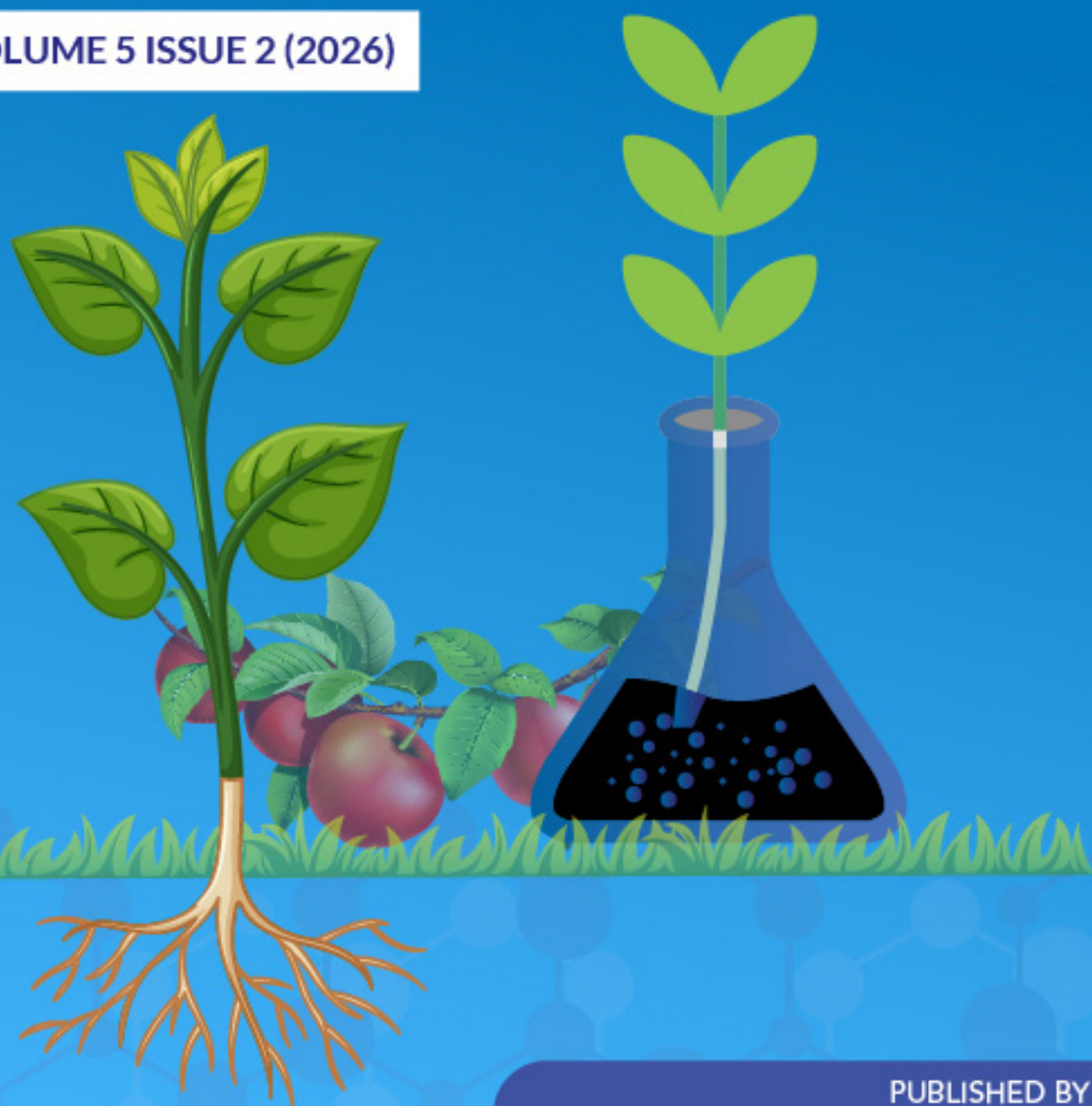




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## Sensory, Nutritional, Functional, Flavour Compounds and Microbial Properties of Novel Seasoning Cubes Produced from Tree Tomato Fruit and Crayfish as Influenced by Fermentation and Drying

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*Crayfish, Fermentation Drying, Flavour Compounds, Functional, Microbial Properties, Nutritional, Seasoning Cubes, Sensory, Tree Tomato*

### ABSTRACT

This study evaluated the effect of fermentation and drying on the core quality attributes of seasoning cubes produced from tree tomato and crayfish blends. Preliminary sensory evaluation was performed on five samples of salted and non-salted seasoning cubes containing varying percentages of tree tomato and crayfish. Preliminary results showed that the salted sample containing 60% tree tomato and 40% crayfish, and the non-salted sample containing 70% tree tomato and 30% crayfish, were most preferred. Portions of these two samples were further subjected to fermentation, drying and fermentation-drying. Sensory results showed that fermented-dried salted seasoning (FDSS) and fermented non-salted (FNSS) samples were the most preferred, with 8.30 and 6.80 acceptabilities, respectively. Proximate analysis revealed FDSS had the highest protein (26.38%) and energy content (394.23 kcal), significantly surpassing the commercial control sample (CCS) (6.98% protein, 354.64 kcal). Functional properties showed FDSS had high oil absorption (2.46 g/g) and bulk density (0.52 g/mL), while FNSS exhibited superior dispersibility (58.35%). GC-MS analysis detected up to thirty-two volatile flavour compounds in FDNS (fermented and dried non-salted seasoning), including terpenes, esters, aldehydes, and phenolics, compared to only two in CCS. Microbial analysis indicated acceptable bacterial loads ( $<10^4$  CFU/g) across all samples but elevated fungal counts (up to  $7.92 \times 10^3$  CFU/g in FNSS), exceeding the recommended limit ( $<10^3$  CFU/g). Salt significantly suppressed microbial growth and enhanced sensory attributes, while fermentation enriched flavour complexity and nutritional value. From the results, it could be concluded that the synergistic application of fermentation and drying significantly improved the nutritional profile, flavour diversity, and functional performance of tree tomato–crayfish seasoning cubes. FDSS emerged as the optimal formulation for the salted samples and FNSS for the non-salted samples, balancing high protein, palatability, and safety, supporting the development of clean-label, nutrient-dense alternatives to synthetic commercial seasonings, aligning with consumer demand for more natural and healthy products.

### INTRODUCTION

In the food sector today, simpler, more natural, less processed products with fewer and more identifiable components are becoming popular. This movement is referred to as the “clean label” concept (Asioli *et al.*, 2017). Consumers show a significant preference for clean-label products, often willing to pay a premium for foods with simpler and more natural ingredients, even if these products have less desirable sensory qualities (Maruyama *et al.*, 2021). “Clean label” prioritises transparency and simplicity in ingredient lists, avoiding artificial additives, preservatives, and chemicals (Cao & Miao, 2023). This movement reflects a shift towards healthier, more natural eating habits and a greater awareness of the impact of food choices on health and well-being. Clean labels on food packaging can positively influence consumer perception, with a preference for products that are perceived as natural and of higher quality (Sá *et al.*, 2023). This quest for clean-label products and consumer demand for natural ingredients and reduction in the use of synthetic additives has driven significant advancements in

food processing technology, particularly in the aspect of flavour enhancement, where consumers are increasingly requesting for natural rather than artificial flavour enhancers (Vasilakia *et al.*, 2022). Flavours are volatile compounds naturally released by foods or produced during various processing operations such as cooking and fermentation (Kane *et al.*, 2024).

Natural flavour enhancers are compounds or extracts obtained from plant, animal, or microbial sources that possess the ability to amplify, modify, or impart specific taste characteristics to food products without necessarily contributing a distinct flavour of their own. These substances interact with taste receptors and olfactory systems to modulate flavour perception, often by enhancing umami taste, masking off-notes, or increasing overall flavour intensity (Sarkar & Choudhury, 2017).

Several types of flavour enhancers are used in the food industry, and they come in various forms, such as spices, herbs, sauces, and seasonings (Davila, 2023). Seasonings are ingredients used to bring out the natural tastes and flavours present in food without a drastic alteration to the

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basic taste and flavour. Seasoning cubes are dehydrated seasonings commonly shaped into small cubes of approximately 4 g (Hong *et al.*, 2016). Seasonings are added to a food before it is ready for serving, either during its manufacture or in its preparation.

Commercial seasoning cubes may be made from single ingredients, such as individual spices and herbs, or they may be complex blends of components, such as salt, sugars, starch, vegetable fats, hydrolysed vegetable proteins, and flavour enhancers such as monosodium glutamate, monosodium guanylate, and disodium inosinate. Seasonings may also contain ingredients that have little or no effect on the flavour of a food but provide other advantages such as physical stability, antimicrobial preservation, or nutritional value (Moretti *et al.*, 2018).

Seasonings have been widely used in many different recipes and dishes across Africa. They possess significant nutritional and antioxidative properties (Śmiechowska *et al.*, 2021). They are equally used as vehicles for micronutrient fortification to address key dietary insufficiencies, particularly of iodine, iron, and provitamin A (De Mejia *et al.*, 2015). They are used to create unique and distinct flavours that are not naturally present in the food. This is especially important in the food industry, where creating unique flavours is crucial for attracting customers (Saguy *et al.*, 2018).

Sodium chloride, commonly called table salt, is the primary ingredient in seasoning cubes. Table salt has been reported to increase sodium levels in the blood, leading to high blood pressure and build-up of plaque on the arteries, raising the risk of stroke and heart diseases (Emurotu *et al.*, 2017). Seasoning cubes are also made up of hydrogenated oils, which, when excessively consumed, could lead to obesity and cancer among other health problems (Nwankwo & Omar, 2021).

Most food seasonings have also been shown to contain high quantities of synthetic flavour enhancers such as sodium glutamate, disodium inosinate, and disodium guanylate. Excess of these substances is associated with cardiovascular diseases and neurodegenerative disorders (Fadimatou *et al.*, 2024). Animal studies carried out by Wijayasekara & Wansapala (2021) demonstrated that monosodium glutamate (MSG) consumption precedes the development of obesity. Niaz *et al.* (2018) demonstrated that MSG induces a significant decrease in liver transaminase levels, indicating hepatic damage associated with chronic inflammation. Disodium inosinate, when broken down, releases uric acid in the body, which could promote gout in predisposed people (Ma *et al.*, 2024). The presence of these substances is often criticised by consumers for their effects on health, especially in developing countries where food standards based on risk analysis using consistent, reliable data are often weak or non-existent.

