Diameter (mm)</td>
<td>18.18</td>
<td>11.84</td>
<td>12.66</td>
<td>12.70</td>
<td>2.46</td>
<td>ns</td>
</tr>
<tr>
<td>Change in Thickness (mm)</td>
<td>1.29</td>
<td>-1.17</td>
<td>-0.13</td>
<td>-1.27</td>
<td>1.39</td>
<td>ns</td>
</tr>
<tr>
<td>Shrinkage (%)</td>
<td>17.71</td>
<td>9.92</td>
<td>11.45</td>
<td>10.36</td>
<td>3.08</td>
<td>ns</td>
</tr>
</tbody>
</table>

Sed= Standard Error of difference, Sig.= significance, Means in the same row with different superscripts are significant, ns= not Significant, **= Significant (p<0.01)

Table 2: Cooking loss, changes in Diameters and Thicknesses of beef burgers after cooking

<table>
<thead>
<tr>
<th>Treatment</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>Sed</th>
<th>Sig.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cooking loss (g)</td>
<td>37.33</td>
<td>25.33</td>
<td>28.67</td>
<td>22.33</td>
<td>6.76</td>
<td>ns</td>
</tr>
<tr>
<td>Change in diameter (mm)</td>
<td>15.86&lt;sup&gt;a&lt;/sup&gt;</td>
<td>10.84&lt;sup&gt;b&lt;/sup&gt;</td>
<td>11.43&lt;sup&gt;b&lt;/sup&gt;</td>
<td>9.80&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.10</td>
<td>***</td>
</tr>
<tr>
<td>Change in thickness (mm)</td>
<td>0.48</td>
<td>-1.93</td>
<td>-0.73</td>
<td>-1.83</td>
<td>1.51</td>
<td>ns</td>
</tr>
<tr>
<td>Shrinkage (%)</td>
<td>14.09&lt;sup&gt;a&lt;/sup&gt;</td>
<td>8.30&lt;sup&gt;b&lt;/sup&gt;</td>
<td>9.89&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>7.40&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.72</td>
<td>**</td>
</tr>
</tbody>
</table>

Sed= Standard Error of difference, Means in the same row with different superscripts are significant, ns= not Significant, **= significant (p<0.01), ***= Significant (p<0.001)
Table 3: Sensory characteristics of Hamburgers

<table>
<thead>
<tr>
<th>Days</th>
<th>Parameters</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>Sed</th>
<th>Sig.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Colour</td>
<td>4.00</td>
<td>3.60</td>
<td>3.80</td>
<td>3.40</td>
<td>0.63</td>
<td>ns</td>
</tr>
<tr>
<td>1</td>
<td>Firmness</td>
<td>3.70^a</td>
<td>2.90^ab</td>
<td>2.10^b</td>
<td>2.50^ab</td>
<td>0.75</td>
<td>*</td>
</tr>
<tr>
<td>1</td>
<td>Juiciness</td>
<td>2.40</td>
<td>2.10</td>
<td>2.20</td>
<td>2.70</td>
<td>0.52</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Cowpea flavour intensity</td>
<td>3.10</td>
<td>3.50</td>
<td>3.30</td>
<td>3.50</td>
<td>0.52</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Flavour liking</td>
<td>2.10</td>
<td>1.90</td>
<td>2.20</td>
<td>2.30</td>
<td>0.73</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Acceptability</td>
<td>1.90</td>
<td>1.70</td>
<td>2.20</td>
<td>2.10</td>
<td>0.71</td>
<td>ns</td>
</tr>
<tr>
<td>7</td>
<td>Colour</td>
<td>3.30</td>
<td>3.50</td>
<td>3.40</td>
<td>3.10</td>
<td>0.44</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Firmness</td>
<td>4.10^a</td>
<td>2.90^ab</td>
<td>2.30^b</td>
<td>2.10^b</td>
<td>0.78</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td>Juiciness</td>
<td>2.50</td>
<td>2.30</td>
<td>2.00</td>
<td>1.80</td>
<td>0.52</td>
<td>ns</td>
</tr>
<tr>
<td>7</td>
<td>Cowpea flavour intensity</td>
<td>2.00</td>
<td>2.60</td>
<td>2.20</td>
<td>2.60</td>
<td>0.77</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Flavour liking</td>
<td>2.30</td>
<td>2.10</td>
<td>2.10</td>
<td>2.00</td>
<td>0.48</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Acceptability</td>
<td>2.00</td>
<td>1.80</td>
<td>1.80</td>
<td>2.00</td>
<td>0.47</td>
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Sed= Standard error of difference, Means in the same row with different superscripts are significant  ns= not Significant, *= significant (p<0.05), ***=significant (p<0.001)
Table 4: Sensory characteristics of beef burgers

