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WEIGHT LOSS, PROXIMATE AND MICROBIAL PROPERTIES OF FERMENTED AND UNFERMENTED SAUSAGES PRODUCED FROM SNAIL AND BEEF BLENDS

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Abstract

Four different batches of sausages were produced from ground snail and beef blends at varying snail: beef ratios (100 : 0, 80 : 20, 60 : 40, 50 : 50) for batch S1 , S2 , S3 and S4 respectively. Another four samples of the same snail: beef ratio were produced and fermented. All the sausage samples were cooked, roasted and then analyzed for quality characteristics. The proximate content showed significant differences ($p \leq 0.05$) among samples. The range of moisture content was 18.54 - 21.81% with unfermented sausages having the highest moisture content, protein content range from 83.73 - 87.02% with fermented sausages having the highest protein content while the fat content ranged from 3.00 - 5.86% with unfermented sausages having the highest value. Cooking losses showed significant difference ($p \leq 0.05$) from roasting losses. Microbial analysis showed that fermented sausages had the highest microbial load of (3.0 x 104 CFU/g). Sensory evaluation of the sausages revealed high mean overall acceptability scores for unfermented and roasted sausages. Generally, the products with 100% snail and 50% snail were most acceptable to the panelists. As such it was concluded that 50% beef or more could be replaced with snail in the production of healthy sausage roll.

Keywords: Snail, beef, sausage, ferment

Introduction

Sausages can be defined as products in which fresh comminuted meats are modified by various processing methods to yield product with great organoleptic and keeping properties (Sanwo, 2017). Sausages are one of the oldest forms of preserved meat products they are usually of high nutritive value, usually produced from parts of carcass (Jihad, 2009). They are either comminuted (particle size reduction/ground) seasoned meats, stuffed or unstuffed into casing. Whereas, Merinoff (2007) defined sausages as food made from ground meat with back fat, salts, herbs and spices.

Typically, sausages are stuffed into cellulose casing or animal intestine. These sausages are cylindrically linked and vary in size depending on the type of sausage. According to an online etymology dictionary, sausages are classified into fresh, cooked, roasted, and dry sausages. They differ depending on their ingredients, shape, production technique, level of dryness and whether fresh or cooked. According to Marchello (2012), the contemporary role of sausage fits conveniently into our modern lifestyles as an elegant appetizer for entertaining as well as the main course in "quick and easy" meals.

Sausages have been produced from different meats such as beef, pork, chicken, fish and buffalo meat (Raju et al., 2003; Salam et al., 2004 and Sachindra et al., 2005) and have been classified as nutritious (FDA, 2001). Ukpong (2009) has reported that the prevalence of malnutrition and iron deficiency in school children could be reduced in her country by incorporating snail meat in their diet. This is because snail meat contains protein, fat (mainly polyunsaturated fatty acid), iron, calcium, magnesium, phosphorus, copper, zinc, vitamins A, B6, B12, K and folate. It also contains the amino acids arginine and lysine at higher levels than in whole egg. It also contains healthy essential fatty acids such as linoleic and linolenic acids. The high-protein, low-fat content of snail meat makes it a healthy alternative food. However, due to the high cost of animal and plant protein sources, the search for cheap and affordable source of protein has increased considerably.

In the last two decades attention has therefore been shifted to heliculture, which is the practice of raising snails in captivity like other farm animals. The reason for this being that snail meat competes favourably with other protein sources in protein amino acid and vitamins (Ademola et al., 2007). Snails are unconventional meats with high protein and low fat contents and are of major dietary importance (Ademola et al., 2007). They are cheap, affordable and readily available source of protein with very low cholesterol and essential fatty acid content. Snails have high percentage of minerals such as calcium, iron, selenium and magnesium. They can be processed and consumed in many ways: roasted, dried or boiled. Onwuka and Okorie (2004) assessed the quality of sausages from combination of fermented and unfermented snail and African oil bean, all the products were generally accepted.

The consumption of snail therefore offers better nutritional benefits as compared to the conventional meat protein used in processed meats which have high cholesterol and saturated fatty acids and have been implicated in coronary heart diseases and arteriosclerosis (Leisner et al., 2002). Consequently, production of snail sausages with blends of beef will afford the various interest groups the opportunity to eat and enjoy the sausage without fear of beef or pork related diseases.

This research will help provide new ideas on how best to formulate snail/beef sausage with high nutritional value, appealing sensory attributes and good aesthetic value. Finally, the production of snail/beef sausages, if perfected and generally accepted, will widen the variety of processed meat produced in food industries, thereby creating more jobs opportunities, improve commercial utilization of snails and make readily available the nutritional benefits of consuming snails.