Plants and animal sources have long been recognised for their ability to produce a wide range of flavour compounds that enhance the taste and aroma of food. Certain vegetables like tomatoes and crustaceans such

as crayfish are rich in organic acids, which enhance the taste and flavour of food products, making them suitable natural sources to produce seasonings (Wijayasekara & Wansapala, 2021).

Tree tomato fruit (*Cyphomandra betacea*) is a tiny shrub or semi-woody tree that grows at elevations of 500–2500 m. The fruit is referred to as ‘tree tomato’ because its flesh resembles that of a tomato. It is a largely unexploited species, presenting a wonderful chance to diversify fruit production as a high-value income crop in many subtropical and mild temperate production locations (Shah *et al.*, 2022). Tree tomatoes can be consumed as juice, fresh fruit, cooked in stews, sauces, and desserts, or made into chutneys and pickles. In Cameroon, particularly in the North-West region, the tree tomato fruit is native. Historically, it was used as seasoning, especially by the Bambui people for the preparation of yellow soup for Achu, a traditional dish (Njoya *et al.*, 2020). However, the tree has now become scarce, and many farmers have stopped cultivating it, making it less common and marketable. It comes in two colours, red and yellow, with the red being the more popular and prevalent.

It contains anthocyanins and carotenoids, showing its biological, therapeutic, and preventative properties (Kong *et al.*, 2013). This fruit is found to have a relatively high content of provitamin A, C, B6, E and antioxidants (Valente *et al.*, 2011). Tree tomato fruits are a rich source of natural pigments with potential antioxidant activity, with phenolics being the main antioxidants (Osorio *et al.*, 2012). The seed of the fruit, together with flesh make up 1.0–1.5 % wet basis of the fruit (50–80 g). The proximate composition of Tree tomato seeds shows that it contains 22.63 % proteins, 21.13 % fat, and 3.15 % ash, with a total carbohydrate content of 43.87 %. Tree tomato seed oil's fatty acid profile revealed that linoleic acid (70.47 %), oleic acid (14.93 %), palmitic acid (9.41 %), stearic acid (2.23 %), and linolenic acid (1.73 %) are the most prevalent fatty acids (Morton, 2013).

Crayfish (*Procambarus clarkia*), a freshwater crustacean, is an important economic aquatic animal product that originated in America. Crayfish is enormously popular because of its delicious taste and high nutritional value. It contains high protein content and is rich in about eight essential amino acids (Li *et al.*, 2021). Crayfish also has a suitable fatty acid composition that contains nearly 50 % unsaturated fatty acids, including eicosatetraenoic acid (EPA) and docosahexaenoic acid (DHA) (Oksuz & Mazlum, 2016). Crayfish is a rich source of mineral elements and provitamins that are essential for human health, such as selenium and retinol (Peng *et al.*, 2021). It has a complex and appealing flavor profile attributed to several volatile flavor compounds such as aldehydes, ketones, and pyrazines, which contribute to its nutty, sweet, fruity, and salty meaty aromas, making it a suitable raw material to produce seasonings (Brian, 2022).

Fermentation and thermal processing, such as boiling and drying, are used traditionally to improve the sensory qualities, digestibility, and nutritional composition of

foods. Fermented foods constitute a substantial part of African diets. Common among the different categories of fermented foods in Africa are condiments used for seasoning food. These seasonings are usually in the form of paste, slurry, or solids. Traditionally, they are wrapped in leaves, making them less appealing and posing some safety concerns. The development of seasoning cubes from these condiments may improve the safety and attract more consumers to the product.

Tree tomato fruit and crayfish represent promising sources of natural flavour enhancers and essential nutrients yet are highly under-utilised in the aspect of seasoning cube formulations. Previous studies have focused on conventional processing methods, overlooking the potential impact of various pre-treatment techniques, such as fermentation and drying, on the development of flavour precursors and bioactive compounds in these natural ingredients. Also, the synergistic effects of combining tree tomato fruit and crayfish in varying proportions, along with the influence of different processing methods on their sensory attributes, shelf stability, and consumer acceptability, remain largely unexplored. This study aimed to evaluate the effect of fermentation and drying on the core quality attributes,

nutritional, functional, sensory, flavour compounds and microbial properties of seasoning cubes produced from tree tomatoes and crayfish.

## MATERIALS AND METHODS

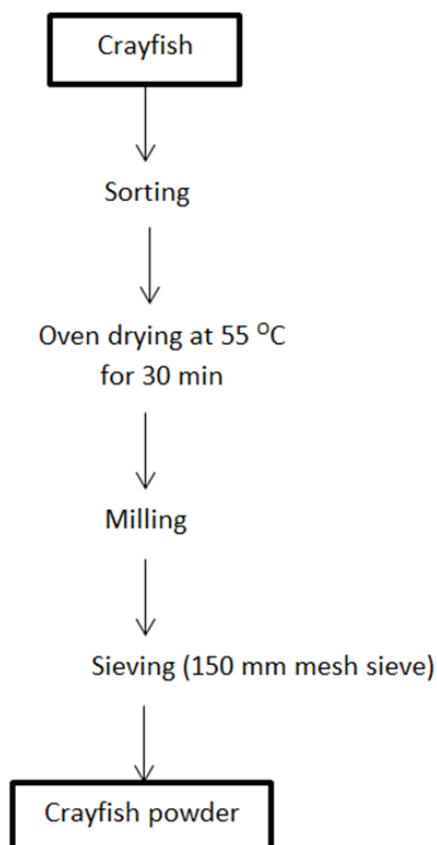
### RAW MATERIALS

Tree tomato fruit (*Cyphomandra betaceae*) was sourced from local farmers in Bamenda, Cameroon. Crayfish (*Procambarus clarkia*), sweet potato (*Ipomoea batatas*), ginger (*Zingiber officinale*), garlic (*Allium sativum*), salt, and hydrogenated fat were sourced from Wurukum market, Makurdi, Benue State, Nigeria. All these raw materials were transported to the Centre for Food Technology and Research (CEFTER) food laboratory, Benue State University, where they were further processed as ingredients for seasoning cubes production.

### Preparation of Raw Materials

#### Crayfish powder

A modified method described by Sengey *et al.* (2016) as shown in figure 1 was adopted to produce crayfish powder. The crayfish was sorted, oven dried at a temperature of 55 °C for 30 min, after which it was milled, sieved to obtain a fine powder, and packaged for subsequent use.



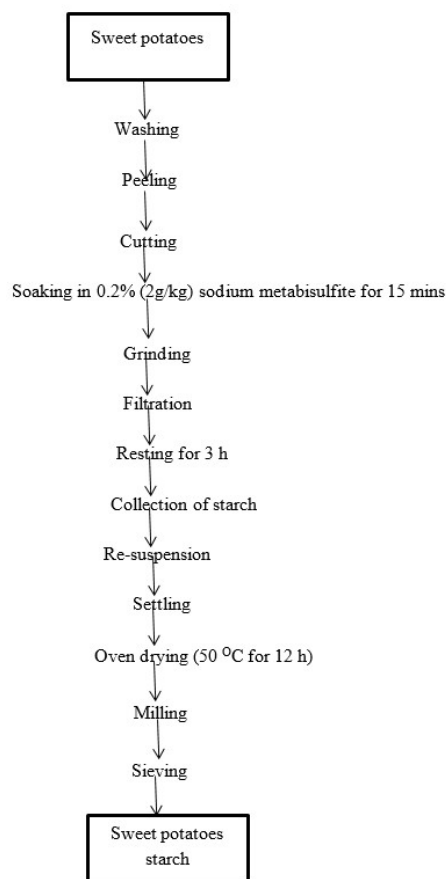
**Figure 1:** Flow chart for the production of crayfish powder

#### Sweet Potato Starch

Starch from sweet potatoes was produced following the method described by Kaur & Sandhu (2016). The processing steps were as shown in Figure 2.

#### Preparation of Fermented and Dried Tree Tomato Fruit Pulp

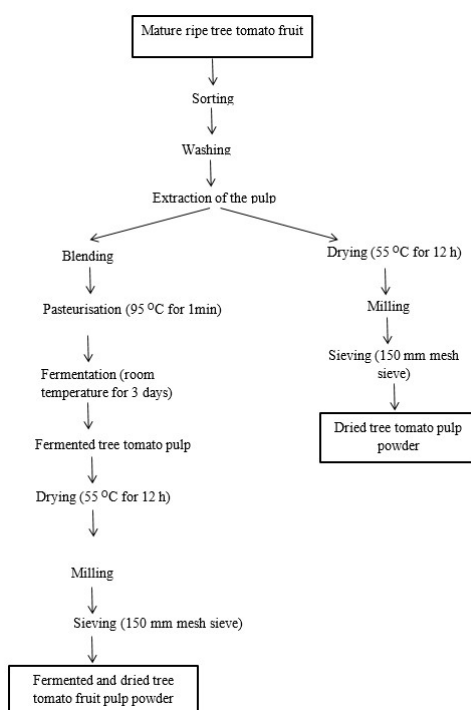
The method adopted from Olasupo & Okorie (2019) was used as shown in Figure 3



**Figure 2:** Flow chart for the production of sweet potato starch  
 Source: (Kaur & Sandhu, 2016)

### Production of Ginger Powder

Ginger powder was produced according to the method described by Ndife *et al.* (2022) with slight modifications, as shown in Figure 4 below. Ginger rhizomes were carefully sorted and washed with clean potable water. They were then peeled manually using stainless steel

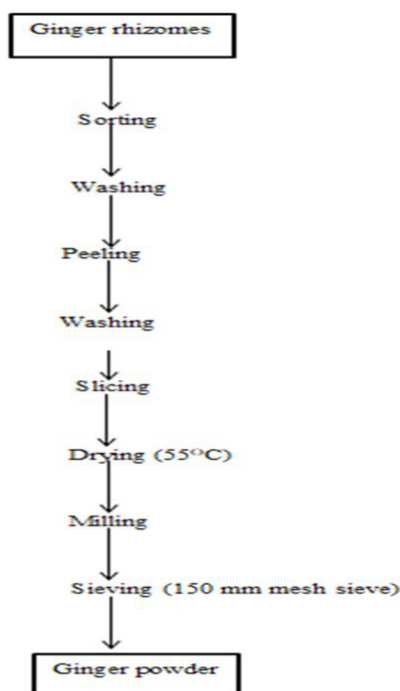


**Figure 3:** Flow chart for the production of fermented and dried tree tomato fruit pulp  
 Source: (Olasupo & Okorie, 2019)

knives, washed, and cut into smaller slices to facilitate drying. The sliced ginger was oven-dried at a temperature of 55 °C for 2 hours. After drying, it was cooled, milled, sieved, and packaged for subsequent use.

