



## Sensory Properties, Proximate, Vitamin and Anti-Nutrient Composition of *Awara*-Based *Kilishi* Analogue with Suya Spice

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### Abstract

*Awara*-based *kilishi* analogue with suya spice was produced and subjected to sensory, proximate, vitamin and antinutrient analysis using standard methods. The *awara*-based *kilishi* formulated with *awara*:suya spice ratio of 83.33:16.67, 79.85:20.15 and 75.62:24.38 showed percentage acceptability index of 82.78, 85.56 and 80.0%, respectively, which means high consumers acceptance and was further analyzed. The *awara*-based *kilishi* with suya spice with ratio 83.33:16.67 had the highest protein [35.04%], fiber (0.63%) and fat [22.92%] level while 75.62:24.38 ratio had the highest ash [3.99%] and carbohydrate [38.88%] with least moisture of 5.08%. Sample ratio 75.62:24.38 contained the highest level of vitamin B<sub>12</sub> [230 µg/100], B<sub>6</sub> [1020µg/100], B<sub>1</sub> [420µg/100] and C [35.40mg 100g<sup>-1</sup>]. Stachyose, trypsin, lectin, phytate, glycine and raffinose content of the *awara*-based *kilishi* analogue samples ranged from 0.54 to 0.83, 1.15 to 1.19, 0.14 to 0.19, 2.19 to 2.25, 0.22 to 0.73 and 0.24 to 0.27mg 100g<sup>-1</sup> and were all within acceptable limits. The addition of the suya spice notably enhanced the ash and crude fiber content of the *awara*-based *kilishi* compared to the control [100% *awara*-based *kilishi* without suya spice] sample. Findings showed the product to be a viable alternative to traditional animal-based *kilishi* when considering health and environmental effect.

**Keyword:** Plant-meat, sustainable protein, high-quality, soybean

### Article History

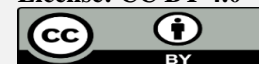
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### Introduction

Even with clear health and environmental concerns, global meat consumption is projected to grow by 14% by 2030 compared to 2018–2020 levels [1]. High meat intake is linked to food security challenges, animal welfare issues, resource depletion, greenhouse gas emissions, and public health risks, including cardiovascular and zoonotic diseases [2]. Red and processed meat consumption increases the risk of heart disease, stroke, type 2 diabetes, and certain cancers, contributing to higher mortality, particularly in high-income countries [3,4,5]. Reducing meat intake enhances human health and environment sustainability. Switching to flexitarian feeding habit also causes a decrease in the annual greenhouse gas emissions to 583 MtCO<sub>2e</sub> in addition to decrease in premature mortality rate of 52,700/year and enhanced obesity management [6,7]. Meat substitutes provide adequate protein with lower saturated fat and no cholesterol [2]. Substitution of five percent beef consumption with plant-based protein such as pea can reduce emissions by eight millions tones yearly. Meats from plant-based showed reduced greenhouse gas

emissions up to 98% with 79% less land use, 95% less water use, 89% less air pollution and 92% less water pollution [8,9]. Diversifying protein sources with plant proteins therefore supports more resilient, resource-efficient, and sustainable food systems. Bonales et al. [9] said that plant-based meat industry is growing quickly, reaching USD 10 billion in 2022 and expected to nearly triple by 2027. This growth is fueled by increasing global interest in meat alternatives driven by health, lifestyle, environmental and ethical concerns. Plant-based proteins are scalable, relying on existing agricultural systems instead of livestock, with costs likely to fall as production expands. It also presents localization opportunities using locally grown crops such as soybean, pea, enabling rapid global expansion.

*Kilishi* is a spiced traditional dried meat delicacy commonly consumed in West Africa. *Kilishi* is known for its unique flavour, high protein content and long shelf stability. However, the increase in the demand for affordable and sustainable sources of protein has encouraged plant-based meat analogues

development that mimic the sensory and nutritional qualities of conventional meat products [10,11]. Soybean is one of the best sources of plant-based accessible and sustainable protein. Soybean is a northern grown and widely used in Nigeria for several traditional food products such as beverages, drinks, *awara* etc. soybean-based food products are mostly used to complement other foods such as akamu. *Awara* has limited use as it is only used traditionally as deep fried snack sold in most local markets in northern parts of Nigeria.

*Awara* [soy curd] is a soy-based protein product locally processed and consumed in northern region of Nigeria and it stands as a promising meat substitute. Nutritionally, *awara* is rich, giving high-quality protein, essential amino acids, minerals and vitamins and also serves as a versatile foundation for meat-analogue innovations and also has potential for utilization in a wide range of other application [12].

Suya spice [yaji] is a peanut and chilli-based condiment blend used in preparing grilled meat such as suya to enhance both flavour and cultural acceptability. It contributes bioactive compounds, minerals and lipids while also delivering characteristic sensory properties [13, 14]. Because it has the ability to enhance flavour and taste, its addition to *kilishi* would be highly complementary and adequate towards enhancing the sensory acceptability of the *awara*-based *kilishi*. Suya spice [yaji] is a composite blend containing nutrient-dense ingredients such as ground peanuts [25–60%], chili pepper, ginger, garlic, and onions, providing a broader range of nutrients and bioactive compounds than single-ingredient spices [15]. The significant level of peanut in suya spice supplies the healthy fats, protein and minerals which are not significantly found in other blends of spice. The sustainable benefit [nutritional and environmental] that suya spice provide is majorly due to its peanut component and peanut is a nitrogen-fixing legume with the ability to improve fertility of the soil which in turn reduces the dependence on synthetic fertilizer use. Also, peanut farming needs less water, land and carbon input compare with other many nuts. The remaining component of the suya spice contributes antioxidants and anti-inflammatory compounds in addition [15].

It is also important to note that soy-based analogues often come with its challenge of harboring antinutrients like phytates, tannins and trypsin inhibitor that reduce mineral bioavailability and protein digestibility [16, 17] and as such determining the anti-nutrient of the product becomes crucial.