Materials and Methods

Material procurement

Life Snails were purchased from a snail farmer at Itam market in Uyo, fresh meat from the thigh muscle of a matured cattle was purchased from a reputable slaughter slab in Itam, Uyo, for sausage production and quality assessment. Other ingredients and seasonings were purchased from Akpan Andem market, Uyo, Akwa Ibom State.

Raw material preparation

The snails were de-shelled to expose the edible flesh and then thoroughly washed with water. The snail meat was cut into smaller pieces and refrigerated for five hours. The beef was also washed thoroughly in conformity to FSA (2002) then cut into smaller pieces and refrigerated for five hours.

Sausage preparation

Four batches of sausages (500g of meat per batch) using varying ratios of snail to beef (100: 0, 80:20, 60:40, 50:50) were formulated. Another four batches with the same snail to beef ratios as the first four batches were replicated and used for fermented sausages. The prepared meat was run through a 5mm plate mincing machine. The minced meat (93.28g) was then mixed with 1.87 of vegetable oil, 3.73 kg starch and 1.12 kg of seasoning as indicated in the recipe table (Table 1.)

The sausage mixtures were stuffed into an already washed goat intestine for roasting and cellulosic casing for cooking. Each batch was labelled and replicated twice. The batches for fermentation were left to ferment for 48 hours at ambient temperature.

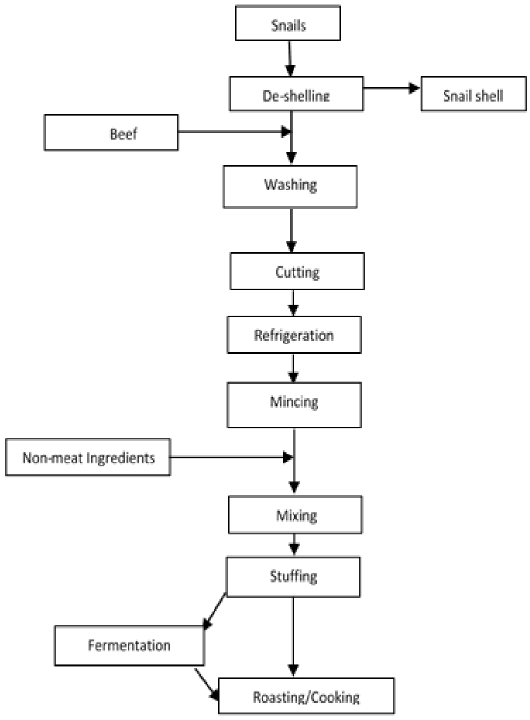

Figure 1: Flow chart for the production of beef/snail sausage

Table 1: Recipe formulation of the sausages.

Ingredients	Fermented				Unfermented			
Samples	1	2	3	4	5	6	7	8
Snail (g)	93.28	46.64	55.97	74.63	93.28	46.64	55.97	74.63
Beef (g)	0	46.64	37.31	18.65	0	46.64	37.31	18.65
Oil (ml)	1.87	1.87	1.87	1.87	1.87	1.87	1.87	1.87
Starch (g)	3.73	3.73	3.73	3.73	3.73	3.73	3.73	3.73
Seasoning (g)	1.12	1.12	1.12	1.12	1.12	1.12	1.12	1.12
Total (g)	100	100	100	100	100	100	100	100

Table 2: Proportions (%) of Snail: Beef combination used for the sausages

Raw Material	S_1	S_2	S_3	S_4
Snail (%)	100	50	60	80
Beef (%)	0	50	40	20

Determination of weight Losses of sausage during cooking or roasting

Samples of sausages were replicated thrice and prepared alike each sample in its casing were dropped in 80ºC. 600ml of hot water and cooked till an internal temperature of 85ºC was attained for the sausages, while another roasted at 85ºC The samples were allowed to cool to room temperature and their final weights taken cooking or roasting weight loss was calculated as follows:

Cooking/Roasting loss (R) = (Weight before cooking/ roasting - Weight after cooking/roasting)

$$Cooking\ or\ Roasting\ loss\ (\%) = \frac{(Wt\ before\ cooking\ or\ roasting - Wt\ after\ cooking\ or\ roasting)}{Wt\ before\ cooking\ or\ roasting}\ x\ 100$$

Proximate evaluations

Moisture, ash, crude protein, carbohydrate and crude fat were determined according to the method of AOAC (2000).

