



Experimental Investigation of the Functional and Proximate Properties of Pigeon Pea (*Cajanus cajan*) Using Fermentation Process

Bosesde Folake Olanipekun^{1*}, Joy A. Ilevbare¹, Olusegun James Oyelade²,
Oluyemisi Elizabeth Adelakun¹ and Ajekigbe Sola Olajire²

¹Department of Food Science, Faculty of Food and Consumer Sciences, Ladoke Akintola University of Technology, Ogbomoso, Oyo State, Nigeria.

²Department of Food Engineering, Faculty of Engineering and Technology Ladoke Akintola University of Technology, Ogbomoso, Oyo State, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. Author BFO designed the study, supervised the project and wrote the first draft of the manuscript. Authors JAI and OJO contributed to the literature search, method of the research and corrected the final manuscript authors OEA and ASO carried out the processing, laboratory analysis and managed the analyses of the study. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/EJNFS/2021/v13i530418

Editor(s):

(1) Dr. Rasha Mousa Ahmed Mousa, University of Jeddah, Saudi Arabia.

Reviewers:

(1) Ravi Kant Avvari, National Institute of Technology, India.

(2) Lucia Maria Jaeger De Carvalho, Rio De Janeiro Federal University, Brazil.

Complete Peer review History: <https://www.sdiarticle4.com/review-history/71991>

Original Research Article

Received 07 June 2021
Accepted 13 August 2021
Published 19 August 2021

ABSTRACT

Pigeon pea (*Cajanus cajan*) is a leguminous crop rich in protein and widely cultivated in Nigeria but underutilized due to high anti-nutritional properties. Several processing methods such as fermentation using non-pathogenic micro fungi have been used to address these problems. This study evaluated the effect of fermentation on the functional and proximate properties of pigeon pea using *Rhizopus* species. The following ranges of values were obtained for bulk density, water absorption, oil absorption and swelling capacities (0.74-0.98g/ml; 71.67-189.00%; 120.00-205.00%; 6.81-10.93g/ml), respectively. Values of proximate composition were in ranges of 6.67-7.87, 16.09-22.09, 2.83-4.00, 0.84-1.23, 1.33- 2.00 and 65.47- 71.35% for moisture, protein, ash, fat, fibre and carbohydrate contents, respectively. Improvement in functional and proximate properties of fermented pigeon pea flour were observed. Hence, it can also serve as functional and supplement ingredients in different food formulations.

*Corresponding author: Email: bfolanipekun@lautech.edu.ng;

Keywords: Pigeon pea; fermentation; water absorption; bulk density; *Rhizopus*.

1. INTRODUCTION

Pigeon pea is recognised as perennial legume having good nutritional values that can be used in the management of metabolic problems in human and animals [1]. According to Venkidasamy *et al.* [2], pigeon peas is a typical legume rich in protein and are served as staple food for large population in India. This accounts for India being the largest supplier of the pigeon pea in the world market [3]. The rate of pigeon pea intake thus has contributed to consumption of reduced fat, and consequently helping in building the body weight [4].

Despite the potentials of pigeon pea, there seems to be problems associated with its processing making it impossible for consumers to adequately subscribe to its consumption. For instance, Feng *et al.* [5] discovered that certain methods for its processing pose some quality defects such as quality loss in the food, taste, flavour and colour to make it unhealthy for consumption. However, optimization of the fermentation parameters gives a better quality of the pigeon pea. More so, Lee *et al.* [6] reported that fermented pigeon pea improves the regulation of blood pressure which helps in reducing cardiovascular health challenges. Based on this, Olagunju *et al.* [7] hydrolysed pigeon pea protein using different enzymes and observed that membrane ultrafiltration exists which has the potential of improving the functional recipes in industry in relation to plant based products.

Many researchers focused on utilizing the pigeon pea towards further enhancement of the nutritional value of some foods. For instance, study by Solomon *et al.* [8] revealed that toasted pigeon pea can be used to improve nutritional quality of cat fish, thereby reducing the cost of catfish production. In view of this, Martinez-Villaluenga *et al.* [9] further applied fermented *Cajanus cajan* powder to improve the quality of protein in pasta. Further studies on pigeon pea showed that different modification methods have effects on both functional and proximate properties to consequently influence the variation in their suitability for varieties of products that can be developed [10-15]. Thus, the aim of this study is to investigate effect of fermentation on the functional and proximate properties of pigeon pea (*Cajanus cajan*).