Findings from the above project could serve as a guiding formulation strategy, improving processing practices and fostering commercial opportunities for *awara*-based *kilishi* with suya spice. Considering the nutritional significance of *kilishi*, its socio-cultural value and growing acceptance of soy products, evaluating the sensory properties, proximate composition, vitamin and antinutrient content of *awara*-based *kilishi* analogue with suya spice is of importance.

## Materials and Methods

### Raw sample procurement and processing

The raw soybean was purchased from Eke-Awka Market in Awka South, Anambra State, Nigeria while the suya spice was

bought from a local producer by name Ijioma [MSc student in Food Science Technology Department, Faculty of Agriculture, Nnamdi Azikiwe University Awka, Anambra State, Nigeria].

### Soymilk processing

The soymilk was produced according to the method of soymilk extraction as described by Adelekan et al. [18] with modifications [use of steeped *Akamu* water, the volume of the steeped water, grinding water used and quantity of soybean used]. The soybean [1058.82 grams] was sorted, washed, soaked for an hour, drained, dehulled, grinded and sieved with 12500ml of water. The extracted soymilk was boiled in an aluminum pot for 20 minutes at 100°C.

### *Awara* [soy curd] processing

According to the method described by Chang and Liu [19] with modifications [type and quantity of coagulant] of *Awara* was produced. A 7260ml of steep water from *Akamu* fermentation was added to the boiled soymilk [20 minutes at 100°C] and allowed to stand for 30 minutes for coagulation to occur. It was then poured into a cheese cloth and lightly pressed by placing a weight on the tied cheese cloth for the water to drain and the *Awara* [curd] was formed after the water was drained. The formed curd is known as *Awara*.

### *Awara*-based *kilishi* analogue production

The *Awara* *kilishi* was produced using the *awara* obtained from coagulating soymilk. The suya spice was added to the *awara* at different ratios as designed, mixed together, cut into desired shapes and oven baked for 45 minutes at 150°C to obtain *awara*-based *kilishi*.

### Sensory evaluation and acceptability index

As described by Odoh et al. [20], a 9-point Hedonic scale using 30 semi-trained panelist was adopted for the sensory evaluation where the appearance, texture, flavour, taste and overall acceptability were scored while the acceptability index was determined by calculation. The samples of *awara*-based *kilishi* with suya spice was evaluated by the semi-trained panelists using a structured evaluation scale, rating each attribute on a 9-point Hedonic scale ranging from 1 [dislike extremely] to 9 [like extremely].

The acceptability index was generated from the mean scores of each sensory parameter using a known formula such as:

$$\% \text{ acceptability index} = \frac{\text{mean score of the product} * 100}{\text{Maximum possible score}}$$

Where acceptability index greater or equal to 70% is generally considered an acceptable level of consumer acceptance for a food product.

### Proximate composition

Proximate composition [moisture, protein, fat, fiber, ash, and available carbohydrate] was determined using standard methods [21]. Moisture and ash were measured by weight difference, fat by petroleum ether extraction, protein by the micro-Kjeldahl method [N × 6.25], and dietary fiber by the gravimetric method. Available carbohydrate was calculated by difference: Carbohydrate [%] = 100 – [protein [%] + fat [%] + ash [%] + fiber [%]].

### Vitamin content determination

Vitamin B<sub>1</sub>, B<sub>6</sub>, B<sub>12</sub> and C were done according to the method of AOAC [22] as described by Odoh et al. [20].

Thiamin [vitamin B<sub>1</sub>]: standard thiamin solutions were reacted with potassium ferricyanide–sodium hydroxide reagent, and absorbance was measured at 367 nm to generate a calibration curve. Sample solution [2 mL] was reacted with the reagent, extracted with isobutyl alcohol, dried over anhydrous sodium sulfate, and the absorbance of the organic layer was measured at 367 nm using isobutyl alcohol as the blank. Thiamin concentration was calculated from the standard curve and expressed as percentage vitamin B<sub>1</sub> using the specified formula.

Vitamin B<sub>6</sub>: Sample [10 g] were homogenized in 0.1 N HCl and autoclaved at 121 °C for 30 min. After cooling, the pH was adjusted to 4.5, followed by enzymatic digestion with takadiastase, acid phosphatase, and β-glucosidase at 37 °C for 18 h. The extract was diluted to volume, filtered [0.4 μm], absorbance measured at 220 nm and calculate percentage value recorded.

In determining vitamin B<sub>12</sub>, 1g of sample was extracted with distilled water, oxidized with potassium ferricyanide and sodium nitrate, heated, treated with formaldehyde, filtered, absorbance read at 361nm and recorded.

Using iodometric titration method vitamin C was determined by extracting the sample with zinc sulfate and acetone, centrifuged and treated with acetic acid. Starch, an indicator was added to the extract, titrated with sodium thiosulfate [0.5N] and calculation expressed as mg/100g.

### Antinutrient content determination

According to Siddiq et al. [23], raffinose and stachyose were determined by oven-drying the samples at 60 °C, milled [0.5mm] and extracted with 70% ethanol [10ml/g dry mass]. HPLC with a refractive index detector was used to analyzed the supernatant of the sonicated and centrifuged extract. Rezex RNM carbohydrate column with water mobile phase [0.4 mL/min] was used for the separation while maintaining the column and detector temperatures at 85 °C and 45 °C, respectively. External raffinose and stachyose standards were used for quantification.

Based on AOAC [22], phytate was determined by extracting 1g of sample with 10 mL of 0.2 N HCl, stirred for 30 min, reacted with ammonium iron [III] sulfate solution, boiled for 30 min, cooled, and centrifuged [3000 rpm] for 30 min. one ml of the supernatant was mixed with 2,2'-bipyridine reagents, and read the absorbance at 519 nm against distilled water. by using the standard curve [3–30 μg/mL phytate-phosphorus] phytate concentration was quantified.