Microbial analysis

Serial dilution: 10g of each sample were weighed aseptically and homogenized in 90ml sterile peptone water. Then, serial dilutions was made by mixing 1.0ml of the suspension in 9.0ml sterile peptone water to obtain 10^{-1} dilution. The dilution was then made to 10^{-5} diluents, then poured into Petri dishes. The agar was then poured into the plate aseptically according to the manufactural's instruction. The agar used are nutrient agar (for total viable counts), Robertson cooked meat medium (for *Clostridium botulinum*), Eosin Methylene blue agar (for enteric bacteria) and Sabouraud dextrose agar. The plates were made in triplicates and incubated at 37ºC for 24 hours. Total number of cells per gram of samples were then estimated after counting the colonies on the plates (Prescott *et al*. 2002; Ezeama, 2007).

Results and Discussion

Weight losses

Significant difference (p<0.05) existed in the batches between fermented and unfermented sausages as shown in Table 2. Percentage weight losses of the sausages ranged from 0.21 to 1.43% in fermented sausages and 0.78 to 0.97% in unfermented sausage with fermented sausages having the highest percentage weight losses of 1.43%. This is likely as a result of the fact that fermentation process requires the use of some nutrients and water. This fell within the acceptable range of less than 10% (Wilson, 1980). However, the low cooking loss may have been as a result of the re-absorption of moisture due to the permeability of the intestine used as packaging material. It was also observed that the lesser the moisture loss, the higher the tenderness of the final products.

There was a significant difference (p<0.05) in roasting weight losses for the batches in fermented and unfermented as shown in Table 2. Percentage weight losses of roasted sausages ranged from 1.81 - 2.12% which falls within the acceptable range of not more than 15% for fermented (dry and semidry) sausages (Rust, 2014). However, weight losses in unfermented batches are greater than that of the fermented batches; this could be due to the activities of micro-organisms in in utilizing some constituents and nutrients in the sausage during the fermentation process. It was also observed that there were shrinkages in the final product; this may have been as a result of moisture loss during roasting.

Proximate composition of snails/beef sausages

From Table 3, there were significant differences (p<0.05) in the proximate composition parameters. The moisture content of the sausages produced ranged from 18.54 - 21.81% with sample S6 having the highest value as compared to 19.20% for sample S3. This range compares reasonably with the maximum 25% recommended for shelf stable sausages (Boyle, 1994). It was observed that the unfermented sausages had higher moisture content than the fermented sausages. This could be attributed to the fermentation process as fermented sausages are often referred to as dry or semi dry sausages.

It was also observed that the lesser the moisture loss, the higher the tenderness of the final products. There was a significant difference (p < 0.05) in roasting weight losses for the batches in fermented and unfermented as shown in Table 2. Percentage weight losses of roasted sausages ranged from 1.81 - 2.12% which falls within the acceptable range of not more than 15% for fermented (dry and semidry) sausages (Rust, 2014). However, weight losses in

unfermented batches are greater than that of the fermented batches; this could be due to the activities of micro-organisms in in utilizing some constituents and nutrients in the sausage during the fermentation process. It was also observed that there were shrinkages in the final product; this may have been as a result of moisture loss during roasting.

Table 2: Weight losses of fermented and unfermented sausages

		Cooking weight losses				Roasting weight losses			
Sample	Snail : beef ratio	Initial weight (g)	Final weight (g)	Weight loss (g)	Weight loss (%)	Initial weight (g)	Final weight (g)	Weight loss (g)	Weight loss (%)
S_1	100:0	85	83.78	1.22	1.43	85	83.24	1.76	2.07
S_2	50:50	85	84.66	0.34	0.40	85	83.40	1.60	1.88
S_3	60:40	85	84.59	0.41	0.48	85	83.46	1.54	1.81
S_4	80:20	85	84.82	0.18	0.21	85	83.20	1.80	2.12
S_5	100:0	90	89.12	0.88	0.97	90	88.94	1.06	1.18
S_6	50:50	90	89.24	0.76	0.84	90	88.23	1.77	2.08
S_7	60:40	90	89.29	0.71	0.78	90	88.24	1.76	2.07
S_8	80:20	90	89.17	0.83	0.92	90	88.28	1.72	1.90

Keys: S_1=Fermented with 100% snail and 0% beef inclusion, S_2=Fermented with 50% snail 50% beef inclusion, S_3 =Fermented with 60% snail 40% beef inclusion, S_4=Fermented with 80% snail 20% beef inclusion, S_5=Unfermented with 100% snail and 0% beef inclusion, S_6=Unfermented with 50% snail and 50% beef inclusion, S7=Unfermented with 60% snail and 40% beef inclusion, S8=Unfermented with 80% and 20% beef inclusion

Proximate composition of snails/beef sausages

From Table 3, there were significant differences ($p<0.05$) in the proximate composition parameters. The moisture content of the sausages produced ranged from 18.54 - 21.81% with sample S_6 having the highest value as compared to 19.20% for sample S_3. This range compares reasonably with the maximum 25% recommended for shelf stable sausages (Boyle, 1994). It was observed that the unfermented sausages had higher moisture content than the fermented sausages. This could be attributed to the fermentation process as fermented sausages are often referred to as dry or semi dry sausages.

