

Effect of cooking methods and temperature on proximate and amino acid composition of breakfast sausage

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ABSTRACT

Aims: The effect of cooking method and temperature on amino acid composition of breakfast sausage (BS) was undertaken in this study.

Methodology: Three batches of prepared BS from beef, were randomly allotted to three cooking methods (CM): (boiling, grilling and frying) each at cooking temperatures (CT) of 80, 90 and 100°C to attain internal temperature of 72°C in a completely randomized design. Samples from each treatment were oven-dried and assayed for amino acid and proximate composition using standard procedures. Data obtained was analysed using descriptive statistic and ANOVA at $\alpha_{0.05}$.

Results: Results showed that grilled sausage at 80 °C had highest total amino acid profile (3.2 %). Grilled sausage at 80 °C had highest crude protein (25.58 %). Grilled BS at 80°C recorded least fat content (15.99 %). Grilled sausage at 80 °C had the higher ash (6.66 %) and least (1.40 %) in boiled sausage at 90°C.

Conclusion: Therefore, breakfast sausage could be best grilled at 80°C due to maintain high amino acid profile, crude protein, ash and lower fat content.

Keywords: cooking methods, amino acid, breakfast sausage, cooking temperatures

1. INTRODUCTION

Meat can be consumed fresh, cured, dried or otherwise processed. About one third of all meat is processed, which means it has been changed from its original fresh cut [1]. Ham and sausage are the most popular processed meat products [1]. Sausages are made from chopped or comminuted lean meat and fat mixed with salt, spices and other ingredients, and then filled into a casing made of animal intestine or cellulose. Sausages can be classified based on country of origin or types of spices added [2]. An example of this is breakfast sausage, categorised under fresh sausage type.

Meat and its products become more edible and digestible when subjected to cooking [3]. During cooking, meat undergoes both physical and chemical changes, such as a decrease in the nutritional value, which are strongly dependent on protein denaturation and water loss [4] and [5]. Heating time, temperature, cooking method and muscle composition are all important variables, which may influence the final desirable characteristics of the meat [6] and [7]. It is well known that an increase in core temperature in meat will promote collagen shrinkage, reduce water holding capacity and increase cooking loss that influences its final quality and acceptability [8]. Although meat changes induced by cooking have been studied for many years and extensively discussed [9] and [5], only few reports have specifically dealt with the influence of different cooking conditions on the amino acid and mineral contents [10]. Therefore, the effect of three cooking methods (frying, boiling and grilling) and three cooking temperatures (80, 90 and 100 °C) on proximate and amino acid profile of breakfast sausage was evaluated in this study.

35 **2. MATERIAL AND METHODS**

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37 **2.1 Experiment site**

38 The experiment was carried out at the Animal Product and Processing (Meat Science) Laboratory,
39 Department of Animal Science, University of Ibadan, Ibadan.

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41 **2.2 Meat Source and sausage preparation**

42 Semi-membranous muscle from mature bull was purchased from the University abattoir, immediately after
43 slaughter. The large intestine of pig was also collected from the Pig slaughter slab at the Piggery Unit of
44 the University Teaching and Research Farm while lard was purchased from Bodija market (Pig Unit)
45 abattoir. The meat was cleaned; connective tissues and fat were properly trimmed out and was kept in
46 the refrigerator $4 \pm 1^\circ\text{C}$ overnight prior to sausage preparation. Meat and fat were run separately through
47 an automated meat mincer, the meat through 6mm plate and the fat through 4mm plate. Then the rest of
48 the meat and the other ingredients were thoroughly mixed and reground through a 4mm plate.

49 Salt, sodium nitrite, phosphate and sugar were dissolved in iced water, added to facilitate the mixing. The
50 blended lard, soybean used as binder and dry spice (Table 2) were mixed. The green spices onion,
51 ginger and garlic in ratio of 3:1:1 (Table 3) were finely chopped and ground for the preparation of green
52 spice mixture. Green spice and dry spice mixture were poured simultaneously into the mixing bowl. All
53 meat and non-meat ingredients were mixed together and ground again to desired consistency.

