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The Effect of Fermentation Time on the Proximate Composition of Maize Grains (*Zea mayz*)

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Abstract: **The proximate composition was carried out by standard methods. The values are all in percentages. The moisture content increased by, 2.50, 5.50, 1.50, and 3.50 on day 1, 2, 3, and 4 of fermentation respectively. In the same order, the ash content increased by 2.50, 6.50, 1.00 and 6.50. Crude protein decreased by 0.88, 0.30, 0.18, and 0.61, on day 1, 2, 3 and 4 respectively. The crude fiber decreased by 0.50, 0.50, 1.50 and 1.00 on the 1st, 2nd, 3rd, and 4th day of fermentation respectively. Crude fat on the other hand increased by 1.50, 1.00, 2.50, and 2.50 respectively. And, finally the carbohydrate decreased by 5.12, 12.19, 3.33 and 10.89 in that order.**

Keywords: **Composition, Fermentation, Food, Maize, Nutrient, Proximate, Time.**

1. INTRODUCTION

Fermentation is a metabolic process that produces chemical changes in organic substrates through the action of enzymes. In biochemistry, it is narrowly defined as the extraction of energy from carbohydrates in the absence of oxygen. In the context of food production, it may more broadly refers to any process in which the activity of microorganisms brings about a desirable change to a foodstuff or beverage (Hui, 2004). The science of fermentation is known as zymology. In microorganisms, fermentation is the primary means of producing adenosine triphosphate (ATP) by the degradation of organic nutrients anaerobically (Klein *et al.*, 2006).

Humans have used fermentation to produce foodstuffs and beverages since the Neolithic age. For example, fermentation is used for preservation, a process that produces lactic acid found in such sour foods as pickled cucumbers, kombucha, kimchi, and yogurt, as well as for producing alcoholic beverages such as wine and beer. Fermentation also occurs within the gastrointestinal tracts of all animals, including humans (Bowen Richard, 2018)

Fermentation is a way of extracting energy from molecules, but it is the only one common to all bacteria and eukaryotes. It is therefore considered the oldest metabolic pathway, suitable for an environment that did not yet have oxygen (Tobin and Dusheck, 2005). Yeast, a form of fungus, occurs in almost any environment capable of supporting microbes, from the skins of fruits to the guts of insects and mammals and deep ocean, and harvests sugar-rich materials to produce ethanol and carbon

dioxide (Bass *et al.*, 2007). The basic mechanism for fermentation remains present in all cells of higher organisms. Mammalian muscle carries out fermentation during periods of intense exercise where oxygen supply becomes limited, resulting in creation of lactic acid (Donald and Judith, 2010). In invertebrates, fermentation also produces succinate and alanine (Broda, 2014).

Fermentative bacteria play an essential role in the production of methane in habitats ranging from the rumens of cattle to sewage digesters and freshwater sediments. They produce hydrogen, carbon dioxide, formate and acetate and carboxylic acids; and then consortia of microbes convert the carbon dioxide and acetate to methane. Acetogenic bacteria oxidize the acids, obtaining more acetate and either hydrogen or formate. Finally, methanogens (in the domain Archea) convert acetate to methane (Ferry, 1992).

Maize grain is readily available in the societies of the world and has high economic value. All over the world, different methods of processing food have been developed. Fermentation of maize grains for the variety of household and industrial foods has been in effect. However, most of these fermentations are done once in a fixed time range. Furthermore, most of the fermentation exercises are done by the locals to merely soften these grains for effective grinding into paste without knowing that fermentation readily occurs in the process. And little or nothing is known of its effect on the nutritional status of the grains. This study, therefore, undertake the novel task of finding out the effect of varying fermentation time on the proximate content of maize grain.

Besides, most research woks involving fermentation were conducted at a fixed time. But, this research work, tends to provide an evidence based scientific data of the effect of varying fermentation time on the proximate composition of maize grain. The information derived from this work may serve as an eye opener to processors of foods on the effect of time intervals of fermentation as a method of food processing on the proximate contents of maize grains.