Determination of trypsin inhibitor was done with azocasein method as described by AOAC [22] where 30g/L azocasein was prepared in 100 mM Tris buffer [pH 8.5] containing 5 mM CaCl<sub>2</sub> at 37 °C. Fresh trypsin of 0.3–0.4 mg/mL was prepared in 1 mM HCl. Samples were dispersed in pH 3.3 acetic acid, homogenized, and serially diluted to 50% inhibition. A 125 μL aliquots were incubated with trypsin or water [control], azocasein added and incubated at 37 °C for 30 min. addition of 15% TCA stopped the reaction, centrifuged [15,000 g, 10 min, 4 °C], and the supernatant neutralized with 1.5 M NaOH. Absorbance read at 450 nm, and calculated the trypsin inhibitory activity from linear regression relative to the positive control and expressed as mg/g sample.

Glycine: following Somiari and Balogh [24], 5g of samples was extracted with 70% aqueous ethanol under agitation, filtered, and reacted with ninhydrin using a distillation setup. The resulting chromophore was developed with chromotropic acid and sulfuric acid, heated, cooled, and quantified colorimetrically.

Lectin: using the method of Harborne [25] 2g of sample was extracted with 0.9% NaCl, allowed to stand, stirred, and the absorbance of the supernatant was measured at 420 nm.

### Statistical analysis

The data obtained were analyzed using ANOVA with SPSS software [version 23]. Means were separated using Duncan's Multiple Range Test to determine if there were significant differences among the various parameters in the awara-based kilishi analogue with suya spice.

## Result

### Sensory evaluation of awara-based kilishi analogue with suya spice

The sensory evaluation of awara-based kilishi analogue with suya spice is shown in Table 1a. The values for the appearance, texture, flavour, taste and overall acceptability ranged from 6.50-7.95, 5.9-7.60, 4.6-7.20, 4.85-7.25 and 5.45-7.7, respectively at 9-point Hedonic scoring. There was no significant difference [ $p > 0.05$ ] in the appearance, texture, flavour, taste and overall acceptability for sample ration [awara:suya spice] 100:0, 83.33:16.67, 91.68:8.32, 79.85:20.15, 75.62:24.38; 83.33:16.67, 91.68:8.32, 79.85:20.15, 75.62:24.38; 100:0, 83.33:16.67, 88.24:11.76, 91.68:8.32, 79.85:20.15, 75.62:24.38; 100:0, 83.33:16.67, 88.24:11.76, 91.68:8.32, 79.85:20.15, 75.62:24.38; 100:0, 83.33:16.67, 79.85:20.15, 75.62:24.38, respectively. The sample ratio 79.85:20.15 [awara:suya spice] had the highest score of 7.7 for overall acceptability.

Table 1b showed that awara-based kilishi with ratio 83.33:16.67, 79.85:20.15 and 75.62:24.38 scored 82.78, 85.56 and 80%, respectively acceptability index.

**Table 1a:** Sensory evaluation of awara-based kilishi analogue with suya spice samples

Sample	Appearance	Texture	Flavour	Taste	Overall acceptability
Awara:SS					
100:0	7.00 <sup>bc</sup> ±1.21	6.55 <sup>abc</sup> ±1.36	6.90 <sup>a</sup> ±0.97	6.85 <sup>a</sup> ±1.14	7.05 <sup>a</sup> ±0.89
71.43:28.57	6.50 <sup>c</sup> ±2.04	5.90 <sup>c</sup> ±2.25	4.60 <sup>b</sup> ±1.85	4.85 <sup>b</sup> ±2.21	5.45 <sup>c</sup> ±1.76
83.33:16.67	7.10 <sup>abc</sup> ±1.12	7.05 <sup>ab</sup> ±1.32	7.20 <sup>a</sup> ±1.15	7.25 <sup>a</sup> ±1.16	7.45 <sup>a</sup> ±1.19
88.24:11.76	6.70 <sup>c</sup> ±1.53	6.75 <sup>abc</sup> ±1.29	6.90 <sup>a</sup> ±1.17	6.60 <sup>a</sup> ±1.39	6.90 <sup>ab</sup> ±0.91
94.80:5.20	6.55 <sup>c</sup> ±1.40	6.45 <sup>bc</sup> ±1.79	5.40 <sup>b</sup> ±1.73	5.50 <sup>b</sup> ±1.61	6.10 <sup>bc</sup> ±1.48
91.68:8.32	7.60 <sup>ab</sup> ±0.94	7.60 <sup>a</sup> ±1.19	6.70 <sup>a</sup> ±1.69	6.80 <sup>a</sup> ±1.54	6.95 <sup>ab</sup> ±1.43
79.85:20.15	7.95 <sup>a</sup> ±0.83	7.50 <sup>ab</sup> ±1.15	7.05 <sup>a</sup> ±1.10	7.25 <sup>a</sup> ±1.02	7.70 <sup>a</sup> ±0.87
75.62:24.38	7.70 <sup>ab</sup> ±1.13	7.10 <sup>ab</sup> ±1.83	7.10 <sup>a</sup> ±1.45	6.80 <sup>a</sup> ±1.88	7.20 <sup>a</sup> ±1.74

Values are mean ± standard deviation of 30 panelists. Data in the same column bearing different superscripts differ significantly at  $p < 0.05$

Where SS= Suya spice

**Table 1b:** Acceptability index [%] of the overall acceptability of *awara*-based *kilishi* analogue with suya spice samples

Sample Awara:SS	Overall acceptability mean score	Acceptability index [%]	Interpretation
100:0	7.05	78.33	Moderately acceptable
71.43:28.57	5.45	60.56	Fairly acceptable
83.33:16.67	7.45	82.78	Highly acceptable
88.24:11.76	6.90	76.67	Moderately acceptable
94.80:5.20	6.10	67.78	Fairly acceptable
91.68:8.32	6.95	77.22	Moderately acceptable
79.85:20.15	7.70	85.56	Highly acceptable
75.62:24.38	7.20	80.00	Highly acceptable

Where SS= Suya spice

### Proximate composition of *awara*-based *kilishi* analogue with suya spice

In Table 2 is the Proximate composition of *awara*-based *kilishi* analogue with suya spice. The moisture, ash, fiber, fat, protein and carbohydrate content ranged from 5.2-76.78, 0.22-3.99, 0.18-0.63, 4.69-24.04, 7.92-37.12 and 10.19-38.88%, respectively. Generally, there was significant difference [ $p<0.05$ ] in all the proximate composition parameters. The moisture content of the wet curd drastically reduced from 76.78-5.08% while the ash, fiber, fat, protein and carbohydrate

increased from 0.22-3.99, 0.18-0.63, 4.69-24.04, 7.92-37.12 and 10.19-38.88%, respectively. The *awara*-based *kilishi* with-out suya-spice appeared to have a significantly [ $p<0.05$ ] higher fat [24.04%] and protein [37.12%] when compared with the ones with suya spice. The inclusion of the suya spice to the *awara*-based *kilishi* analogue led to the reduction of fat [24.04-22.92%], protein [37.12-35.04%] and moisture [5.89-5.61%] while the ash [3.79-3.99%], crude fiber [0.51-0.63%] and carbohydrate [28.64-38.88%] increased.