54 The prepared sausage was stuffed into presoaked natural casing (pig intestine) that was presoaked in
55 brine using an automated stuffer. The stuffed casings were divided into links (units) by twisting; each
56 sausage link was 10cm in length. The finished sausages were then packaged in Ziploc bags and frozen
57 at -4°C .

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59 **Table 1: Sausage composition**

Ingredient	Composition %
Beef	65.00
Lard	20.00
Binder	3.50
Curing salt *	2.00
Sugar	1.00
Phosphate	0.30
Ice water	4.00
Dry spices **	2.00
Green spices ***	2.20
Total	100.00

60 * Sodium chloride and Sodium nitrite(99% and 1% respectively)

61 ** Table 2

62 *** Table 3

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64 **Table 2: Composition of dry spices for breakfast sausage**

Spice	Inclusion level %
Black pepper	20.00
Nutmeg	7.00
Calabash nutmeg	3.00
Red pepper	20.00
Monosodium glutamate	15.00

Thyme	20.00
Curry powder	10.00
Total	100.00

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Table 3: Composition of green spices for breakfast sausage

Spices	Inclusion level %
Onion (<i>Allium cepa</i>)	60
Ginger (<i>Zingiberofficinale</i>)	20
Garlic (<i>Allium sativum</i>)	20
Total	100

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68 **2.3 Cooking methods**

69 Sausages were subjected to three cooking methods which were grilling using gas grilling machine, frying
70 and boiling using water bath at three cooking temperature; 80, 90, and 100 °C until internal temperature
71 of 72 °C was reached.

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73 **2.4 Amino acid composition**

74 Essential amino acids were determined by the spectrophotometric method using Ninhydrin chemical
75 reaction according to [11].

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77 **2.5 Determination of proximate composition**

78 Proximate composition was done according to the procedure of [12].

79 **2.6 Experimental design**

80 3 X 3 factorial arrangement in complete randomized design.

81 **2.7 Statistical Analysis**

82 Data were subjected to analysis of variance using [13]. Means were separated using Duncan's Multiple
83 Range Test option of the same software.