2. MATERIALS AND METHODS

Sample collection and preparation

The maize grain sample was collected from Samaru market Sabongari LGA, Zaria Kaduna state, Nigeria in a clean polythene bag container. It was manually sorted and washed in order to remove the spoilt grains, dust, and extraneous matter from the grain. The sorted grain was divided into 5 equal portions. One portion was not fermented and served as a control. Each of the remaining 4 portions was sucked in distilled water at the ratio of 1:4(w/v) grain to water and allowed to ferment for 24, 48, 72 and 96 hours respectively. At the end of each fermentation period the grain was separated from the steeping water by decantation and grind to paste.

Proximate analysis

The proximate analysis was carried out in all the samples for the following nutrients: protein, carbohydrate, lipids, moisture, ash and crude fiber using standard methods described by AOAC (2005) etc.

Determination of Moisture Content

Aluminium dishes were washed and dried to a constant weight in an oven at 100^{o}C. They were removed, cooled and weighed (W1). 2 grams of the paste or flour sample was placed in the weighed moisture dish (W2). The dish containing the sample was kept in an oven for about 3 hours, then removed, cooled and weighed (W3). The % of moisture was calculated as: $\frac{W2-W3}{W2-W1}\times 100$

Determination of Fibre

Two grams of the sample was placed in a beaker containing 1.2ml of H_2SO_4 per 100ml of solution and boiled for about 5 minutes, the residue was filtered and washed with hot water then transferred to a beaker containing 1.2 grams NaOH per 100ml of solution and boiled for about 5 minutes. The residue was washed with hot water and dried in an oven and weighed (C2). The weighed sample was incinerated in a furnace of about 550^{o}C then removed, cooled and weighed (C3). The % fibre was calculated as: $\frac{C2-C3}{W}\times 100$

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Determination of Ash

Crucibles were cleansed and dried in the oven, after drying, they were cooled and weighed (W1). 2g of the paste or flour was placed in the crucibles and weighed (W2). They were transferred into the Muffle furnace of about 550°C then removed, cooled and weighed (W3). The % Ash was calculated as: $\frac{W3-W1}{W2-W1} \times 100$

Determination of Lipids (Fats)

250ml clean boiling flask was dried in oven, and cooled. Empty filter paper was weighed and labeled (W1). Two grams of sample was weighed into the labeled filter paper (W2). The boiling flask was filled with petroleum ether. The soxhlet apparatus was assembled and allowed refluxing for 8 hours then, removed and transferred to an oven and dried. It was then cooled and weighed (W3). The % fat was calculated as: $\frac{W2-W3}{W2-W1} \times 100$

Determination of Protein

Digestion: 2g of sample was weighed into a Kjeldahl flask. Copper catalyst and 15ml of concentrated sulfuric acid were added. It was heated in a fume cupboard till solution assumed a green colour. It was cooled and black particles showing at the mouth and neck of the flask were washed down with distilled water. The digest then washed thoroughly with distilled water.

Distillation: the Markham distillation apparatus was steamed through for about 15 minutes before use. 100ml conical flask containing 10ml of boric indicator was placed under the condenser. 10ml of the digest was pipetted into the body of the apparatus via the small funnel aperture; washed down with distilled water followed by 10ml of 40% NaOH solution.