<table>
<thead>
<tr>
<th>Days</th>
<th>Parameters</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>Sed</th>
<th>Sig</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Colour</td>
<td>3.80</td>
<td>3.20</td>
<td>2.40</td>
<td>2.70</td>
<td>0.82</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Firmness</td>
<td>4.00</td>
<td>2.90</td>
<td>3.40</td>
<td>3.10</td>
<td>0.54</td>
<td>ns</td>
</tr>
<tr>
<td>1</td>
<td>Juiciness</td>
<td>2.80</td>
<td>2.90</td>
<td>2.90</td>
<td>2.80</td>
<td>0.65</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Cowpea flavour intensity</td>
<td>2.30</td>
<td>3.00</td>
<td>2.70</td>
<td>3.00</td>
<td>0.77</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Flavour liking</td>
<td>1.80</td>
<td>2.90</td>
<td>2.50</td>
<td>3.00</td>
<td>0.84</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Acceptability</td>
<td>1.80</td>
<td>2.60</td>
<td>2.50</td>
<td>3.20</td>
<td>0.91</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Colour</td>
<td>4.30</td>
<td>4.00</td>
<td>3.90</td>
<td>4.00</td>
<td>0.41</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Firmness</td>
<td>4.40&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.80&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.00&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>2.70&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.74</td>
<td>***</td>
</tr>
<tr>
<td>7</td>
<td>Juiciness</td>
<td>3.60</td>
<td>2.70</td>
<td>3.00</td>
<td>3.20</td>
<td>0.74</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Cowpea flavour intensity</td>
<td>2.40</td>
<td>2.60</td>
<td>2.90</td>
<td>3.10</td>
<td>0.74</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Flavour liking</td>
<td>2.30</td>
<td>2.10</td>
<td>2.10</td>
<td>2.20</td>
<td>0.63</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Acceptability</td>
<td>2.20</td>
<td>2.40</td>
<td>2.70</td>
<td>3.30</td>
<td>0.73</td>
<td>ns</td>
</tr>
</tbody>
</table>

Sed= Standard error of difference, Means in the same row with different superscripts are significant  ns= not Significant, ***= Significant (p<0.001)

Table 5: Proximate composition of Beef burgers

<table>
<thead>
<tr>
<th>Parameters (%)</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>Sed</th>
<th>Sig</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture</td>
<td>71.55</td>
<td>76.37</td>
<td>78.11</td>
<td>75.75</td>
<td>1.71</td>
<td>ns</td>
</tr>
<tr>
<td>Fat (ether extract)</td>
<td>6.73</td>
<td>5.87</td>
<td>5.44</td>
<td>4.80</td>
<td>1.67</td>
<td>ns</td>
</tr>
<tr>
<td>Crude Protein</td>
<td>17.89&lt;sup&gt;b&lt;/sup&gt;</td>
<td>18.85&lt;sup&gt;b&lt;/sup&gt;</td>
<td>22.35&lt;sup&gt;a&lt;/sup&gt;</td>
<td>23.67&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.63</td>
<td>**</td>
</tr>
</tbody>
</table>

Sed= Standard error of difference, Means in the same row with different superscripts are significant  ns= not Significant, **= Significant (p<0.01)
Table 6: Proximate composition of Hamburgers

<table>
<thead>
<tr>
<th>Parameters (%)</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>Sed</th>
<th>Sig.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture</td>
<td>64.48</td>
<td>61.94</td>
<td>65.58</td>
<td>66.39</td>
<td>0.98</td>
<td>ns</td>
</tr>
<tr>
<td>Fat (ether extract)</td>
<td>14.68^a</td>
<td>10.87^ab</td>
<td>8.94^bc</td>
<td>7.80^c</td>
<td>0.59</td>
<td>**</td>
</tr>
<tr>
<td>Protein</td>
<td>17.88</td>
<td>20.69</td>
<td>20.09</td>
<td>23.44</td>
<td>1.37</td>
<td>ns</td>
</tr>
</tbody>
</table>

Sed = Standard error of difference, Means in the same row with different superscripts are significant

sig = significant, ns = not significant, ** = Significant (p<0.01)
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