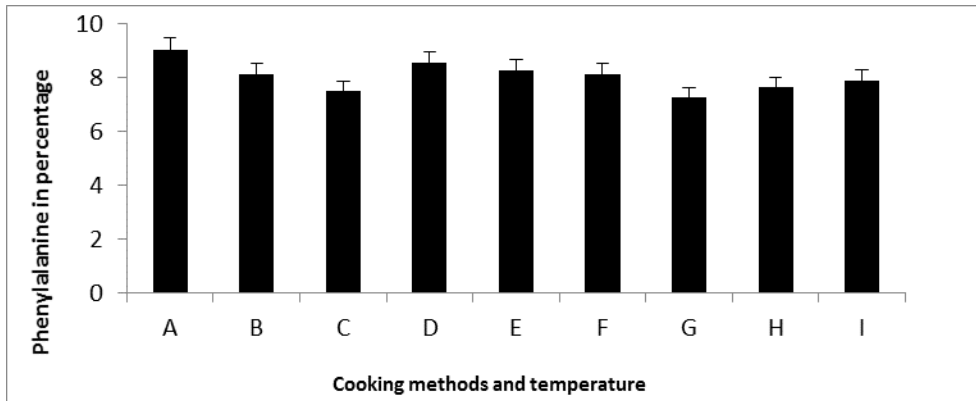
84 **3 RESULTS AND DISCUSSION**

85 **3.1 Essential amino acid score of breakfast sausage**

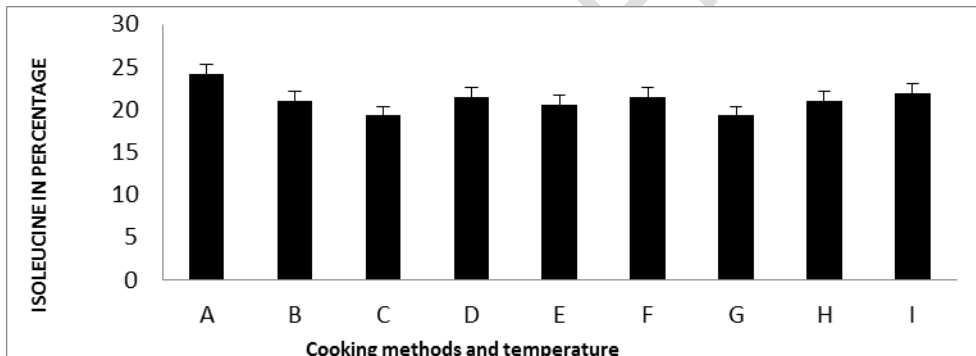
86 Presented in Figure 1 to 12 was the essential amino acid profile of breakfast sausage cooked at different
87 temperatures. From the result, it was observed that grilled sausage at 80°C had the highest total amino
88 acid profile (3.2%), followed by fried sausage at 80°C (2.8%) while boiled sausage at 80°C had the
89 lowest value (2.5%).

90 Grilled sausage had highest amino acid profile among the three different cooking methods (Figure 2),
91 followed by frying while boiling had the lowest rating. This could be due to higher water content found in
92 boiled sausage. Water molecules are highly polar and attracted to the muscle proteins by ionisable basic
93 and acidic groups such as arginine, histidine, lysine, glutamic acid. The meat protein was denatured by
94 heat and a part of its ionized basic or acid groups were broken. The possible reason for this might be that
95 the high temperature caused rapid denaturation of meat protein when compared with low temperature,
96 resulting in loss of more water molecules [14]. Although, the boiling of meat at 90 and 100°C showed
97 increased amino acid contents when compared with that of 80°C. Boiling resulted in denaturation of
98 proteins in meat and the denatured meat proteins could be easily hydrolyzed. When dissolved in
99 hydrochloric acid for determination of amino acids, the meat boiled for longer time with high content of
100 denatured proteins easily hydrolyzed resulting in high amino acid contents than when boiled for shorter

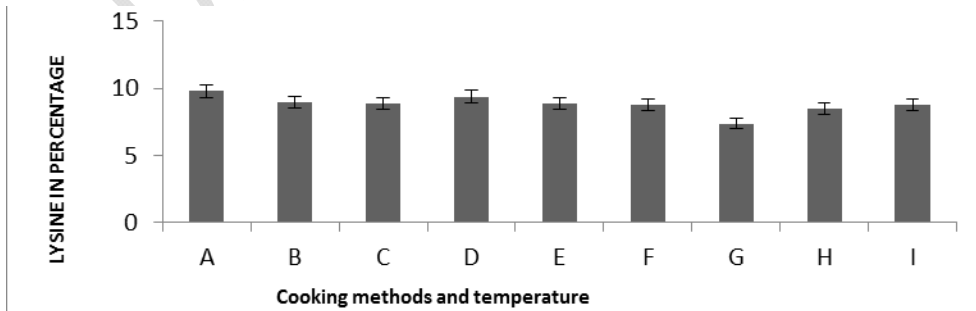
101 time. This perhaps accounted for the increased amino acid contents with increase in boiling time.
 102 However, prolonged boiling at 100°C resulted in more protein loss in meat and therefore the amino acid
 103 contents in meat boiled for 100°C were less than that of meat boiled for 90°C. This observation agreed
 104 with that of [15] that an endothermic transition at a temperature range of 59.6 to 68.4°C for collagen in
 105 chicken breast patties resulted in an increase of soluble protein. The soluble protein dissolved in water
 106 causing decrease in protein content of chicken breast patties.
 107 The amino acid score of breakfast sausage decreased with increased grilling and frying temperature. The
 108 possible reason is that grilling and frying led to formation of heterocyclic aromatic amines [16] and a hard
 109 and dry surface on the meat [17], which made the meat proteins difficult to hydrolyze during amino acid
 110 analysis [18] and caused the meats fried for longer duration to have a low amino acid content.
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 113 **Figure 1: Phenylalanine in percentage**
 114 A= Grilling 80 °C, Grilling B= 90 °C, C= Grilling 100 °C, D= Frying 80 °C, E= Frying 90 °C, F= Frying
 115 100 °C, G= Boiling 80 °C, H= Boiling 90 °C, I= Boiling 100 °C
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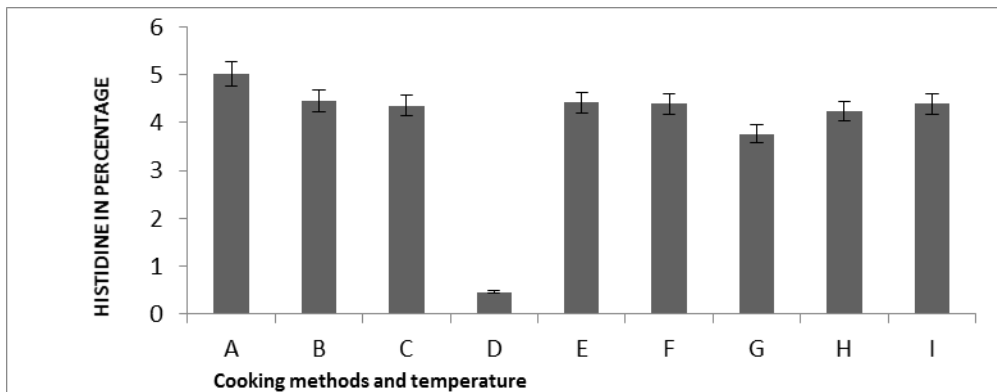