The protein content was significantly increased for sausages with higher inclusion level of snail, this agrees with Fagbuaro *et al.*, (2006) and Ademosun and Imerbore (1988). As observed in Table 3, the percentage of protein content in fermented sausages was higher in fermented sausages than in the unfermented. The increases in the percentage of protein could be attributed to the known fact that fermentation improves the nutritional quality of food products through the increase in microbial population and microbial mass during fermentation with extensive hydrolysis of the protein molecules to amino acids and other simple peptides (Saldanha et al., 2001). The Ash content of snail/beef sausages ranged from 5.83 - 4.40% with sample S1 having the highest value, compared to the unfermented sample with the same inclusion level. The high ash content in the product showed that the sausages are rich in minerals and this agrees with Saldanha et al. (2001) and Ademolu et al. (2007) that snail has rich mineral content.

The fat content of the produced sausages as recorded in Table 3 range from 5.86 - 30.00% with S6 having the highest value. These were significant differences ($p<0.05$) among the samples except for S4 (fermented sample with 80% snail + 20% beef) and S5 (Unfermented sample with 100% snail + 0% beef). From the result, unfermented samples S5, S6, S7 and S8 (unfermented with 100% snail and 0% beef, unfermented with 50% snail and 50% beef inclusion, unfermented with 60% snail and 40% beef and unfermented with 80% and 20% beef) had higher fat values than S1, S2, S3 and S4 (fermented with 100% snail+ 0% beef, sample fermented with 50% snail + 50% beef, sample fermented with 60% snail + 40% beef, and sample fermented with 80% snail + 20% beef). The lower fat content of the fermented samples (S1, S2, S3 and S4) could be attributed to the activities of micro-organisms during fermentation which degrade fats and convert the lipoprotein to micro-protein subsequently reducing the fat content in fermented sausages. It was also observed that the higher the inclusion levels of beef, the higher the fat percentage.

The crude fibre and carbohydrate ranged from 2.89 - 2.12% and 3.95 - 1.60% respectively and there were significant differences ($p < 0.05$) amongst the samples for both determinations.

Microbial analysis of sausages

Total fungal count of sausages as shown in Table 4 indicated contamination with spoilage micro-organisms for sample S1 to S8 with the exception of S4 as the permissible limit is 105 cfu/g (Kheyri et al., 2014). The highest count being 1.2 x 105cfu/g. It was also observed that the higher the inclusion level of beef the higher the fungal count. This could be attributed to higher microbial contamination of the beef than the snail due to micro-organisms contamination during abattoir operations and also due to the fact that beef is prone to rapid spoilage (Benain et al., 2014). Unfermented samples (S5, S6, S7 and S8) were all observed to have higher counts than the fermented samples (S1, S2 and S3). This could be as a result of the chance fermentation which allowed the growth of spoilage micro-organism and subsequent contamination. However, the total plate count did not exceed 105cfu/g; which would have been highly suggestive of unwholesomeness according to the Department of Health (Sofos, 1994) and USDA (1999). Total bacterial counts were higher in fermented sausages than in the unfermented sausages.

Table 3: Proximate composition of fermented and unfermented sausages

Parameters	**S_1**	**S_2**	**S_3**	**S_4**	**S_5**	**S_6**	**S_7**	**S_8**
Moisture (%)	18.54^{g} ± 0.02	20.70^{d}±0.04	19.20^{e}±0.05	18.69^{f}±0.06	21.08±0.04^{c}	21.81±0.01^{a}	21.19±0.04^{b}	21.10±0.07c
Protein (%)	87.0.2^{a}±0.01	85.63^{e}±0.06	85.7^{d}±0.01	85.87^{c}±0.03	85.97±0.01^{b}	83.73±0.02^{h}	83.87±0.01^{g}	84.56±0.02^{f}
Fat (%)	3.00^{h}±0.06	3.74^{e}±0.02	3.59^{f}±0.01	3.36^{d}±0.07	4.15±0.03^{d}	5.86±0.01^{a}	5.79±0.01^{b}	5.18±0.03^{c}
Ash (%)	5.83^{a}±0.01	4.73^{a}±0.04	4.89^{c}±0.01	4.72^{d}±0.03	5.30±0.02^{b}	4.43±0.03^{f}	4.40±0.05^{f}	4.58±0.01^{e}
Crude Fibre (%)	2.52^{b}±0.02	2.22^{c}±0.02	2.19^{c}±0.05	2.16±0.01^{d}	2.89±0.03^{a}	2.20±0.01^{c}	2.18±0.05^{c}	2.12±0.02^{d}
Carbohydrate (%)	1.60^{f}±0.02	3.67^{c}±0.01	3.62^{c}±0.01	3.95±0.02^{a}	1.69±0.07^{e}	3.78±0.01^{b}	3.76±0.05^{b}	3.56±0.02^{d}