**Table 2:** Proximate composition [%] of *awara*-based *kilishi* analogue with suya spice

Sample ratio [A:S]	Moisture	Ash	Crude fiber	Fat	Crude protein	CHO
Fresh Curd	76.78 <sup>a</sup> ±0.02	0.22 <sup>d</sup> ±0.01	0.18 <sup>c</sup> ±0.00	4.69 <sup>e</sup> ±0.02	7.92 <sup>e</sup> ±0.01	10.19 <sup>e</sup> ±0.01
100:0	5.89 <sup>b</sup> ±0.01	3.79 <sup>c</sup> ±0.01	0.51 <sup>d</sup> ±0.01	24.04 <sup>a</sup> ±0.06	37.12 <sup>a</sup> ±0.02	28.64 <sup>d</sup> ±0.07
83.33:16.67	5.61 <sup>c</sup> ±0.02	3.82 <sup>b</sup> ±0.01	0.63 <sup>a</sup> ±0.01	22.92 <sup>b</sup> ±0.01	35.04 <sup>b</sup> ±0.05	31.97 <sup>c</sup> ±0.04
79.85:20.15	5.20 <sup>d</sup> ±0.01	3.84 <sup>b</sup> ±0.01	0.60 <sup>b</sup> ±0.00	21.10 <sup>c</sup> ±0.01	33.41 <sup>c</sup> ±0.01	35.83 <sup>b</sup> ±0.01
75.62:24.38	5.08 <sup>e</sup> ±0.01	3.99 <sup>a</sup> ±0.01	0.55 <sup>c</sup> ±0.01	20.39 <sup>d</sup> ±0.01	31.10 <sup>d</sup> ±0.01	38.88 <sup>a</sup> ±0.01

Values are mean scores ± standard deviation of triplicate determination. Data in the same column bearing different superscripts differ significantly at  $p<0.05$   
Key: A= Awara, S= spice and CHO= Carbohydrate

### Vitamin composition of *awara*-based *kilishi* analogue with suya spice

The vitamin composition of *awara*-based *kilishi* analogue with suya spice as shown in Table 3 showed that, the vitamin B<sub>12</sub>, B<sub>6</sub>, B<sub>1</sub> and C of the raw fresh *awara* significantly generally decreased with processing from 270-90, 1100-850, 500-310µg/100 and 3730-2290mg 100g<sup>-1</sup> when compared with the *awara*-based

*kilishi* without suya spice and further increased significantly [ $p<0.05$ ] from 90-230, 850-1020, 310-420µg/100 and 22.90-35.40mg 100g<sup>-1</sup>, respectively with the inclusion of the suya spice to the *awara*-based *kilishi*. The *awara*-based *kilishi* with suya spice with ratio 75.62:24.38 had the highest value of vitamin B<sub>12</sub> [230µg/100], B<sub>6</sub> [1020µg/100], B<sub>1</sub> [420µg/100] and C [35.40mg 100g<sup>-1</sup>].

**Table 3:** Vitamin composition of *awara*-based *kilishi* analogue with suya spice

Sample ratio [A:S]	Vit. B <sub>12</sub> (µg/100)	Vit. B <sub>6</sub> (µg/100)	Vit. B <sub>1</sub> (µg/100)	Vit. C (mg 100g <sup>-1</sup> )
Fresh Curd	270 <sup>a</sup> ±0.01	1100 <sup>a</sup> ±0.02	500 <sup>a</sup> ±0.02	37.30 <sup>a</sup> ±0.03
100:0	90 <sup>e</sup> ±0.01	850 <sup>d</sup> ±0.01	310 <sup>e</sup> ±0.02	22.90 <sup>e</sup> ±0.02
83.33:16.67	150 <sup>d</sup> ±0.02	850 <sup>d</sup> ±0.01	360 <sup>d</sup> ±0.04	31.10 <sup>d</sup> ±0.09
79.85:20.15	180 <sup>c</sup> ±0.01	960 <sup>c</sup> ±0.01	400 <sup>c</sup> ±0.01	33.50 <sup>c</sup> ±0.04
75.62:24.38	230 <sup>b</sup> ±0.01	1020 <sup>b</sup> ±0.01	420 <sup>b</sup> ±0.01	35.40 <sup>b</sup> ±0.04

Values are mean scores ± standard deviation of triplicate determination. Data in the same column bearing different superscripts differ significantly at  $p<0.05$   
Key: A= Awara and S= Suya spice

### Antinutrients content of *awara*-based *kilishi* analogue with suya spice

Table 4 shows the antinutrients content of *awara*-based *kilishi* analogue with suya spice. It was observed that the stachyose [0.88-0.57mg 100g<sup>-1</sup>], trypsin [2.14-1.03mg 100g<sup>-1</sup>], lectin [0.76-0.07mg 100g<sup>-1</sup>], phytate [2.26-2.16mg 100g<sup>-1</sup>], glycine [0.95-0.63mg 100g<sup>-1</sup>] and raffinose [0.31-0.15mg 100g<sup>-1</sup>] of the fresh wet curd significantly [ $p<0.05$ ] decreased with processing into *kilishi* analogue. The 100 percent *kilishi* analogue without suya spice was lowest in trypsin, lectin,

phytate, glycine and raffinose value compared to the *kilishi* analogue samples with suya spice which had an increased value. This increase in antinutrients with the inclusion of suya spices is an indication that the suya spice may contain some minute antinutrients. *Awara*-based *kilishi* with suya spice with formulation ratio of 83.33:16.67 had the lowest level of stachyose [0.54mg 100g<sup>-1</sup>], lectin [0.14mg 100g<sup>-1</sup>], glycine [0.22mg 100g<sup>-1</sup>] and raffinose [0.24mg 100g<sup>-1</sup>] while 75.62:24.38 had the lowest trypsin inhibitor [1.15mg 100g<sup>-1</sup>] and phytate [2.19mg 100g<sup>-1</sup>].