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 118 **Figure 2: Isoleucine in percentage**
 119 A= Grilling 80 °C, Grilling B= 90 °C, C= Grilling 100 °C, D= Frying 80 °C, E= Frying 90 °C, F= Frying
 120 100 °C, G= Boiling 80 °C, H= Boiling 90 °C, I= Boiling 100 °C
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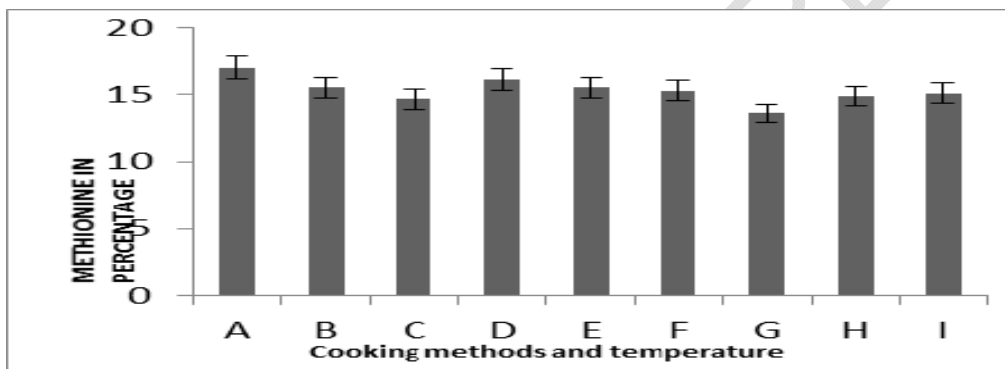


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 123 **Figure 3: Lysine in percentage**