NB: Values are means ± SD of triplicates. Values with different superscripts within the same row are significantly different ($p < 0.05$).
Keys: S_1=Fermented with 100% snail and 0% beef inclusion, S_2=Fermented with 50% snail 50% beef inclusion, S_3 =Fermented with 60% snail 40% beef inclusion, S_4=Fermented with 80% snail 20% beef inclusion, S_5=Unfermented with 100% snail and 0% beef inclusion, S_6=Unfermented with 50% snail and 50% beef inclusion, S7=Unfermented with 60% snail and 40% beef inclusion, S8=Unfermented with 80% and 20% beef inclusion

Table 4: Fungi count/taxonomy of produced sausages

Sample code	Total coliform count (cfu/g)	Total Heterotrophic Bacterial Count (cfu/g)	Total Clostridium Count (cfu/g)	Total Fungal Count (cfu/g)
S_1	2.05 x 10^3	2.5 x 10^4	2.9 x 10^4	1.0 x 10^5
S_2	2.5 x 10^3	3.0 x 10^4	2.4x 10^4	1.3 x 10^5
S_3	2.01 x 10^3	2.8 x 10^3	2.4 x 10^4	1.2 x 10^5
S_4	2.00 x 10^3	2.5 x 10^4	2.2 x 10^4	-
S_5	1.0 x 10^3	1.2 x 10^3	1.30 x 10^3	1.0 x 10^5
S_6	1.2 x 10^3	1.4 x 10^3	1.20 x 10^3	1.2 x 10^5
S_7	1.0 x 10^3	1.5 x 10^3	1.0 x 10^5	1.2 x 10^5
S_8	1.13 x 10^3	1.4 x 10^3	1.5 x 10^5	1.0 x 10^5

Key: S_1 = Fermented with 100% snail and 0% beef inclusion, S_2 = Fermented with 50% snail 50% beef inclusion, S_3 = Fermented with 60% snail 40% beef inclusion, S_4 = Fermented with 80% snail 20% beef inclusion, S_5 = Unfermented with 100% snail and 0% beef inclusion, S_6=Unfermented with 50% snail and 50% beef inclusion, S_7 = Unfermented with 60% snail and 40% beef inclusion, S_8 = Unfermented with 80% and 20% beef inclusion.

Coliform count had a range of 1.0×10^3 - 2.5×10^3. The unfermented sausages (S5, S6, S7 and S8) had lower counts than the fermented samples (S1, S2, S3 and S4). The count decreased with decrease in beef inclusion in fermented and unfermented samples. This suggests that the beef may have had higher level of contamination than the snail.

Total Clostridium count ranged from 1.20× 10^3 - 1.5 × 10^5. The counts also reduced with reduction in the amount of beef just as observed in Coliform count. The unfermented sausage with 80% snail + 20% beef (S_8) had the highest count of 1.5 × 10^5 cfu/g while unfermented sample with 50% snail + 50% beef had the lowest value of 1.20× 10^3 cfu/g.

Conclusion and Recommendation

The study showed that the unfermented sausage with 0% beef inclusion level was rated best based on its organoleptic properties. Followed by the sausage with 46.64% beef inclusions. The fermented sausages due to its sour flavor and undesirable taste was least preferred. This was as a result of the fermentation process which provides optimum conditions and time for the activities of lactic acid producing bacteria, thus the off-flavour. This microbiological change however had impressive results on the nutritional composition of the fermented sausages such as the increase in protein content. Microbial counts were also higher in fermented sausages than in the unfermented sausages.

Based on the results from this study, its recommended that the production of sausages with snail/beef inclusion should be done at the level of S_1 and S_2. Also, fermentations of sausages should be done in temperature-controlled rooms and not left to chance fermentation, and/or starter cultures should be used for its fermentation in order to reduce microbial load.

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