**Table 4:** Antinutrients and glycine content [mg 100g<sup>-1</sup>] of *awara*-based *kilishi* analogue with suya spice

Sample ratio [A:S]	Stachyose	Trypsin	Lectin	Phytate	Raffinose	Glycine
Fresh Curd	0.88 <sup>a</sup> ±0.01	2.14 <sup>a</sup> ±0.01	0.17 <sup>a</sup> ±0.03	2.26 <sup>a</sup> ±0.01	0.31 <sup>a</sup> ±0.01	0.95 <sup>c</sup> ±0.03
100:0	0.57 <sup>d</sup> ±0.01	1.03 <sup>b</sup> ±0.03	0.07 <sup>b</sup> ±0.01	2.16 <sup>ab</sup> ±0.14	0.15 <sup>c</sup> ±0.01	0.63 <sup>ab</sup> ±0.01
83.33:16.67	0.54 <sup>e</sup> ±0.01	1.19 <sup>ab</sup> ±0.04	0.14 <sup>ab</sup> ±0.01	2.25 <sup>a</sup> ±0.01	0.24 <sup>b</sup> ±0.01	0.22 <sup>b</sup> ±0.01
79.85:20.15	0.81 <sup>c</sup> ±0.01	1.17 <sup>ab</sup> ±0.03	0.19 <sup>ab</sup> ±0.01	2.19 <sup>ab</sup> ±0.01	0.27 <sup>ab</sup> ±0.01	0.73 <sup>ab</sup> ±0.01
75.62:24.38	0.83 <sup>b</sup> ±0.01	1.15 <sup>ab</sup> ±0.03	0.16 <sup>ab</sup> ±0.01	2.19 <sup>ab</sup> ±0.01	0.26 <sup>ab</sup> ±0.01	0.70 <sup>ab</sup> ±0.01

Values are mean scores ± standard deviation of triplicate determination. Data in the same column bearing different superscripts differ significantly at  $p < 0.05$ . Key: A= Awara and S= spice

## Discussion

### Sensory evaluation of *awara*-based *kilishi* analogue with suya spice

From Table 1a, it was observed that the sample ratio 71.43:28.57 for *awara*:suya spice was scored the lowest values in all the sensory parameters evaluated. This could be attributed to the fact that the ratio of the suya spice [28.57%] used was higher compared to other samples, which indicates poor acceptance for such level of suya spice in the formulated *kilishi* analogue. The no significant difference  $p > 0.05$  in the appearance, texture, flavour, taste and overall acceptability for all the *awara*-based *kilishi* with suya spice shows that these samples competed with each other. However, *awara*-based *kilishi* with suya spice with ratio 79.85:20.15 ranked the highest score of 7.7 for overall acceptability and this means 'liked moderately' on 9-point Hedonic scale. Notably, it was observed that the *kilishi* analogue with 100 percent *awara* [soy curd] was also well accepted by the sensory panelists and competed favorably with the other samples that had suya spices.

The sensory performance of the plant-based *kilishi* analogue varied with soy curd–suya spice ratio, reflecting the balance between structure, flavour intensity, and consumer familiarity. The highest appearance score of 7.95 was recorded by the *awara*-based *kilishi* with the formulation ratio of 79.85:20.15 and this may be due to surface colour enhancement from the suya spice pigments and darker colour due to the roasted peanut particles giving the traditional *kilishi*-like appearance. Consumers acceptance for traditional *kilishi* is critically driven by visual attributes such as colour, signaling authenticity and quality [26].

The formulation with ratio 91.68:8.32 rated highest score of 7.6 in texture, which could be due to higher soy curd content. Formation of cohesive gel networks with solid moisture-binding ability by soy proteins gives a firmer meat-like texture. The improvement in the chewiness and structural integrity of dried meat analogues is achieved by minimizing matrix disruption due to lower spice inclusion [2].

The *awara*-based *kilishi* with formulation ratio of 83.33:16.67 had the highest flavour score of 7.2 which shows best balance between soy curd and suya spice. The capsaicinoids, sulfur compound and gingerols are complex aromatic compounds contributed by the suya spice while the excessive pungency was moderated by the soy curd. The flavour perception can be dominated by too much spice while little spice decreases the sensory appeal due to weakened suya note characteristic [27].

*Awara*-based *kilishi* with formulation ratio of 79.85:20.15 and 83.33:16.67 scored 7.25, the highest in taste, showing the

synergy between the flavour and umami compounds from soy proteins and the nutty spicy component of the suya spice. The palatability enhancement through fat-mediated flavour release without too much heat is from peanut fraction [28].

The *awara*-based *kilishi* with formulation ratio of 79.85:20.15 scored 7.7 being the highest overall acceptability, showing that consumers favorite is subject to sensory agreement rather than the peak performance in a single attribute [26].

The observations from the study showed that inclusion of 16-20% suya spice is optimal for plant-based *kilishi* analogues. This indicate that consumer acceptance could be maximized without compromising the texture or causing flavour overload. Generally, on a 9-point hedonic scale, scores above 7 indicates strong acceptability which compares with traditional meat *kilishi* showing the possibility of plant-based *kilishi* as a cultural appropriate alternative [27]. From a product development perspective, formulations can be tailored toward texture-focused products using higher soy curd levels or flavour- and appearance-driven products with higher spice inclusion. Importantly, combining high sensory appeal with nutritional benefits such as increased fiber, lower fat, and plant protein enhances market potential among health-conscious and flexitarian consumers [2].