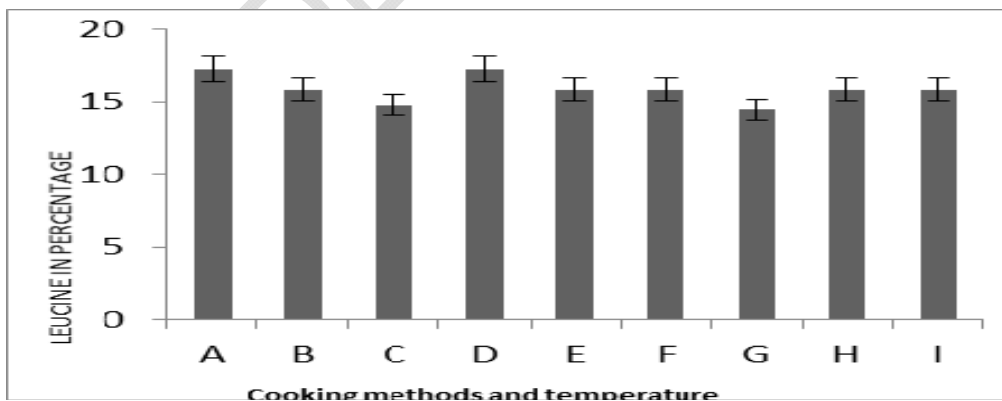
124 A= Grilling 80 °C, Grilling B= 90 °C, C= Grilling 100 °C, D= Frying 80 °C, E= Frying 90 °C, F= Frying
 125 100 °C, G= Boiling 80 °C, H= Boiling 90 °C, I= Boiling 100 °C
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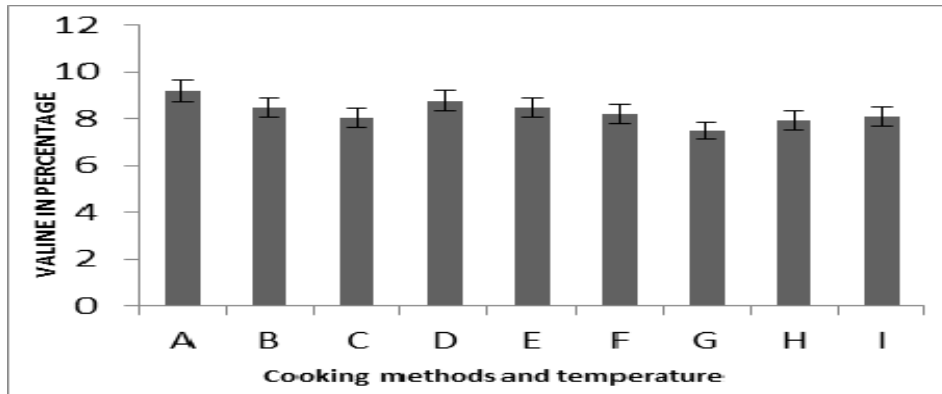
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 129 **Figure 4: Histidine in percentage**
 130 A= Grilling 80 °C, Grilling B= 90 °C, C= Grilling 100 °C, D= Frying 80 °C, E= Frying 90 °C, F= Frying
 131 100 °C, G= Boiling 80 °C, H= Boiling 90 °C, I= Boiling 100 °C
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 136 **Figure 5: Methionine in percentage**
 137 A= Grilling 80 °C, Grilling B= 90 °C, C= Grilling 100 °C, D= Frying 80 °C, E= Frying 90 °C,
 138 F= Frying 100 °C, G= Boiling 80 °C, H= Boiling 90 °C, I= Boiling 100 °C
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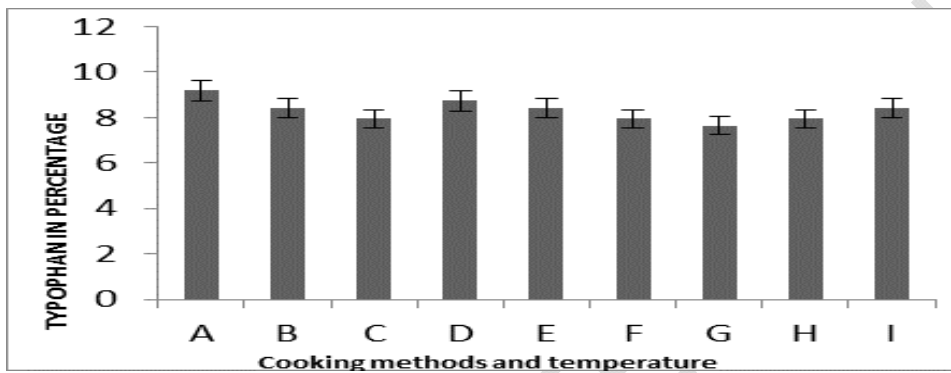
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 141 **Figure 6: Leucine in percentage**
 142 A= Grilling 80 °C, Grilling B= 90 °C, C= Grilling 100 °C, D= Frying 80 °C, E= Frying 90 °C,
 143 F= Frying 100 °C, G= Boiling 80 °C, H= Boiling 90 °C, I= Boiling 100 °C
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Figure 7: **Valine in percentage**