#### Production of Garlic Powder

Garlic powder was produced according to the method described by Mukhia *et al.* (2023), as shown in Figure 5 below, with some modifications.

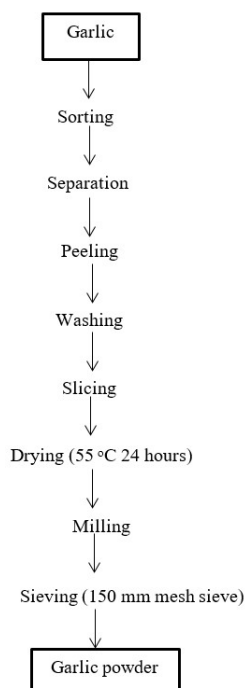


**Figure 4:** Flow diagram for the production of ginger powder source: Ndife *et al.*, (2022)

#### Experimental Design

The seasoning powder was prepared from fresh ingredients as depicted in Figure 4. The powder blends were then

used together with fresh untreated tree tomato fruit pulp to produce salted and non-salted seasoning cubes as described by Ndife *et al.* (2022) with modifications.



**Figure 5:** Flow chart for the production of garlic powder

Prepared seasoning cubes were then subjected to sensory analysis, where the two most preferred blends were selected. The two were used to formulate fermented,

dried and fermented-dried seasoning cubes. Formulated seasoning cubes were then subjected to quality evaluation using Star Maggi as the control sample.

### Production of Seasoning Cubes

A modification of the method described by Ndife *et al.* (2022) was used to produce the seasoning cubes. One tablespoonful of salt and sugar were dissolved in water

and vortexed for 60 seconds. The solution was placed in a kitchen blender, and proportions of the prepared tree tomato and crayfish were added and ingredients mixed at low speed for 10 minutes to ensure proper blending.

**Table 1:** Formulation of seasoning blends

| Sample | Tree tomato fruit% | Crayfish% |
|--------|--------------------|-----------|
| A      | 70                 | 30        |
| B      | 60                 | 40        |
| C      | 50                 | 50        |
| D      | 40                 | 60        |
| E      | 30                 | 70        |

**Table 2:** Recipe for seasoning cube production

| Sample | Salt % | Seasoning mix % | Sugar % | Ginger % | Garlic % | Potato starch % | Hydrogenated fat % | Water % |
|--------|--------|-----------------|---------|----------|----------|-----------------|--------------------|---------|
| S      | 15     | 40              | 10      | 5        | 5        | 15              | 10                 | 30      |
| NS     | 0      | 50              | 10      | 5        | 5        | 15              | 10                 | 30      |

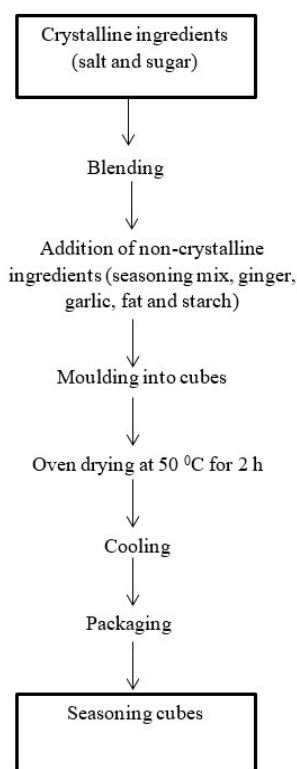
Where S= salted and NS= non-salted

Melted hydrogenated fat (10ml) was added to the mixture followed by 5 g potato starch and thoroughly mixed. The resultant paste was then placed in cubic moulds (5 cm<sup>3</sup>) and oven dried at 50 °C for 2 h. The cubes were then cooled and packaged in polythene Ziplock bags for subsequent quality evaluation. The process was as summarised in Figure 6.

Following the formulation in Table 4 and recipe on

table, 5 seasoning cubes were produced and subjected to sensory evaluation using Maggi Star as the control. The results obtained are shown in Table 3. The results showed that sample SB (60% tree tomato and 40% crayfish) for the salted seasoning and sample NSA (70% tree tomato and 30% crayfish) for the non-salted seasonings were most preferred and used for further treatment.

### Proximate Analyses



**Figure 6:** Flowchart for the production of seasoning cubes

Adopted from: (Ndife *et al.*, 2022)

The moisture, fat, protein, fibre, ash and carbohydrate content of tree tomato fruit pulp and crayfish powder, together with the seasoning blends was analysed following

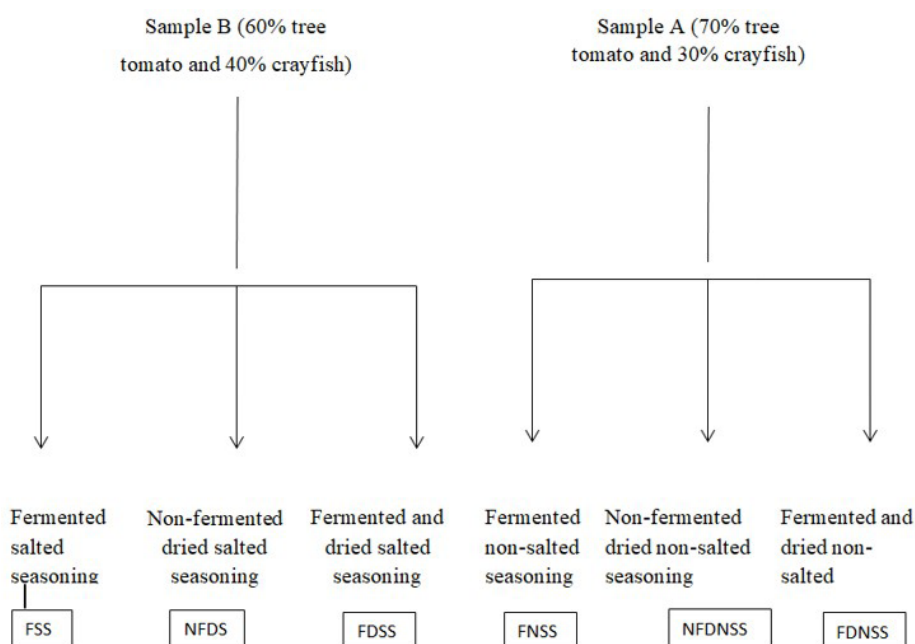
the standard Association of Analytical Chemists (AOAC, 2015) methods as described by Nkeudem *et al.* (2018).

**Table 3:** Preliminary Sensory Evaluation of Seasoning Cubes

| Sample | Appearance                | Aroma                      | Taste                       | Texture                  | Overall acceptability       |
|--------|---------------------------|----------------------------|-----------------------------|--------------------------|-----------------------------|
| SA     | 6.43 <sup>b</sup> ± 2.13  | 7.33 <sup>abc</sup> ± 1.60 | 7.10 <sup>abc</sup> ± 1.92  | 6.87 <sup>a</sup> ± 1.98 | 7.03 <sup>bcd</sup> ± 1.83  |
| SB     | 6.97 <sup>ab</sup> ± 1.94 | 7.70 <sup>bc</sup> ± 1.78  | 7.67 <sup>a</sup> ± 1.90    | 7.23 <sup>a</sup> ± 2.05 | 7.97 <sup>a</sup> ± 1.75    |
| SC     | 7.37 <sup>ab</sup> ± 1.47 | 7.87 <sup>a</sup> ± 1.38   | 7.63 <sup>a</sup> ± 1.63    | 7.63 <sup>a</sup> ± 1.63 | 7.73 <sup>ab</sup> ± 1.51   |
| SD     | 7.13 <sup>ab</sup> ± 1.55 | 7.40 <sup>abc</sup> ± 1.52 | 7.17 <sup>ab</sup> ± 1.60   | 7.43 <sup>a</sup> ± 1.10 | 7.17 <sup>abcd</sup> ± 1.51 |
| SE     | 7.37 <sup>ab</sup> ± 1.65 | 7.40 <sup>abc</sup> ± 1.69 | 7.27 <sup>ab</sup> ± 1.62   | 7.57 <sup>a</sup> ± 1.59 | 7.10 <sup>abcd</sup> ± 1.60 |
| NSA    | 7.27 <sup>ab</sup> ± 1.78 | 7.00 <sup>abc</sup> ± 1.72 | 6.73 <sup>abcd</sup> ± 1.74 | 6.97 <sup>a</sup> ± 1.85 | 7.43 <sup>abc</sup> ± 1.65  |
| NSB    | 7.10 <sup>ab</sup> ± 1.83 | 6.83 <sup>ab</sup> ± 1.58  | 6.50 <sup>bcd</sup> ± 1.66  | 7.27 <sup>a</sup> ± 1.53 | 6.47 <sup>d</sup> ± 1.59    |
| NSC    | 7.27 <sup>ab</sup> ± 1.74 | 6.67 <sup>c</sup> ± 1.56   | 6.13 <sup>cd</sup> ± 1.74   | 7.00 <sup>a</sup> ± 1.76 | 6.37 <sup>d</sup> ± 1.56    |
| NSD    | 7.70 <sup>a</sup> ± 1.39  | 6.83 <sup>ab</sup> ± 1.53  | 6.13 <sup>cd</sup> ± 1.57   | 7.60 <sup>a</sup> ± 1.30 | 6.47 <sup>d</sup> ± 1.46    |
| NSE    | 7.37 <sup>ab</sup> ± 1.56 | 6.70 <sup>b</sup> ± 1.32   | 6.03 <sup>d</sup> ± 1.71    | 7.43 <sup>a</sup> ± 1.45 | 6.77 <sup>cd</sup> ± 1.33   |
| CS     | 7.90 <sup>a</sup> ± 0.92  | 7.73 <sup>a</sup> ± 1.01   | 7.40 <sup>a</sup> ± 1.56    | 6.50 <sup>a</sup> ± 1.01 | 8.07 <sup>a</sup> ± 0.98    |

Key: A= 70% tree tomato and 30% crayfish, B= 60% tree tomato and 40% crayfish, C= 50% tree tomato and 50% crayfish, D= 40% tree tomato and 60% crayfish, E= 30% tree tomato and 70% crayfish.