From Table 1b, all the *awara*-based *kilishi* with suya spice samples with >80% acceptability index means that the product is highly acceptable and were selected and subjected to proximate composition, vitamin and antinutrient content.

The overall acceptability of the *awara*-based *kilishi* with suya spice [ranked highly acceptable on acceptability index] were within the same value as reported by Iyiola et al. [29] for beef *kilishi* [7.5] and above the 6.75 score for plant-based meat substitute deep-fried product by Archana et al. [30] on 9-point Hedonic scale on overall acceptability.

### Proximate composition of *awara*-based *kilishi* analogue with suya spice

The significant [ $p < 0.05$ ] drastic reduction of moisture content of fresh raw *awara* from 76.78-5.08% showed the effect of cooking which would have led to evaporation of moisture during the oven baking. This low level of moisture content is an important factor in considering the shelf stability of the *kilishi* samples [31].

The increase in ash, fiber, fat, protein and carbohydrate of the *awara*-based *kilishi* analogue could be due to the oven baking leading to concentration effect while the highest moisture [5.89%], fat [24.04%], protein [37.12%] with lowest ash [3.79%], fiber [0.51%] and carbohydrate [28.64%] in the

*awara*-based *kilishi* with no suya spice could be linked to the inherent ability of the suya spice to increase those nutrient. The reduced moisture in the *kilishi* analogue with suya spice could be that the spices had some inherent ingredient with the ability to bind moisture, reduction in fat and protein content could be attributed the negative interaction of the suya spice with the *awara* in the presence of heat.

The reduction in moisture content would further enhance shelf stability and microbial safety, a critical quality attribute for dried meat analogues [32].

The reduction in fat content is nutritionally advantageous, as reduced-fat meat are associated with improved cardiovascular health and lower susceptibility to lipid oxidation during storage [2]. The reduction in protein content can be attributed to a dilution effect from suya spice inclusion; however, the substantial peanut fraction in suya spice provides complementary plant protein and healthy lipids, partially offsetting this reduction [33].

The increase in ash, fiber and carbohydrate of the *kilishi* analogue with suya spice could be due to the enhancement capability of the spices which contain some nutrient.

Increased ash content reflects enhanced mineral contribution from peanuts and spice ingredients such as chili pepper, ginger, garlic, and onion, which are recognized sources of essential micronutrients [28]. The observed increase in crude fiber and carbohydrate contents highlights the plant-based nature of suya spice, offering dietary fiber that supports digestive health, satiety, and glycemic regulation, nutritional benefits largely absent in conventional animal-based *kilishi* [34]. Iyiola et al. [29] and Iyiola et al. [35] reported 9.54, 43.69, 17.46, 2.23, 4.53%; and 11.14-13.89, 62.56-69.32, 6.29-8.68, 5.28-7.45% for moisture, protein, fat, crude fiber and ash content, respectively for beef *kilishi* made with groundnut slurry; while beef, chicken and pork *kilishi* is higher than the values obtained from this research expect for the fat content [20.39-24.04%]. The produced *awara kilishi* with suya spice were of higher values of protein, fat and ash except for crude fiber when compared with Aaliya et al. [36] and Hamid et al. [37], who reported 5.15-12.12% [protein], 11.43-18.21% [fat], 1.09-1.20% [crude fiber], 1.94-3.83% [ash]; and 20.67 and 14.25% [protein], 3.41 and 1.67% [crude fiber], 3.76 and 2.88% [ash] in plant-based meat analogue; and jackfruit-based meat analogues and commercial plant-based meat analogue, respectively. This shows that the protein value of the produced *awara*-based *kilishi* competed favourably higher than the commercial plant-based meat analogue. The high level of fat [20.39-24.04%] content in the *awara*-based *kilishi* is in agreement with the statement of Kyriakopoulou et al. [38] and Egbert and Boarders [39] who reported that relatively high fat content in plant-based meat analogue enhances sensory attributes such as juiciness, tenderness and flavour retention. Overall, suya spice incorporation shifts the *kilishi* analogue toward a fiber-rich, mineral-enhanced, lower-fat, and more shelf-stable product, supporting the development of healthier, functional, and more sustainable meat alternatives.

### Vitamin composition of *awara*-based *kilishi* analogue with suya spice

Vitamin composition of *awara*-based *kilishi* analogue with suya spice is shown in Table 3.

In Table 3, the significant decreased in the vitamin B<sub>12</sub>, B<sub>6</sub>, B<sub>1</sub> and C of the fresh *awara* when processed to *awara*-based *kilishi* with no suya spice indicates that the heating process negatively affected the vitamins. These vitamins are known to be heat-sensitive. The significant increase of these vitamins with the inclusion of suya spice into the *awara*-based *kilishi* indicates that the suya spice had some inherent appreciable level of these vitamins. It was also noted that the formulation ratio of 75.62:24.38 *awara*-based *kilishi* with suya spice had the highest values of these vitamins. This ratio of 24.38 [suya spice] was actually the highest ratio used and showed to be the best formulation.

Vitamin B<sub>12</sub> is naturally absent in unfortified plant foods; thus, its presence in *awara*-based *kilishi* analogue likely originates from fortification, microbial synthesis during fermentation, or analytical analogues rather than intrinsic plant sources. Cobalamin is sensitive to heat and light, and thermal drying during *kilishi* processing may reduce retention; however, low moisture and limited oxygen exposure can aid preservation. The soy protein-lipid matrix does not markedly inhibit absorption, but bioavailability depends on the chemical form of B<sub>12</sub> released during digestion. The reported value of 230µg/100 [75.62:24.38] far exceeds the adult Recommended Dietary Allowance [RDA] of 2.4 µg/day, suggesting either fortification (fermented multi-grain liquor used for *awara* coagulation) or analytical overestimation. This confirms that plant-based *kilishi* can meaningfully contribute to B<sub>12</sub> intake only when fortified or validated for true bioactive cobalamin forms [40, 41, 42].