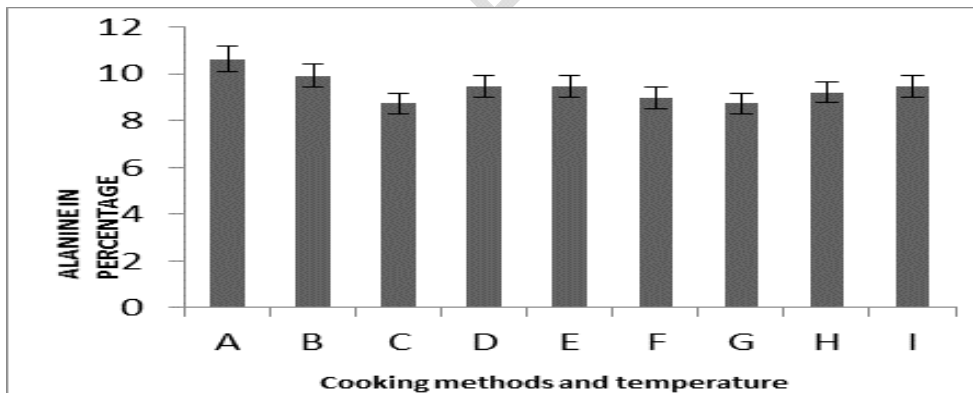
A= Grilling 80 °C, Grilling B= 90 °C, C= Grilling 100 °C, D= Frying 80 °C, E= Frying 90 °C, F= Frying 100 °C, G= Boiling 80 °C, H= Boiling 90 °C, I= Boiling 100 °C



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Figure 8: **Tryptophan in percentage**

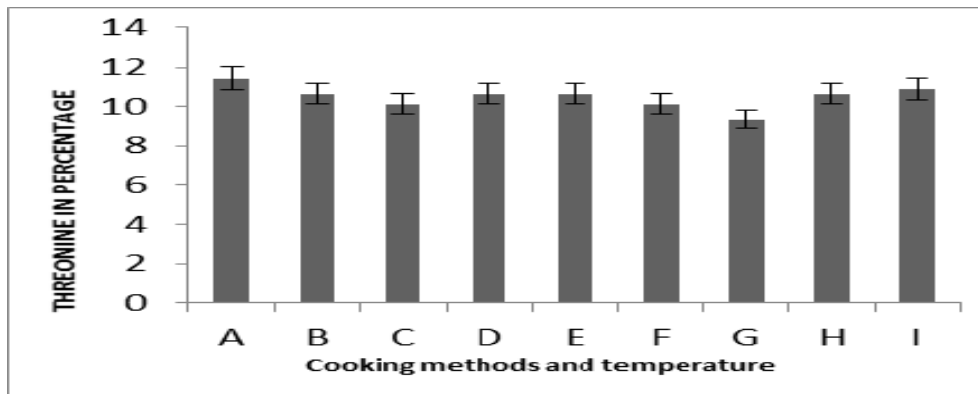
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Figure 9: **Alanine in percentage**

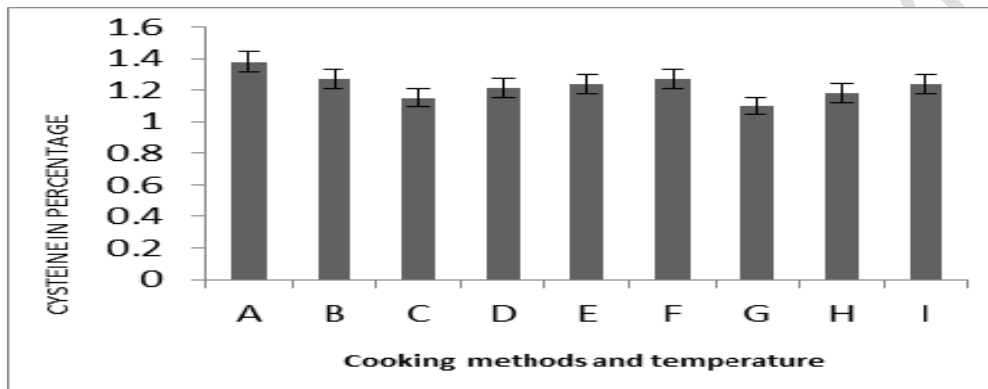
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Figure 10: **Threonine in percentage**