S= Salted and NS= None-salted



**Figure 7:** Experimental designs for seasoning cube production

### Determination of Moisture Content

Moisture content was determined through the oven drying method. For each sample, two grams of a thoroughly mixed sample were accurately weighed into a pre-dried and clean crucible (initial weight  $W_1$ ). The crucible containing the sample was heated at 103.5°C for 4-5 hours until a constant weight was achieved. After heating, the crucible was allowed to cool in a desiccator for 3 min. Once cooled, the final weight ( $W_2$ ) was recorded and the percentage moisture calculated using the following formula:

$$\% \text{Moisture} = (W_1 - W_2) / (\text{Weight of sample}) \times 100 \quad (1)$$

Where;  $W_1$  = initial weight of crucible + sample,  $W_2$  = final weight of crucible + sample

### Determination of Crude Fat

This was done using a Soxhlet extraction device. Intermittent extraction was used to evaluate the crude fat contents. With this technique, 2 grams of a moisture-free sample were wrapped in a filter paper and placed in a fat-free thimble and inserted into the extraction tube. A pre-weighed and dried round-bottom flask was filled with petroleum ether and connected to the apparatus. The extraction process was allowed for six hours, after which

the ether was allowed to evaporate. After washing the extract with ether and transferring it to a clean glass dish, the ether was evaporated over a water bath. The dish was then placed in an oven at 105 °C for 30 min, removed, and cooled in a desiccator. The percentage of crude fat was further calculated using the following formula:

$$\frac{(\text{Weight of flask with sample} - \text{Weight of empty flask})}{(\text{Weight of sample})} \times 100 = \% \text{ Crude fat} \quad (2)$$

### Determination of Protein

The Kjeldahl method was used to determine the amount of protein in each sample. This technique involved filling a digestion flask with 0.5 g of dehydrated material and adding 10 mL of concentrated  $\text{H}_2\text{SO}_4$ , 2 g of  $\text{Na}_2\text{SO}_4$ , and 0.05 g of copper tuning. The components were properly mixed with a swirl, and the flask was heated to initiate digestion and to turn the combination into a clear, blue-green liquid. The digestion process was done for 120 min. After allowing the digest to cool down, it was transferred into a 100 mL volumetric flask, and distilled water was added until the desired total volume was reached. The digest then underwent distillation using the Markham Still Distillation apparatus described by Khalil and Manan in 1990. 10 mL of the digested sample was placed into the distillation tube, and 10 mL of 40 % sodium hydroxide (NaOH) solution was gradually introduced. Distillation then followed for 10 min, resulting in the collection of 75 mL of ammonia ( $\text{NH}_3$ ) as ammonium hydroxide ( $\text{NH}_4\text{OH}$ ) in a conical flask containing 20 mL of 2 % boric acid solution with a few drops of methyl red and bromocresol green indicators added. The  $\text{NH}_4\text{OH}$  caused a yellowish color to appear during the distillation. The distillate was titrated with a standard 0.01 M hydrochloric acid (HCl) solution until a pink color was observed. A blank sample was processed following the same procedure. The percentage of nitrogen (N) present in the original sample was calculated from the following formula:

$$\% \text{ N} = \frac{((S-B) \times M \times 0.014 \times D)}{(\text{Weight of the sample} \times V)} \times 100 \quad (3)$$

where;

S is the sample titration reading, B is the blank titration reading, M is the molarity of HCL, D is the dilution of the sample after digestion, V is the volume taken for distillation and 0.014 is the milliequivalent of Nitrogen (Nkeudem *et al.*, 2018).

The percentage crude protein is then obtained by multiplying the percentage Nitrogen obtained by the constant 6.25.

$$\% \text{ Crude Protein} = 6.25 \times \% \text{ N} \quad (4)$$

### Determination of Crude Fibre

The crude fibre content of the samples was determined using the AOAC (2015) methodology. Here, 2 grams (W1) of the material was weighed and transferred into a 1000 mL conical flask, then heated for 30 min with 200 mL of a 1.25 % sulfuric acid solution and filtered through a funnel. The residue was washed with boiling water until it

reached a neutral pH. Next, 200 mL of a 1.25 % sodium hydroxide solution was added, and the residue was boiled again for another 30 min. The residue was filtered three times using hot water and methylated spirit. After transferring the residue to a crucible, it was baked for an hour to dry completely. The crucible with its contents was cooled in a desiccator and weighed (W2). Subsequently, the residue was placed in a furnace and ashed for an hour at 600 °C, removed, cooled in a desiccator, and weighed once more (W3). The percentage of crude fibre was then calculated using the following formula:

$$\% \text{ fibre content} = \frac{(W_2 - W_3)}{W_1} \times 100 \quad (5)$$

where:

$W_1$  is the weight of the original sample,  $W_2$  is the weight of the crucible and residue, and  $W_3$  is the weight of the final ashed sample.

### Determination of Total Ash Contents

Following the method described by Nkeudem *et al.* (2018), an unused ceramic container was thoroughly cleansed, then subjected to intense heat at a temperature of 600 °C for 1 hour. It was allowed to cool down gradually in a moisture-free environment. Once cooled, the mass of the container was precisely measured and recorded as W1. 2g of each sample was placed in the crucible (W2). Using a blowpipe and a burner, the sample was lit and burned until it turned black. The crucible was heated to 600 °C for 3 hours in a muffle furnace. The organic material in the sample was fully burned, evidenced by a grey-white ash residue. The furnace used for ashing was subsequently switched off. After allowing the crucible to cool in a desiccator, its mass (W3) was measured. The proportion of ash was then quantified using the following calculation:

$$\% \text{ Ash} = \frac{(\text{Difference in Wt of Ash})}{(\text{Weight of sample})} \times 100 \quad (6)$$

### Determination of Carbohydrate Content

The percentage of carbohydrates in each sample was determined by subtracting the combined total of all other analysed nutrient components from 100 (representing the complete nutrient profile) (Nkeudem *et al.*, 2018). Mathematically,

$$\% \text{ Carbohydrate} = 100 - [\text{Protein}(\%) + \text{Fat}(\%) + \text{Ash}(\%) + \text{Fibre}(\%) + \text{moisture}(\%)] \quad (7)$$

### Energy Values

Energy values were determined from the fat, carbohydrate, and protein contents using Atwater's conversion factors (Nkeudem *et al.*, 2018).

$$\text{Energy (kcal/100g)} = 4 \times \text{Carbohydrate} + 4 \times \text{Protein} + 9 \times \text{Fat} \quad (8)$$

### Functional Properties

#### Oil Absorption Capacity

The oil absorption capacity of the seasoning samples was determined following the method described by Kefale *et al.* (2023). 1 g of powder sample was mixed with 10 mL

of oil in a centrifuge tube and allowed to stand at a room temperature of 30 °C for 1 hr. It was then centrifuged at 200 rpm for 30 min, and the supernatant was measured in a 10 ml measuring cylinder.

#### Water Absorption Capacity

To determine this, 10 mL of distilled water was added to 1 g of the sample in a weighed centrifuge tube. The tube was agitated on a vortex mixer for 2 min and then centrifuged at 4000 rpm for 20 min. The clear supernatant was then decanted and discarded, and the adhering drops of water were removed and weighed. Water absorption capacity was calculated using the following formula.

$$\text{WAC (ml/g)} = (\text{water absorbed}) / (\text{weight of sample}) \quad (9)$$

#### Bulk Density

It was determined according to the method described by Kefale *et al.* (2023). 50 g of powdered sample was added to a 100 ml measuring cylinder, and the volume measured and calculated using the following formula;

$$\text{Bulk density (g/ml)} = (\text{weight of powder}) / (\text{volume of powder}) \quad (10)$$

#### Dispersibility

Dispersibility of the seasoning powder was determined according to Kefale *et al.* (2023). This was measured by placing 10 g of the sample in a 100 mL stoppered measuring cylinder, and distilled water added to 100 mL. It was stirred vigorously and allowed to settle for 3 hrs. The volume of settled particles was then subtracted from 100 and the difference reported as percentage dispersibility following the formula;

$$\% \text{ Dispersibility} = 100 - \text{Volume of settled particles} \quad (11)$$

#### Determination of Flavour Compounds

This was done following the method described by Tuşa *et al.* (2012). In a bottle, 10 mL of seasoning extract and 20 µl of n-octan as surrogate standard (IS) with 1.5 mL of CH<sub>2</sub>Cl<sub>2</sub> were vigorously shaken for 5 min using a Vortex mixer to extract the free volatile fraction. The mixture was centrifuged at 4000 rpm for 5 min. The watery phase was removed, and the organic phase (CH<sub>2</sub>Cl<sub>2</sub> + volatile compounds) was adjusted from 1 ml to 0.1 ml in a dry flow of nitrogen and then injected (1 µl of extract) in a GC/MS system.

GC/MS analysis was then performed on a Thermo Finnigan Trace GC interfaced with a Polaris Q ion trap mass spectrometer, operated in electron impact ionization (EI) mode. Electron energy was set at 70 eV and ion source at 230 °C. Separations were carried out using a DB-5MS fused silica capillary column (30 m x 0.25 mm inner diameter, film thickness 0.5 µm). Helium was employed as the carrier gas with flow rate of 1.5 ml/min at constant flow. 1 µL of extract sample was injected in splitless mode at 250 °C using the following program: 60 °C (2 min), then 6 °C/min ramp to 240 °C (Tuşa *et al.*, 2012). The compound separation was obtained with a good resolution on full time chromatogram.