Vitamin B<sub>6</sub> [Pyridoxine] is widely distributed in soybeans and peanuts. Although heating and drying may cause 10–30% losses, protein denaturation during processing improves bioaccessibility. The highest reported value [1020µg/100, 75.62:24.38] is below the adult RDA of 1.3 mg/day, indicating that *awara*-based *kilishi* could serve as a reliable dietary source of B<sub>6</sub> with good post-processing bioavailability. Soy-based foods often provide B<sub>6</sub> levels comparable to or higher than many animal products on a per-calorie basis [43, 42].

Vitamin B<sub>1</sub> [Thiamin] is heat-labile and sensitive to alkaline conditions; nevertheless, short drying times and mildly acidic conditions during *kilishi* processing may limit degradation. Soybeans and peanuts are natural thiamin sources, and matrix disruption enhances enzymatic accessibility. The reported value [420µg/100; 75.62:24.38] below the adult RDA of 1.1–1.2 mg/day, demonstrating that plant-based protein foods can contribute substantially to daily thiamin intake, often surpassing levels found in processed meats [44, 42].

Vitamin C [Ascorbic Acid] is highly sensitive to heat, oxygen, and prolonged drying, so substantial losses are expected during *kilishi* production. However, residual vitamin C from suya spice ingredients [e.g., chili pepper, garlic, onion] may persist, and low-moisture conditions limit oxidative degradation during storage. The highest reported value [35.40 mg/100 g; 75.62:24.38] below the RDA of 75 mg/day for women and 90

mg/day for men, indicating strong potential for antioxidant activity and enhanced non-heme iron absorption when present [45, 46].

The values obtained for vitamin B<sub>1</sub>, B<sub>6</sub>, B<sub>12</sub> and C in all the awara-based *kilishi* were lower than the values of 4.07 [vitamin B<sub>1</sub>] and 4.07mg 100g<sup>-1</sup> [vitamin B<sub>6</sub>] and lower to 7380 and 38400mg 100g<sup>-1</sup> for vitamin B<sub>12</sub> and C, respectively reported by Emmanuel et al. [47] for beef *Kilishi* [Jerky Meat].

The vitamin profile of awara-based *kilishi* indicates that suya spice contributes not only sensory enhancement but also indirect fortification, particularly for vitamins B<sub>6</sub>, B<sub>1</sub>, and C. The possible contribution of fermented multigrain water used in soy curd coagulation may further explain the presence of B-vitamins, especially B<sub>12</sub>, which is otherwise absent in plant foods. These findings suggest that awara-based *kilishi* functions as a nutrient-enhanced, plant-based snack, though validation of true bioactive B<sub>12</sub> forms remains essential for nutritional claims.

#### **Antinutrients and glycine content of awara-based *kilishi* analogue with suya spice**

Stachyose and raffinose [ $\alpha$ -galactooligosaccharides] are indigestible carbohydrates commonly found in soybeans, a legume. It possesses a challenge as humans lack the enzyme  $\alpha$ -galactosidase needed to hydrolyze them, so they are fermented by colonic microbiota, producing gas and discomfort. When soybean or other legumes are processed poorly, it led to its high intake which reduces consumer tolerance and acceptability of legume-based products [48, 49].

Trypsin inhibitors interfere with trypsin and chymotrypsin [proteolytic enzymes], reducing protein digestibility and amino acid availability. Chronic consumption of inadequately processed soy products can impair growth and protein utilization [50, 51].

Lectins are carbohydrate-binding proteins capable of binding to intestinal epithelial cells, thereby disrupting nutrient absorption and causing gastrointestinal irritation. Raw or under processed soy bean contain biologically active lectins that pose nutritional and toxicological concerns [52, 53].

Phytate chelates essential minerals such as iron, zinc, calcium, and magnesium, reducing their bioavailability. High-phytate diets are associated with mineral deficiencies, especially in populations relying heavily on plant-based staples [54, 55].

Glycine is a non-essential amino acid. However, in plant matrices, excessive free amino acids may participate in Maillard reactions during high-temperature processing, potentially affecting protein quality if uncontrolled. Proper drying and roasting conditions help manage this effect [56].

As seen in Table 4, the decreased in the value of stachyose, trypsin, lectin, phytate, glycine and raffinose of the fresh raw soy curd when processed to *kilishi* without suya spice inclusion and later increased slightly for most samples except few exceptions. This showed that the soaking, grinding, heating, and coagulation steps used in awara [soy curd] preparation significantly reduce antinutrients. Heat denatures trypsin inhibitors and lectins, while soaking and aqueous extraction lower raffinose and stachyose through leaching

[57]. The microbial  $\alpha$ -galactosidase activity from the fermented water or multigrain liquor used during coagulation, further hydrolyzes raffinose and stachyose and may have partially degraded phytate, while improving mineral bioavailability [58, 59]. The high-temperature [150°C, 45m] used during the drying and roasting steps in *kilishi* production cause extensive thermal inactivation of residual trypsin inhibitors and lectins, bringing them below biologically active levels. Controlled drying also limits excessive Maillard reactions involving amino acids such as glycine [60, 51]. Suya spice used in the production of awara-based *kilishi* contain organic acids and phenolic compounds that can enhance mineral solubility and counteract phytate–mineral binding. The spice may also stimulate endogenous digestive enzymes, indirectly improving nutrient absorption [54, 55].

The component of the suya Spices such as ginger and garlic possess carminative and digestive-enhancing properties, which may alleviate gastrointestinal discomfort associated with residual oligosaccharides like raffinose and stachyose [61].