A= Grilling 80 °C, Grilling B= 90 °C, C= Grilling 100 °C, D= Frying 80 °C, E= Frying 90 °C,
F= Frying 100 °C, G= Boiling 80 °C, H= Boiling 90 °C, I= Boiling 100 °C

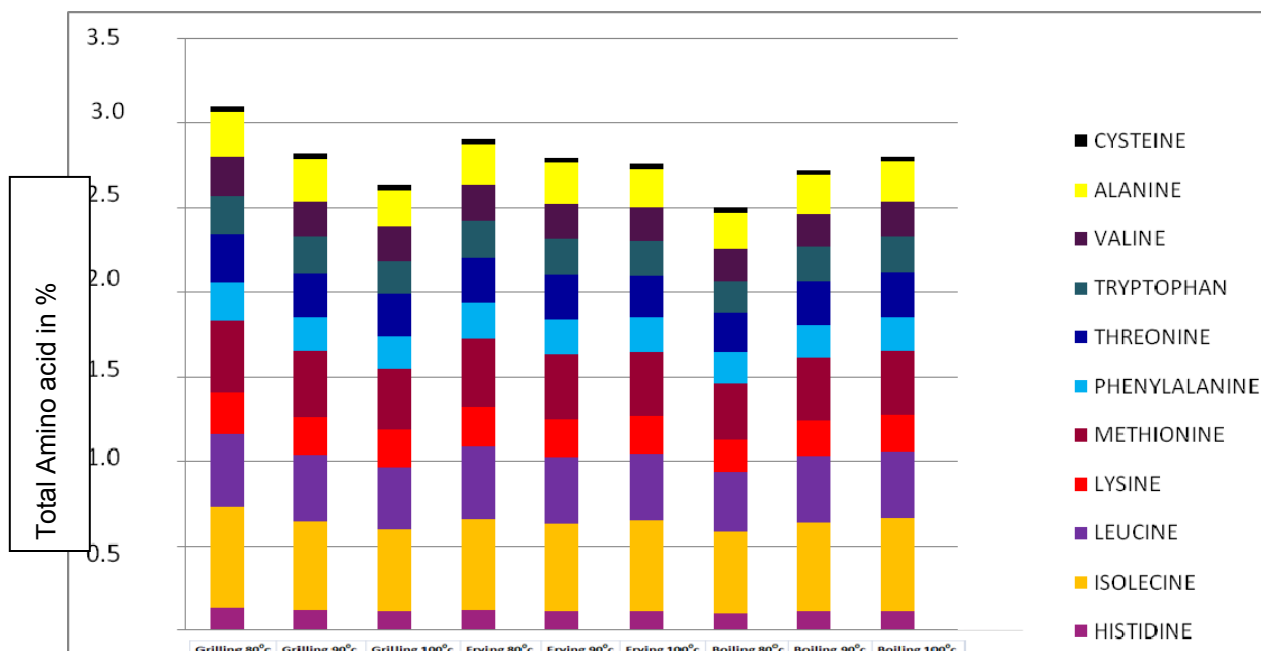


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Figure 11: **Cycteine in percentage**

169 A= Grilling 80 °C, Grilling B= 90 °C, C= Grilling 100 °C, D= Frying 80 °C, E= Frying 90 °C,
170 F= Frying 100 °C, G= Boiling 80 °C, H= Boiling 90 °C, I= Boiling 100 °C

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177 **Figure 12: Essential amino acid score of breakfast sausage cooked at different**
178 **temperature**
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180 **3.2 Proximate composition of breakfast sausage cooked at different**
181 **temperature**