#### Microbial Analyses

##### Cultivation and Enumeration of Bacteria

The seasoning samples were evaluated for bacterial contamination through the serial dilution technique as described by (Akhigbemidu *et al.*, 2015). 1 g of each sample was transferred into bottles containing 9 ml of sterile distilled water. The mixture was thoroughly stirred for about 5 min. The mixture was further heated by boiling each of the samples at 100 °C for 10 min. The pour plate method was used to determine the microbial load in each sample. From the first dilution, 1 ml of the sample was pipetted into another sterile diluents containing 9 ml to obtain 10<sup>-2</sup> dilution. The samples were serially diluted up to 10<sup>-6</sup>. 0.1 ml of appropriate dilutions was then inoculated into sterile Petri dishes and molten nutrient agar added and left to solidify. The samples were inoculated triplicates and incubated at 37 °C for 24 hrs. The mean counts for triplicate cultures were recorded as the bacterial counts in the samples.

##### Cultivation and Enumeration of Fungi

Fungi (yeast and mould) were cultivated and enumerated using the pour and spread plate method on agar plates of potato dextrose. This was done following the method described by (Akhigbemidu *et al.*, 2015). 1 g of each sample was aseptically transferred into 9ml of sterile distilled water. This was stirred thoroughly for about 5 min and boiled at 100 °C for 10 min. From the first dilution, 1 ml of the sample was inoculated into another 9 ml. of the sterile diluents, representing the 10<sup>-2</sup> dilution. 0.1 ml of this dilution was then transferred into sterile Petri dishes and molten potato dextrose agar added and left to solidify. Each sample was made in triplicates and incubated at 28 °C for 48 h and colonies formed were counted. The mean counts for triplicate cultures were then recorded as the fungal counts in the samples.

#### Sensory Attributes Analyses

The appearance, aroma, taste, texture, and overall acceptability of the samples were evaluated according to the method described by Rodrigues *et al.* (2016). First, the seasoning cubes were used to prepare rice which will then be subjected to sensory analyses. 10 g of seasoning cubes, 3 ml of vegetable oil and 600 ml of water was used to prepare 100 g of rice for about 30 min. A commercial seasoning (maggi star) was used as a control sample. The samples were randomly coded and presented to 30 untrained panellists who are all consumers of maggi for the assessment. They were pre-instructed on how to complete the sensory evaluation form. Water was provided to the panellists to rinse their mouths after testing each sample. A 9-point hedonic scale was used to assess the appearance, aroma, taste, texture, and overall acceptability where 1 will represent dislike extremely and 9-like extremely.

#### Statistical Analysis

Statistical package for social science (SPSS) v26 computer

software was used to analyse raw data. Mean and standard deviation was calculated where appropriate. Analysis of variance (one-way ANOVA) used to determine treatment different from others in the various parameters to be tested. Duncan's Multiple Range Test was performed to determine the difference of mean. Differences was considered at 95 % ( $p < 0.05$ ) significant level (Ndife *et al.*, 2022).

## RESULTS AND DISCUSSION

### Results

#### Sensory Analysis of Seasoning Cube Samples

The results of sensory evaluation obtained are shown in table 4. The appearance of the seasoning showed that there was no significant difference between the formulated samples and the control sample (maggi star). Sample FSS scored highest with a mean score of  $7.80 \pm 1.11$  while sample DNSS scored the least with a mean score of  $7.00 \pm 1.21$ . From the 9-point hedonic scale, the results showed that the appearance of all samples were at least liked moderately

For aroma, a significant difference was recorded between the salted and non-salted seasonings. CSS had the highest score of  $7.60 \pm 1.05$ , followed by FDSS ( $7.55 \pm 1.47$ ), DSS ( $7.30 \pm 1.08$ ) and FSS ( $7.25 \pm 1.33$ ). CNS, FNSS, DNSS, and FDNSS had lower scores, suggesting that salt plays a crucial role in enhancing the aroma of the seasoning cubes.

The taste of the seasoning samples varied significantly from salted to non-salted and with treatments. FDSS received the highest score of ( $8.00 \pm 1.17$ ) followed by DSS ( $7.55 \pm 1.15$ ), CSS ( $7.50 \pm 0.95$ ), and FSS ( $7.15 \pm 1.18$ )

1.18). CNS, FNSS, DNSS, and FDNSS had lower scores of  $6.30 \pm 1.26$ ,  $6.45 \pm 1.50$ ,  $6.10 \pm 1.77$  and  $6.45 \pm 1.39$ , respectively

Results obtained for texture showed that there was no significant difference between the texture of the salted seasoning samples and the commercial control seasoning, with FDSS scoring highest with a mean value of  $7.50 \pm 1.36$ , and the least by DSS with a mean score of  $7.30 \pm 1.13$ . On the other hand, there was a significant difference between the non-salted samples and the commercial control, with CNS scoring highest with a mean value of  $6.90 \pm 0.79$  and FNSS and DNSS scoring  $6.35 \pm 1.14$ .

The overall acceptability of the seasoning samples showed a similar pattern to that of taste with a significant difference between samples. For the salted seasoning samples, FDSS was the most accepted sample with a score of  $8.30 \pm 0.86$  which was accepted due to the combined effect of fermentation, drying and salt. For the non-salted samples, FNSS was the most accepted with a score of  $6.80 \pm 1.06$ . This could be attributed to the production of new flavour compounds during fermentation.

From the overall acceptability of the seasoning samples, the 4 most preferred samples (CSS-7.60, FDSS-8.30, FNSS-6.80 and FDNSS-6.65) and the commercial control were then used for further analysis.

#### Proximate Composition of Tree Tomato, Crayfish and Seasoning Cube Samples

The results of proximate analyses are as shown in table 5. Tree tomato fruit recorded a high moisture content of 78.33 % which is typical of fresh fruits, significantly

**Table 4:** Sensory Evaluation Results of the Seasoning Cube Samples

| Samples | Appearance        | Aroma                | Taste                 | Texture           | Overall acceptability |
|---------|-------------------|----------------------|-----------------------|-------------------|-----------------------|
| CSS     | $7.55^a \pm 1.36$ | $7.60^a \pm 1.05$    | $7.50^{ab} \pm 0.95$  | $7.45^a \pm 0.69$ | $7.60^{ab} \pm 0.75$  |
| FSS     | $7.80^a \pm 1.11$ | $7.25^{ab} \pm 1.33$ | $7.15^{abc} \pm 1.18$ | $7.40^a \pm 0.99$ | $7.30^{bc} \pm 1.08$  |
| DSS     | $7.50^a \pm 0.76$ | $7.30^{ab} \pm 1.08$ | $7.55^{ab} \pm 1.15$  | $7.30^a \pm 1.13$ | $7.40^{bc} \pm 0.68$  |
| FDSS    | $7.65^a \pm 1.09$ | $7.55^a \pm 1.47$    | $8.00^a \pm 1.17$     | $7.50^a \pm 1.36$ | $8.30^a \pm 0.86$     |
| CNS     | $7.40^a \pm 0.94$ | $6.55^b \pm 0.89$    | $6.30^{cd} \pm 1.26$  | $6.90^b \pm 0.79$ | $6.50^d \pm 0.61$     |
| FNSS    | $7.15^a \pm 1.18$ | $6.55^b \pm 1.10$    | $6.45^{cd} \pm 1.50$  | $6.35^b \pm 1.14$ | $6.80^{cd} \pm 1.06$  |
| DNSS    | $7.00^a \pm 1.21$ | $6.55^b \pm 1.39$    | $6.10^d \pm 1.77$     | $6.35^b \pm 1.53$ | $6.10^d \pm 1.62$     |
| FDNSS   | $7.25^a \pm 1.25$ | $6.60^b \pm 1.35$    | $6.45^{cd} \pm 1.39$  | $6.40^b \pm 1.47$ | $6.65^{cd} \pm 1.42$  |
| CCS     | $7.35^a \pm 1.31$ | $6.95^{ab} \pm 1.57$ | $7.05^{bc} \pm 1.57$  | $7.65^a \pm 0.81$ | $7.35^{bc} \pm 1.53$  |

Key: CSS=Control salted seasoning, FSS= Fermented salted seasoning, DSS= Dried salted seasoning, FDSS= Fermented and dried salted seasoning, CNS= Control non-salted seasoning, FNSS= Fermented non-salted seasoning, DNSS= Dried non-salted seasoning, FDNSS= Fermented and dried non-salted seasoning and CCS= Commercial control seasoning

higher than that of crayfish powder which was quite low with a value of 5.36 %. The results showed a significant difference between the moisture content of the seasoning samples. The highest moisture content, 9.65 % was recorded in FDNSS while the lowest 6.75 % was recorded in sample CSS. The results of protein content showed that crayfish have a high protein content of 17.22 %

which is characteristic of sea foods. The protein content of tree tomato was 2.15 %. For the seasoning samples, the fermented and dried salted seasoning (FDSS) showed the highest protein content at 26.38 %, which could be because of the combined effects of fermentation and drying while the commercial control seasoning (CCS) had the lowest protein content at 6.98 %. The results

showed a significant difference in the protein content of the seasoning samples. For the fibre content, the results showed that Tree Tomato (TT) had a fibre content of 1.66 %, while Crayfish (CF) had a low fibre content of 1.48 % as expected for an animal product. The fibre content between the seasoning samples ranged from 1.40 % in sample FDNSS to 2.14 % CCS. No significant difference in fibre content was observed among the seasoning samples. The fat content of tree tomato was recorded as 0.53 % consistent with its fruit composition while that of crayfish was 1.78 %. There was some significant difference in the fat content of the seasoning cubes samples ranging from 4.68 % in sample CCS to 11.03 % in sample FDSS. The fermented and dried salted seasoning (FDSS) had the highest fat content, while the commercial control seasoning (CCS) had the lowest. Results of ash content showed that crayfish is high in minerals as shown by its high ash content of 7.35 %. It also revealed that tree tomato is not very rich in minerals as reflected in the value 1.41 % recorded. For the seasoning samples, no

significant difference was recorded among them. The ash content ranged from 5.78 % in sample FNSS to 6.63 % in CSS showing that crayfish contributed consistently to the mineral profile across all formulations.

The results of the proximate analysis also showed that tree tomato has a carbohydrate content of 15.92 % typical for fruits and crayfish a carbohydrate content of 66.82 %. Among the seasoning cubes, the carbohydrate content ranged from 46.76 % in FDSS to 71.15 % in CCS showing a significant difference between the samples.