The antimicrobial activity of the suya spice components such as garlic, ginger and chili reduce the microbial load thereby adding to the shelf-stability of the dried awara-based *kilishi*. In addition to the low moisture content in the *kilishi* bringing about microbiological stability [62, 63]. The combined effects of thermal processing, fermentation-assisted coagulation, drying, and suya spice inclusion substantially mitigate the negative effects of stachyose, raffinose, trypsin inhibitors, lectins, and phytate. As a result, improved protein digestibility, enhanced mineral bioavailability, reduced the risk of gastrointestinal discomfort, strengthened the microbial and shelf stability. These outcomes confirmed that awara-based *kilishi* is nutritionally safe, functionally improved, and suitable for regular consumption, supporting its development as a high-quality, culturally adapted plant-based meat analogue. However, the range value for phytate [2.16-2.25mg 100g<sup>-1</sup>], stachyose [0.54-0.83mg 100g<sup>-1</sup>], trypsin [1.03-1.19mg 100g<sup>-1</sup>], glycine [0.22-0.73mg 100g<sup>-1</sup>] and raffinose [0.15-0.27mg 100g<sup>-1</sup>] were within the acceptable values of below 1% to avoid significant mineral binding effects particularly iron absorption [64] and 2000-2600mg day<sup>-1</sup> [phytate], 0.5-3.0g day<sup>-1</sup> [0.2%] [glycine] in finished beverages, below 200-400 HAU [lectins], less than 0.5-3g day<sup>-1</sup> [stachyose], 0.67-2.56mg 100g<sup>-1</sup> [raffinose in soybean] and 1.2-3.8mg g<sup>-1</sup> [trypsin inhibitors] [65, 66, 67, 68, 69, 70, 71, 72, 73].

#### **Conclusion**

This study showed that awara-based *kilishi* with suya spice is a highly acceptable, nutritionally healthy, and environmentally sustainable alternative to conventional meat-based *kilishi*. The most preferred formulation [79.85:20.15 awara:suya] achieved high consumer acceptability, while compositional analysis discovered substantial levels of plant protein, dietary fiber, essential vitamins [including B-complex and vitamin C] and antinutrient contents within safe limits. This places awara-based *kilishi* flavored with suya spice among healthy food alternatives with the ability to give adequate nutrient without negative burdens related with traditional *kilishi* from animal source.

In addition to the nutritional benefits of this study, it is in line with the focus of the sustainable food systems and food sustainable connect agenda, by using a traditionally obtained soybean to produce plant-based kilishi thereby supporting food securing and reducing post-harvest losses and in turn reduces greenhouse gas emissions, land use and water demand relative to livestock-based kilishi. Furthermore, its scalability is anchored on its simple processing method involved, dependency on traditional preservation, local value addition with income chances, supporting robust and complete food systems.

Although this study is limited in its nutrient bioavailability validation, shelf-life study and market consumer testing, however, the findings showed that awara-based kilishi is rooted culturally as an inexpensive and climate-smart protein novelty. Notably, the findings of the study are in line with the sustainable food system principles, showing that traditional food analogue can promote human health, sustainable environment and socio-economic resilience, making awara-based kilishi a future sustainable diet and should be part of policy-supported nutrition intervention.

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### Figures originality

All figures used in the manuscript are original and not reproduced from any published articles

### List of abbreviation

HCl Hydrochloric acid  
RPM: Revolution per minute  
NaCl Sodium chloride  
ANOVA: Analysis of variance  
SPSS: Statistical package for the social science  
SS: Sua spice  
CHO: Carbohydrate  
A: Awara  
S: Spice  
RDA: Recommended dietary allowance

### Author contributions

Odoh, Eunice Ngozi: conceptualization, preliminary data collection and supervision of data collection, methodology verifications, writing of manuscript, corrections of the reviews.  
Ojiabo Chisom Chimdi: statistical data analysis, manuscript review  
Ezegbe Clement Chinedum: validation of methodology, manuscript review and corrections, literature review and updating.  
Onyenwenwa Kenekwaku Cynthia: laboratory data collection [proximate and sensory] and first stage result report writing, funding acquisition from families and friends.  
Orji Jennifer Oluebube: laboratory data collection [vitamin and anti-nutrient composition] and first stage result report writing, funding acquisition from families and friends.  
Igwe, Ernest Chukwusoro: review of the concept, methodology, collected data, manuscript writing and supervision.

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### Availability of data and materials

The data supporting the findings of this study are included within the article.

### Conflict of interest

The authors declare no conflicts of interest regarding this manuscript.

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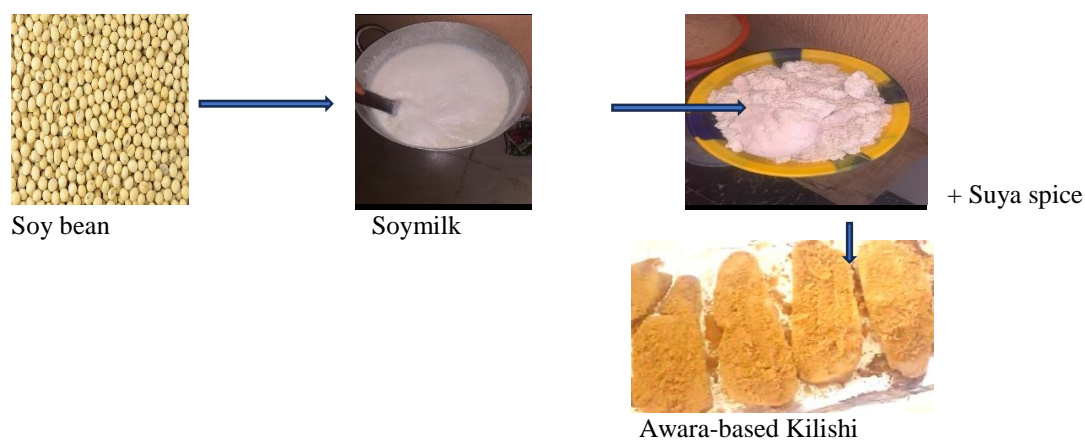
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Plates 1: Awara-based kilishi analogue with suya spice sample



Sensory Evaluation + Proximate + Vitamins + Antinutrient Analysis

**Result:** Sample ratio 83.33:16.67 had 82.7% Acceptability Index; protein, fiber & fat (35.04, 0.63 & 22.92%); 79.85:20.15 had 85.56% acceptability index while 75.62:24.38 had 80%; ash (3.99%), CH0 (38.88%), moisture (5.08%), vit. B<sub>12</sub>, 6, 1 & C (230, 1020, 420 & 3540mg100g<sup>-1</sup>). All antinutrient within acceptable limit.

**Conclusion:** Awara-based kilishi is a viable alternative to traditional animal-based kilishi when considering health & environment.