182 The summary presented in Table 4 gives the proximate composition of breakfast sausage
183 cooked at different temperature. The moisture content ranged from 49.16% to 55.38%.
184 Grilled sausage at 80°C had the highest crude protein (25.58%) followed by fried sausage
185 (21.36%) with least crude protein in boiling (10.59%). The fat content of proximate
186 composition of breakfast sausage cooked at different temperature revealed significant
187 differences in fat content but was not following a consistent pattern. The least fat content
188 was recorded for sausage grilled at 80°C (15.99%). The ash content were statistically
189 ($p < 0.05$) different. Grilled sausage at 80°C had the highest ash content (6.66%) with least
190 ash content in boiled sausage at 90°C (1.40 %).
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192 **3.2.1 Moisture content**

193 The moisture content of breakfast sausage at 80 and 90°C across the cooking methods was
194 significantly higher at 54.76 and 53.33%, respectively while grilled sausage recorded highest
195 moisture content at 100°C as shown in Table 4. This revealed that boiling had less influence
196 on moisture content of breakfast sausage cooked at difference temperature compared to
197 grilling and frying that resulted in severe moisture loss depending on the temperature and
198 cooking time.
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200 **3.2.2 Crude protein**

201 Crude protein of breakfast sausage cooked at different temperature revealed that, sausage
202 grilled at 80°C had the highest value (25.58 %) as shown in Table 4, followed by frying
203 (21.36%) with least value observed for boiling (10.59%). The crude protein values revealed
204 that as the cooking temperature increased, there was decreased crude protein in grilled and
205 fried breakfast sausage while the reverse was the case for boiling. This could be as a result
206 of protein denaturation ability of high temperature, time and the amount of water loss during
207 cooking.
208

3.2.3 Ether extract and ash content

209 The fat content of breakfast sausage cooked at different temperature revealed significant
 210 differences but did not follow a consistent pattern. The least fat content (15.99%) was
 211 recorded for grilled sausage at 80°C while ash content was (p<0.05) different as shown in
 212 Table 4, which followed the same pattern with crude protein. Grilling at 80°C had the highest
 213 value for ash which could be as a result of water loss, impact of temperature and cooking
 214 time. Since microwave cooking and grilling proceed in the absence of water (dry cooking
 215 methods), these cooking methods allowed for a high retention of ash than boiling (wet
 216 cooking method). [19] also found a decreased in ash content of rabbit meat after boiling and
 217 roasting. Similar ash values for cooked meat (1.3 to 1.9 g/100 g) were reported by [20].
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Table 4: Proximate composition of breakfast sausage cooked at different temperature

Parameters (%)	Temperature (°C)	Grilling	Frying	Boiling	SEM
Moisture content	80	50.58 ^{cz}	52.19 ^{by}	54.76 ^{ax}	0.51
	90	51.75 ^{by}	49.16 ^{cz}	53.33 ^{cx}	0.61
	100	55.38 ^{ax}	52.81 ^{az}	54.50 ^{by}	0.38
	SEM	0.62	0.56	0.22	
Crude protein	80	25.58 ^{ax}	21.36 ^{ay}	10.59 ^{cz}	2.75
	90	20.27 ^{bx}	18.59 ^{by}	12.29 ^{bz}	1.21
	100	16.86 ^{cy}	18.32 ^{bx}	13.78 ^{az}	0.66
	SEM	1.91	0.48	0.46	
Ether extract	80	15.99 ^{cz}	17.50 ^{cy}	26.24 ^{ax}	1.31
	90	27.99 ^{ax}	24.91 ^{az}	26.14 ^{by}	0.45
	100	20.53 ^{bz}	22.65 ^{by}	24.57 ^{cx}	0.58
	SEM	1.29	1.11	0.27	
Ash	80	6.66 ^{ax}	4.73 ^{bz}	5.66 ^{ay}	0.31
	90	4.82 ^{bx}	1.53 ^{cy}	1.40 ^{cz}	0.58
	100	4.46 ^{by}	5.66 ^{ax}	2.73 ^{bz}	0.64
	SEM	0.34	0.63	0.58	

220 ^{xyz...} Means on the same column with different superscripts are significantly different (p<0.05)

221 ^{abc...} Means on the same row with different superscripts are significantly different (p<0.05)

222 SEM: standard error mean

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225 4. CONCLUSION

226 Breakfast sausage grilled at 80°C had the highest essential amino acid scores, crude protein
 227 and ash.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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UNDER PEER REVIEW