A significant difference was observed in the energy content of tree tomato, crayfish and between the seasoning cube samples and the control sample. The results showed that tree tomato has an energy content of 77.19 Kcal and crayfish, 318.81 Kcal. For the seasoning cube samples, the energy content ranged from 354.64 Kcal in CCS to 394.23 Kcal in FDSS. The fermented and dried salted seasoning (FDSS) had the highest energy content, attributed to its balanced macronutrient composition.

**Table 5:** Proximate Composition of Tree Tomato, Crayfish and Seasoning Cube Samples

| SAMPLES | Moisture %               | Protein %                | Fibre %                 | Fat %                    | Ash %                   | CHO %                     | Energy KJ/Cal               |
|---------|--------------------------|--------------------------|-------------------------|--------------------------|-------------------------|---------------------------|-----------------------------|
| TT      | 78.33 <sup>d</sup> ±2.91 | 2.15 <sup>a</sup> ±0.66  | 1.66 <sup>a</sup> ±0.66 | 0.53 <sup>a</sup> ±0.01  | 1.41 <sup>a</sup> ±0.66 | 15.92 <sup>a</sup> ±2.24  | 77.19 <sup>a</sup> ±11.51   |
| CF      | 5.36 <sup>a</sup> ±1.65  | 17.22 <sup>d</sup> ±0.66 | 1.48 <sup>a</sup> ±0.66 | 1.78 <sup>ab</sup> ±0.66 | 7.35 <sup>b</sup> ±1.65 | 66.82 <sup>c</sup> ±0.66  | 318.81 <sup>b</sup> ±52.67  |
| CSS     | 6.75 <sup>a</sup> ±1.17  | 17.93 <sup>d</sup> ±1.65 | 1.50 <sup>a</sup> ±1.16 | 10.68 <sup>c</sup> ±2.33 | 6.63 <sup>b</sup> ±1.65 | 56.51 <sup>c</sup> ±2.34  | 393.88 <sup>c</sup> ±18.21  |
| FDSS    | 8.15 <sup>b</sup> ±1.16  | 26.38 <sup>f</sup> ±2.33 | 1.42 <sup>a</sup> ±1.16 | 11.03 <sup>c</sup> ±2.33 | 6.26 <sup>b</sup> ±1.65 | 46.76 <sup>b</sup> ±0.69  | 394.23 <sup>c</sup> ±24.02  |
| FNSS    | 8.42 <sup>b</sup> ±2.33  | 13.45 <sup>e</sup> ±1.16 | 1.58 <sup>a</sup> ±1.16 | 10.75 <sup>c</sup> ±2.33 | 5.78 <sup>b</sup> ±1.65 | 60.02 <sup>d</sup> ±3.01  | 390.63 <sup>c</sup> ±4.29   |
| FDNSS   | 9.65 <sup>c</sup> ±2.33  | 23.57 <sup>e</sup> ±1.16 | 1.40 <sup>a</sup> ±1.16 | 10.89 <sup>c</sup> ±2.33 | 6.02 <sup>b</sup> ±1.16 | 48.50 <sup>bc</sup> ±5.82 | 386.29 <sup>bc</sup> ±53.88 |
| CCS     | 8.95 <sup>b</sup> ±1.65  | 6.98 <sup>b</sup> ±1.16  | 2.14 <sup>a</sup> ±1.16 | 4.68 <sup>b</sup> ±2.33  | 6.10 <sup>b</sup> ±1.16 | 71.15 <sup>c</sup> ±2.82  | 354.64 <sup>bc</sup> ±14.33 |

Values are means ± standard deviations of triplicate determinations. Mean values in the row followed by different superscripts are significantly different ( $p < 0.05$ ).

Key: TT= Tree Tomato, CF= Crayfish, CSS=Control salted seasoning, FDSS= Fermented and dried salted seasoning, FNSS= Fermented non-salted seasoning, FDNSS= Fermented and dried non-salted seasoning and CCS= Commercial control seasoning

### Functional Properties of the Seasoning Cubes

The functional properties of the seasoning cubes samples such as bulk density, water absorption capacity, oil absorption capacity and dispersibility were evaluated. The results gotten were compared with that of a commercial seasoning (maggi star) and the outcome was as shown in table 6 below. The bulk density of the seasoning samples showed that sample FDSS and CCS had the highest bulk density 0.52 g/ml with no significant difference between them implying that they have a denser structure. The lowest bulk 0.41 g/ml density was recorded in sample FNSS indicating a less dense structure. A significant difference was recorded between the other samples and sample FDSS and CCS. For the water absorption capacity, there was no significant difference recorded between all

samples. The water absorption capacity ranged from 2.96 g/g for FDNSS which is the highest followed by 2.84 g/g for FDSS, 2.82 g/g for CCS, 2.69 g/g for CSS and 2.50 g/g for FNSS which is the lowest. Results obtained for oil absorption capacity showed that FDSS has the highest oil absorption capacity (2.46 g/g), followed closely by CSS (2.37 g/g) and FDNSS (2.28 g/g). The FNSS sample had the lowest oil absorption capacity (1.98 g/g), which could be attributed to the lack of salt and potential changes in lipid interactions. No significant difference was recorded between the formulated seasoning samples and the commercial control seasoning sample. The percentage dispersibility of the seasoning samples was equally evaluated and the results revealed a significant difference between the samples and the commercial control. The

**Table 6:** Functional Properties of the Seasoning Cubes

| Samples | Bulk density g/ml       | Water absorption capacity g/g | Oil absorption capacity g/g | Dispersibility %         |
|---------|-------------------------|-------------------------------|-----------------------------|--------------------------|
| CSS     | 0.49 <sup>c</sup> ±0.00 | 2.69 <sup>a</sup> ±1.65       | 2.37 <sup>a</sup> ±2.33     | 51.62 <sup>a</sup> ±1.65 |
| FDSS    | 0.52 <sup>d</sup> ±0.01 | 2.84 <sup>a</sup> ±1.65       | 2.46 <sup>a</sup> ±1.16     | 54.78 <sup>b</sup> ±2.33 |

|       |                          |                          |                          |                            |
|-------|--------------------------|--------------------------|--------------------------|----------------------------|
| FNSS  | 0.41 <sup>a</sup> ± 0.01 | 2.50 <sup>a</sup> ± 1.22 | 1.98 <sup>a</sup> ± 1.87 | 58.35 <sup>c</sup> ± 1.16  |
| FDNSS | 0.46 <sup>b</sup> ± 0.01 | 2.96 <sup>a</sup> ± 1.65 | 2.28 <sup>a</sup> ± 1.65 | 57.04 <sup>bc</sup> ± 1.16 |
| CCS   | 0.52 <sup>d</sup> ± 0.01 | 2.82 <sup>a</sup> ± 1.65 | 2.05 <sup>a</sup> ± 1.16 | 49.59 <sup>a</sup> ± 0.83  |

Values are means ± standard deviations of triplicate determinations. Mean values in the row followed by different superscripts are significantly different ( $p < 0.05$ ).

CCS=Control salted seasoning, FDSS= Fermented and dried salted seasoning, FNSS= Fermented non-salted seasoning, FDNSS= Fermented and dried non-salted seasoning and CCS= Commercial control seasoning

FNSS exhibited the highest percentage dispersibility of 58.35 % implying better solubility while the lowest percentage dispersibility (49.59 %) was seen in CCS. The results showed that the formulated seasoning samples had better solubility than the commercial control.

### Flavour Compounds of the Seasoning Cubes

The different flavour compounds present in the seasoning samples were evaluated using the gas chromatography-mass spectrometry (GC-MS) method. The GC-MS analysis of the seven samples revealed significant differences in volatile flavour compound profiles. Different categories of flavour compounds were found present in the seasoning samples as shown in table 7.

The results revealed the presence of several volatile flavour compounds such as aldehydes.

Terpenes, terpenoids, esters, alcohols, ketones, and phenolic compounds in the seasoning cubes as influenced by fermentation, drying and the composition of the various samples. The FDNSS sample recording the highest with 32 flavour compounds including aldehydes (4), terpenes (13), terpenoids (3), esters (5), alcohols (1), ketones (4), and phenolic compounds (1). This was followed by the FNSS with 30 compounds, and the least of this category of compounds shown by the CCS sample with only 2 categories of compounds identified one terpenoid and one alcohol. The salted samples CSS and FDSS recorded 4 and 7 volatile flavour compounds, respectively.

**Table 7:** Flavour compounds determined in the seasoning cube samples

| Sample | Category of flavour compounds       | Number of compounds |
|--------|-------------------------------------|---------------------|
| CSS    | Aldehydes/Ketones                   | 3                   |
|        | Esters                              | 1                   |
| FDSS   | Aldehydes/Ketones (derivatives)     | 3                   |
|        | Terpenes/Terpenoids (sesquiterpene) | 1                   |
|        | Aldehydes/Ketones                   | 2                   |
|        | Phenolic compounds (derivative)     | 1                   |
|        |                                     |                     |
| FNSS   | Aldehydes                           | 2                   |
|        | Terpenes                            | 11                  |
|        | Terpenoids                          | 7                   |
|        | Esters                              | 4                   |
|        | Alcohols                            | 1                   |
|        | Ketones                             | 3                   |
|        | Phenolic compounds                  | 1                   |
|        | Sulphur compounds                   | 1                   |
|        |                                     |                     |
| FDNSS  | Aldehydes                           | 4                   |
|        | Terpenes                            | 13                  |
|        | Terpenoids                          | 3                   |
|        | Esters                              | 5                   |
|        | Alcohols                            | 1                   |
|        | Ketones                             | 4                   |
|        | Phenolic compounds                  | 1                   |
| CCS    | Terpenoids                          | 1                   |
|        | Alcohol                             | 1                   |

CCS=Control salted seasoning, FDSS= Fermented and dried salted seasoning, FNSS= Fermented non-salted seasoning, FDNSS= Fermented and dried non-salted seasoning and CCS= Commercial control seasoning

### Effect of Treatments on the Microbial Load of the Seasoning Cubes

The microbiological quality of the formulated seasoning cubes from tree tomato fruit and crayfish together with the commercial control seasoning was evaluated based on the total bacteria count and total fungi count expressed in colony forming units per gram. For total bacteria count, the results obtained showed that sample CSS scored the lowest count  $4.24 \times 10^3$  cfu/g while the highest count  $8.78 \times 10^3$  cfu/g was recorded in the FNSS seasoning. Statistical analysis showed a significant difference ( $p < 0.05$ ) between the formulated seasoning samples and the commercial control seasoning. Between treatments, the fermented and dried salted seasoning recorded a

total bacteria count of  $6.76 \times 10^3$ , the fermented and dried non-salted recorded  $7.62 \times 10^3$  cfu/g while the commercial control seasoning recorded a total bacteria count of  $7.63 \times 10^3$  cfu/g.