Titration: the solution was titrated in the receiving flask using N/100 (0.01N) hydrochloric acid and the Nitrogen content and hence the protein content of the sample was calculated. The blank was run through along with the sample. The % protein was calculated as:

$$final\ reading - initial\ reading - blank(0.2) \times \frac{standard\ number\ of\ nitrogen(1.4)}{initial\ weight(0.5)} \times standard\ number\ of\ protein(6.25)$$

Determination of carbohydrate (CHO)

CHO content was determined by difference: $100 - (\%moisture + \%ash + \%protein + \%fat)$

3. RESULTS

Table 1: The effect of fermentation time on the proximate composition of maize grain

Duration (hours)	Moisture (%)	Ash (%)	Crude Protein (%)	Crude Fiber (%)	Crude Fat (%)	Carbohydrate (%)
0 (Unfermented)	17.40	5.90	8.53	11.50	12.50	44.16
24	19.90	8.40	7.65	11.00	14.00	39.04
48	25.40	14.90	7.35	10.50	15.00	26.85
72	26.90	15.90	7.17	9.00	17.50	23.52
96	30.40	22.40	6.56	8.00	20.00	12.63

Table 2: Increases/decreases in proximate values for each of the fermentation period respectively

Proximate parameter	Day 1 (%)	Day 2 (%)	Day 3 (%)	Day 4 (%)
Moisture	2.50	5.50	1.50	3.50
Ash	2.50	6.50	1.00	6.50
Crude protein	-0.88	-0.30	-0.18	-0.61
Crude fiber	-0.50	-0.50	-1.50	-1.00
Crude fat	1.50	1.00	2.50	2.50
Carbohydrate	-5.12	-12.19	-3.33	-10.89

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Table 3: Increases/decreases of proximate values for each of the fermentation period from day 0 respectively

Proximate parameter	Day 1 (%)	Day 2 (%)	Day 3 (%)	Day 4 (%)
Moisture	2.5	8.00	9.50	13.00
Ash	2.50	9.00	10.00	16.00
Crude protein	-0.88	-1.18	-1.36	-1.97
Crude fiber	-0.50	-1.00	-2.50	-3.50
Crude fat	1.50	2.50	5.00	7.50
Carbohydrate	-5.12	-17.31	-20.64	-31.53

4. DISCUSSION

Table 1 gives the raw values, depicting the effect of fermentation time on the proximate parameters. Table 2 gives the percentage increase or decrease in values of proximate nutrients on each day of the fermentation process. While table 3 gives the overall percentage increase or decrease on proximate nutrients for each day of fermentation from day 0.

The highest increase in value is 6.50% of the ash content on day 2 and 4. The highest decrease in value is 10.89% of carbohydrate only on day 4 fermentation period. The lowest increase in value is 1.00% of the ash content on day 3 and crude fat on day 2. While the lowest decrease is 0.3% of crude protein on day 2 fermentation period (Table 2).

On the other hand, the overall highest increase from day 0 is 16.00% of Ash content. While the overall highest decrease from day 0 is 31.53% of carbohydrate (Table 3)

The result obtained from proximate analysis shows how fermentation affects the proximate composition of maize grain. Generally, it is observed that the fermentation reduces the crude protein, crude fiber and carbohydrate contents of the grain and increases the moisture, ash, and crude fat contents. The reduction may be due to the fact that most of the chemical constituents have been converted to the products of fermentation (ethanol and CO_2)

The crude protein content decreases as the fermentation time progresses. The organisms that facilitate fermentation utilize protein as a source of energy. For instance, using a protein building block, an amino acid, called sereine can be oxidatively deaminated to pyruvate. Pyruvate can be converted to lactic acid or acetylcoa which enters citric acid cycle to generate energy (Ambugus, 2019).

The crude fiber decreases as the fermentation time increases. This may be attributed to enzymatic degradation of the fibrous materials during fermentation (Koko and Ingram, 1986).

crude fat content increases with increase fermentation period as expected, this is because of the conversion of soluble carbohydrate into lactic acid during anaerobic glycolysis also known as fermentation (*Gajera et al.*, 2008).

The carbohydrate content decrease as the fermentation time increases. The reduction in carbohydrate content in fermentation could be attributed to the utilization of fermentable sugar by lactic acid bacteria for growth and other metabolic activities. (Davelin, 2011)

5. CONCLUSION

In this research, it is observed that fermentation process significantly changed the nutritional value of maize grain by affecting the energy value and nutrient densities of the grains.

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