The results for total fungal count showed that values ranged from  $4.16 \times 10^3$  cfu/g in CCS to  $7.92 \times 10^3$  cfu/g in FNSS. The lowest fungal count was observed in CCS, while FNSS recorded the highest. The results showed a significant difference ( $p < 0.05$ ) between the samples: CCS ( $4.16 \times 10^3$ ) < CSS ( $4.86 \times 10^3$ ) < FDSS ( $6.84 \times 10^3$ ) < FDNSS ( $7.03 \times 10^3$ ) < FNSS ( $7.92 \times 10^3$ ). This indicates a progressive increase in fungal load with the absence of salt and the application of fermentation and drying.

**Table 8:** Effect of treatment on the microbial load of the seasoning cubes

| Sample | Total bacteria count (cfu/g)  | Total fungi count (cfu/g)     |
|--------|-------------------------------|-------------------------------|
| CSS    | $4.24^a \times 10^3 \pm 0.02$ | $4.86^b \times 10^3 \pm 0.03$ |
| FDSS   | $6.76^b \times 10^3 \pm 0.03$ | $6.84^c \times 10^3 \pm 0.02$ |
| FNSS   | $8.78^d \times 10^3 \pm 0.03$ | $7.92^e \times 10^3 \pm 0.02$ |
| FDNSS  | $7.62^c \times 10^3 \pm 0.02$ | $7.03^d \times 10^3 \pm 0.02$ |
| CCS    | $7.63^c \times 10^3 \pm 0.12$ | $4.16^a \times 10^3 \pm 0.01$ |

Values are means  $\pm$  standard deviations of triplicate determinations. Mean values in the row followed by different superscript are significantly different ( $p < 0.05$ ).

Key: CSS=Control salted seasoning, FDSS= Fermented and dried salted seasoning, FNSS= Fermented non-salted seasoning, FDNSS= Fermented and dried non-salted seasoning and CCS= Commercial control seasoning

### Discussion

#### Sensory Evaluation of the Seasoning Cube Samples

The sensory evaluation data provided valuable insights into the quality attributes of seasoning cubes produced from tree tomato fruit and crayfish. Five (5) sensory parameters: appearance, aroma, taste, texture, and overall acceptability were evaluated.

The results indicated that the addition of salt in combination with fermentation and drying significantly improved multiple sensory attributes of the seasoning cubes' quality. They enhanced both appearance and aroma which is critical factors influencing consumer perception. The CSS sample recorded the highest score for aroma because of the natural aroma tree tomato and crayfish. Also, fermentation, drying and salting significantly enhanced the taste and overall acceptability of the seasoning cubes. These findings align with previous studies of Smith *et al.* (2020), who produced seasoning cubes from citrus extracts and reported that salt enhanced both taste and aroma but did not significantly affect texture.

On the other hand, the absence of salt resulted in lower scores across all sensory parameters, highlighting the potential drawbacks of using unsalted seasoning cubes. This could be due to the absence of salt's ability to enhance flavor and texture, leading to less appealing and less palatable products. These results align with those of Johnson & Lee (2021), who evaluated a seafood-based seasoning cube and noted that the absence of salt led to a stronger emphasis on natural flavors but reduced overall palatability.

However, the acceptance of both salted and non-salted samples, especially sample FNSS and FDNSS, shows that fermentation plays a key role in enhancing the sensory attributes of the seasoning cube. This could be explained by the fact that during fermentation, there is the breakdown of cell walls releasing new flavor compounds. Chemical reactions also take place such as the conversion of sugars into organic acids which further enhance the sensory attributes of the seasoning cubes.

Conclusively, the inclusion of salt in the production process of seasoning cubes derived from tree tomato fruit and crayfish is essential for achieving optimal sensory qualities. Also, the synergistic effect of fermentation which produces new flavor compounds and drying which concentrates the flavor compounds significantly improved the sensory attributes of both salted and non-salted seasoning samples thereby addressing the needs of both hypertensive and non-hypertensive persons.

#### Proximate Composition of Tree Tomato, Crayfish and Seasoning Cube Samples

The proximate composition analysis gives information about the nutritional quality of Tree Tomato, Crayfish, and the various seasoning cube samples produced. The results showed that the treatments applied (fermentation and drying) significantly influenced the chemical composition, which in turn affected the nutritional, functional, and sensory properties of the seasoning cubes.

The low moisture content below 10 % recorded in the

seasoning cube samples showed the effectiveness of the drying method employed. This low moisture content is important for ensuring stability and microbial safety of the product. The moisture content of this study was slightly lower than the 7.42 % to 12.62 % reported by Ndife *et al.* (2020). The low moisture content below 10 % reported in this work also conforms to that of Oladiran *et al.* (2021), for dried food products implying their stability during storage.

Results of the protein content showed that fermentation and drying significantly enhanced the protein content of the seasoning cubes. The fermented and dried salted seasoning (FDSS) exhibited the highest protein content (26.38 %). This could be attributed to the synergistic effect of fermentation which improves protein digestibility and bioavailability through the process of hydrolysis that breaks down proteins into peptides and amino acids making them more available, drying which concentrates nutrients during processing and the high protein content of crayfish. This makes FDSS a nutritionally superior option compared to the commercial control seasoning (CCS), which had the lowest protein content (6.98 %). The untreated salted seasoning sample CSS had a higher protein content than FDNSS because of a higher percentage of crayfish in the sample. The results of this study are higher than that reported by Olaide *et al.* (2020), who recorded a protein content of 3.49 % to 9.93 % for bouillon cubes produced from fermented condiments of *Ricinus communis* L. seeds.

The fibre content in the formulated seasoning cubes was lower than in the commercial control seasoning though not significantly, ranging from 1.40 % to 2.14 %. This is because of the low fibre contribution from crayfish and tree tomato and the limited presence of fibrous components in the seasoning formulation. The low fibre content of the seasoning samples in this work aligns with those of Singh *et al.* (2022), who reviewed the fibre content in seasonings. This is still recommended as seasonings are produced to be concentrated flavour enhancers rather than sources of dietary fibre.

The fat content of the seasoning samples was significantly higher than that of the commercial control seasoning due to the addition of 10 % hydrogenated fat during formulation. The fat content equally shows that fermentation has minimal effect on the fat content while drying helped to concentrate the fat making it more stable. The fat content ranging from 4.68 % in CCS to 11.03 % in FDSS can improve the sensory attributes of the seasoning cubes but can equally be detrimental as it can result to rancidity, as such proper packaging and storage conditions are necessary. These results are significantly lower than the 22.77 % to 29.33 % fat reported by Olaide *et al.* (2020), for bouillon cubes produced from fermented condiments of *Ricinus communis* L. seeds using cassava as a binder and closely related to the 3.28 % to 10.64 % fat reported by Ndife *et al.*, (2020) for seasoning mix produced from spice-leaves of Utazi, Uziza, Uda and Nchuanwu.

The results obtained for ash showed that crayfish contributed significantly to the mineral content of the seasoning cubes, as reflected in its high ash content of 7.35 %. The ash content in the seasoning samples ranged from 5.78 % to 6.63%, with no significant differences observed among them. This indicates that crayfish provide a consistent source of essential minerals such as calcium, phosphorus, and iron across all formulations. The low ash content of the FNSS sample showed that some minerals were consumed in the process of fermentation. The slight increase in the fermented and dried samples could also be explained by the fact that during fermentation as the organic compounds' breakdown, the remaining inorganic minerals become more concentrated in addition to drying. These results are lower than 58.77 and 67.27 % in Tahmaz *et al.* (2022), for vegetable food seasoning powders.

The results for carbohydrate content showed a great significant difference between the samples with the commercial control seasoning (CCS) having the highest content (71.15 %) and the fermented and dried salted seasoning (FDSS) having the lowest (46.76 %). The fermented and dried salted seasoning (FDSS) had the lowest carbohydrate content which could be attributed to the breakdown of carbohydrates during fermentation. The commercial control seasoning (CCS) has the highest carbohydrate content, suggesting the use of carbohydrate-rich ingredients such as starch or sugars for texture and flavor enhancement. Ndife *et al.* (2020), equally reported a high carbohydrate content of 39.71 to 58.43 % for spice-mix seasonings from functional leaves of Utazi, Uda, Nchanwu and Uziza.

The energy content of the seasoning cubes ranged from 354.64 KJ/cal (CCS) to 394.23 KJ/cal (FDSS). The FDSS sample had the highest energy content, reflecting its balanced macronutrient composition (high protein, moderate fat and lower carbohydrates). This makes FDSS a more nutrient-dense option compared to the commercial control seasoning, which had lower energy due to its higher carbohydrate content.

This research study showed that combining tree tomato and crayfish with appropriate fermentation and drying conditions of temperature and time yielded a versatile and nutritious seasoning product. The FDSS sample stands out as a promising candidate due to its high protein, moderate fat, and balanced energy content.

### Functional Properties of the Seasoning Cubes

Functional properties are essential physicochemical properties that describe the behavior of ingredients during preparation and cooking and how they affect the quality of finished products in terms of appearance, feel and taste (Awuchi *et al.*, 2019). The effect of treatments (fermentation and drying) on the bulk density, water absorption capacity, oil absorption capacity and dispersibility of the seasoning samples produced from tree tomato and crayfish were evaluated.

In this study, fermentation led to lower bulk density (0.41g/ml) as seen in FNSS. This could be because of the

production of gases and enzymes that alter the matrix structure. However, when combined with drying (FDSS), the bulk density increased (0.52 g/ml), suggesting that drying creates more compact particles compensating for the porosity induced by fermentation (Jayaraman & Gupta, 2020).

It could also be seen from the results that the salted samples have higher bulk densities than the non-salted samples implying that salt enhances structural cohesion thereby increasing the overall density of the mixtures. This result aligns with that of Kumar & Singh (2020), which indicated that drying has the tendency to restore the bulk density of fermented food products.

The bulk density of a food material is a critical factor in determining the type and amount of packaging material required. A higher bulk density implies that the food product is denser, requiring the use of more robust and thicker packaging materials to ensure adequate protection and containment. This relationship between bulk density and packaging material is essential for optimizing packaging efficiency and cost-effectiveness (Iwe *et al.*, 2016).

For water absorption capacity, it could be seen that fermentation enhanced water absorption in FDNSS, possibly due to the degradation of cell walls and the release of hydrophilic components. The water absorption capacity could also be as a result of the presence of hydrophilic compounds (polysaccharides) and fibre in tree tomato that readily absorb water. This aligns with the report of Li *et al.* (2022), on the effect of fermentation with lactic bacteria on the structural characteristics and physicochemical and functional properties of soluble dietary fiber from prosomillet bran where they concluded that fermentation increased the solubility and water holding capacity of its soluble dietary fiber.

Oil absorption was lower in non-salted samples compared to salted samples, indicating that salting plays a role in enhancing lipid interactions and possible oil absorption. The high oil absorption capacity in the fermented and dried salted samples could be explained by the fact that fermentation and drying breaks down proteins, modifying their structures and exposing hydrophobic sites that can enhance oil absorption while salt equally helps in modifying the protein structure and stabilizing the protein oil interactions.

These results agree with the results of research conducted by Li *et al.* (2022), in which they found out that salt can improve the texture and binding properties in plant-based meat analogues.

Water and oil absorption capacities are critical properties in the formulation of seasoning cubes, influencing their texture, stability, and flavor release. These properties determine how well the cubes can integrate with other ingredients during cooking and how they maintain their structural integrity. The balance between water and oil absorption is essential for achieving the desired culinary effects and consumer satisfaction (Oyedemi *et al.*, 2021).

The results obtained for dispersibility showed that fermentation improved dispersibility in both FNSS and

FDNSS. This is as a result of the breakdown of large molecules into smaller, more soluble compounds. This aligns with findings in a study by Chen *et al.* (2021), where fermentation enhanced the bioavailability and solubility of soy sauce. Also, salting reduced dispersibility, possibly due to the formation of stronger ionic bonds that hindered solubility. This is consistent with the results of García *et al.* (2023), where salt reduced the water solubility in seasoned meat products.

The commercial control seasoning (CCS) had intermediate values for most properties, indicating that industrial processes may optimize certain attributes but not necessarily surpass the functional properties achieved through natural treatments like fermentation and drying. The higher dispersibility of FNSS compared to CCS suggests that natural fermentation offers advantages in terms of solubility and functionality over commercially processed seasonings.

The dispersibility of seasoning cubes is an important parameter as it ensures that the seasoning cube dissolves quickly and evenly, preventing clumping and ensuring a consistent flavor throughout the dish (Mohd Asri *et al.*, 2021).

### Effect of Treatments on the Flavor Compounds of the Seasoning Cubes

The flavor of a food can be defined as the subtle interaction of taste and aroma that imparts a pleasing or displeasing sensory experience to an individual (Ojinnaka & Agubolum, 2013). The GC-MS analysis of the five samples revealed significant differences in volatile compound profiles. Pleasant volatile compounds from fermentation were distributed among all fermented samples (FDNSS, FDSS and FNSS), but they were higher in the FDNSS sample. The fermentation volatile compounds included alcohol, aldehydes, esters, ethers, ketones, and acids (Pico *et al.*, 2015).

The FDNSS and FNSS Sample displayed the greatest chemical diversity with 32 and 30 identified flavor compounds respectively, predominantly terpenes and terpenoids, signifying the rich profile of tree tomato fruits and crayfish. FDNSS exhibited the highest percentage of flavor active compounds at 64.5% of total peak area, dominated by monoterpenes including  $\alpha$ -phellandrene (7.12 %), p-cymene (7.43 %), and D-limonene (1.18 %), alongside sesquiterpenes such as  $\gamma$ -muurolene (6.20 %) and germacrene D (2.42 %). This shows the great synergistic effect of fermentation and controlled drying at temperatures of 55 oC which lead to the production of new compounds and minimal losses. Also, the absence of salt in the samples shows that there was extensive hydrolysis of proteins, carbohydrates and volatiles without much inhibition (Adebo & Medina, 2020).

The salted samples CSS and FDSS had fewer flavor compounds 4 and 7 respectively than the non-salted samples. This suggests a possibility of inhibition of hydrolytic enzymes by salt particularly of lactic acid bacteria and proteolytic enzymes responsible for liberating

volatile precursors (Nwachukwu *et al.*, 2021). Salt is reported to lower water activity, slow metabolic activity and reduce hydrolysis reactions. This could explain why the samples recorded just few volatile flavor compounds. These results are in line with the research conducted by Eze *et al.* (2023) on fermented fish powders where they recorded that salt reduced volatile flavor compounds by over 70 % than non-salted samples.

Looking at the commercial control seasoning, the results showed that only 2 volatile compounds-1 terpenoid and 1 alcohol. This shows the industrial preference of commercial seasoning on high temperature processing and the use of synthetic flavor enhancers such as monosodium glutamate, monosodium guanylate and some nucleotides.

The results obtained in this study aligns with that of Xing *et al.* (2025) who indicated that the microbial fermentation process transforms flavor compounds, leading to a richer aroma and the masking or reduction of some off flavors present in the raw fruit material. Further, the researchers identified that fermented products are more attractive than other products because of their unique flavors, which are largely derived from the metabolic activities of the fermenting strains. Ge *et al.* (2025) in another study found out that the fermentation process increases volatile aroma components, such as esters, ketones, alcohols, and terpenes, in addition to retaining beneficial compounds like phenols and flavonoids from the original fruit.

From the results gotten, it could be concluded that the seasoning samples produced from tree tomato and crayfish had a more versatile and natural flavor profile than the commercial control seasoning emphasizing the value proposition of the produced seasoning samples.

### Effect of Treatments on the Microbial Quality of the Seasoning Cubes

The results obtained showed clearly that the different treatments (fermentation and drying) and salting significantly influenced the microbiological quality of the formulated seasonings. The CSS seasoning recorded the lowest bacterial count, showing that salt reduced the water activity of the sample by dehydrating the bacteria cells through osmotic pressure. The treated samples showed higher bacteria count than the non-treated sample (CSS) showing that fermentation resulted in the proliferation of microorganisms in the sample.

However, the total bacteria count in the fermented and dried samples was lower than in the fermented samples. This shows that drying equally contributed to the reduction of the moisture content which inhibits the proliferation of spoilage microorganisms. The results of the total bacteria count ranging from  $4.24 \times 10^3$  cfu/g to  $8.78 \times 10^3$  cfu/g is below the  $1 \times 10^5$  cfu/g threshold which is an accepted limit for dried food products as stated by the International Commission on Microbiological Specifications for Foods (ICMSF, 2022). This could be because of the effect of salt, drying and equally the influence of ginger which has been reported to have antimicrobial properties.

Also, the Codex Alimentarius Commission (2021) and European Union regulations consider total aerobic microbial counts (TAMC) of  $\leq 10^5$  cfu/g acceptable for dried spices and condiments. From the results, it could be seen that the highest total bacteria count score is close to the safe limit suggesting that microbial inhibition was not properly done and equally suggesting that there must have been some level of post-production contamination. This calls for improvement in the aseptic processing conditions and proper packaging and handling after production.

These results obtained are similar to the 6.94 and 7.74 log<sub>10</sub> CFU/g for “Soumbara” powder reported by Massamba *et al.* (2024). The results were equally lower than the  $6.6 \times 10^3$  cfu/g to  $33 \times 10^3$  cfu/g for instant noodles and the accompanying seasonings reported by Lipşa *et al.* (2020).

For the total fungal count, the CCS recorded the lowest ( $4.16 \times 10^3$  cfu/g) fungal count. This could be attributed to the effectiveness of the industrial processing methods, possibly involving thermal treatment, strict hygiene, and chemical preservatives. The highest fungal count  $7.92 \times 10^3$  cfu/g and  $7.03 \times 10^3$  cfu/g recorded in the FNSS and FDNSS shows that the absence of salt created a conducive environment for the proliferation of fungi. Salt is reported to act as a humectant, reducing water activity and inhibiting the growth of spoilage microorganisms (Olasupo *et al.*, 2019).

The range of total fungal count ( $4.16 \times 10^3$  to  $7.92 \times 10^3$  cfu/g) obtained in these results is higher than the recommended fungal count limit of less than  $1 \times 10^3$  cfu/g reported by FAO/WHO, 2022 to prevent microbial spoilage and risk of mycotoxins. This could be attributed to postproduction contamination and improper packaging and storage conditions. The results call for proper handling and storage conditions to prevent the pathogenicity of the samples.

The results obtained in this research are in alignment with the results reported by Nwachukwu *et al.* (2023). They reported that the microbial load of fermented crayfish-tomato blends without salt were higher than salted counterparts, attributing it to the loss of osmotic inhibition. Similarly, a study conducted on ogiri-based condiments by Anukam *et al.* (2021) reported that drying after fermentation without salting resulted in recontamination and elevated fungal counts, reinforcing the need for combined preservation techniques.

### CONCLUSIONS

To conclude, sensory evaluation showed that the inclusion of salt significantly enhanced the sensory properties of the formulated seasoning cubes as compared to the commercial seasoning. Meanwhile, the synergistic application of fermentation and drying resulted to new flavour compounds and equally the concentration of already existing ones. This hurdle technology effectively addresses the needs of both hypertensive and non-hypertensive persons. The proximate analysis highlighted

the effectiveness of fermentation and drying in enhancing the nutritional quality of seasoning cubes as the fermented and dried salted seasoning (FDSS) emerged as the most promising formulation, offering high protein and balanced micronutrients. This research provides valuable insights into developing sustainable and nutritious food products from locally available food crops, aligning with current trends in food science and technology.

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