2. MATERIALS AND METHODS

2.1 Materials

Pigeon pea seeds were obtained from a local market in Ogbomoso while the *Rhizopus species* were obtained from Microbiology Laboratory of Food Science and Engineering Department LAUTECH, Ogbomoso.

2.2 Methods

2.2.1 Preparation of subculture for pigeon pea fermentation

Rhizopus oligosporus, *Rhizopus oryzae* and *Rhizopus nigricans* were sub cultured according to the procedure of Olanipekun *et al.* [16]. Five hundred millilitre (500ml) of Potato Dextrose Agar (PDA) was obtained through dissolution of 18 g of PDA to 500 ml of distilled water, homogenised and subsequently sterilized. After cooling, 15 ml of PDA was dispensed into McCartney bottles, and allowed to set in slant form. The pure cultures of *Rhizopus oligosporus*, *Rhizopus oryzae* and *Rhizopus nigricans* were sub cultured singly into McCartney bottles and incubated at 30°C for 4 days.

2.2.2 Procedure of pigeon pea fermentation

The pigeon pea seeds were cleaned, steeped (28 ±2 °C for 24 h), dehulled, boiled (100 °C for 15 min), drained and cooled. Subsequently, the seeds were inoculated using different *Rhizopus species* (*Rhizopus oryzae*, *Rhizopus oligosporus* and *Rhizopus nigricans*), sealed in polyethylene bag, incubated (32 °C for 72 h), pulverized, dried (55 °C for 24 h), cooled, milled and sieved to produce fermented pigeon pea flour. The unfermented pigeon pea (0 h fermentation) serves as the control and at regular intervals of 12 h; sample was taken out for appropriate analysis.

2.3 Analysis of the Samples

Water absorption capacity was carried out using the procedure of Owuamanam *et al.* [17] and bulk density was determined by using the method of Nwanekezi *et al.* [18]. Swelling capacity of the samples was determined by Tester and Morrison [19]. The proximate composition was determined by the method

described by AOAC [20] while carbohydrate was calculated by difference. Oil absorption capacity was determined using the method of Adelokun et al. [21]. All the analyses were performed in triplicates.

3. RESULTS AND DISCUSSION

3.1 Functional Properties of Fermented Pigeon Pea

The results of functional properties of fermented pigeon pea presented in Table 1 showed decreasing trend in water absorption of pigeon pea fermented using different species of *Rhizopus nigricans*, *Rhizopus oryzae* and *Rhizopus oligosporus* singly within 24-72 h, respectively. The trend in water absorption was observed to follow decreasing pattern of 189.00-74.67, 179.00 -71.67 and 174.00-73.33%, respectively. Consequently, the findings are in agreement with the studies by Olanipekun et al. [22], and Oyelade et al. [23] as water absorption was found to decrease for cocoyam and soya beans due to fermentation.

Swelling capacity exhibited slight decrease in their values which ranged as 8.27- 6.81, 10.93 - 7.11 and 10.74- 7.23 g/ml, respectively using *Rhizopus nigricans*, *Rhizopus oryzae* and *Rhizopus oligosporus*. Fermenting pigeon pea with *Rhizopus oryzae* gave the highest value of 10.93 g/ml whereas the least value of 6.81 g/ml was obtained using *R. nigricans* at 72 h of fermentation period. There were significant differences between the samples (p<0.05). It was observed that swelling capacity decreased with

increase in fermentation period. The result obtained is in agreement with the report of Lawal et al. [24] on effects of selected Hofmeister anions on functional properties of protein isolate prepared from lablab seeds. Adebowale and Maliki [25] found similar trend on the chemical composition and functional properties of fermented pigeon pea (*Cajanus cajan*) flour. Variation in swelling power observed could be due to effect of particle size and nature of unmodified starch in the pigeon pea as probably affected by fermentation which invariably may affect stickiness.

The oil absorption capacity of fermented flour decreased as the fermentation period increases except when using *Rhizopus nigricans* that showed contrary trend. The values ranged from 172.00-183.00, 199.00 -194.00 and 205.00-120.00%, respectively using *Rhizopus nigricans*, *R. oligosporus* and *Rhizopus oryzae*. The control sample had the value of 185.00%. Fermenting pigeon pea with *Rhizopus oligosporus* for 48 h gave the highest value while the least value obtained when using *Rhizopus oryzae* at 72 h. The decreasing trend agreed with the report of Adebowale and Malaiki [25] on the pigeon pea flour.

Bulk density of fermented pigeon pea increased as the fermentation period increases. This is indicated in the values 0.74 – 0.98, 0.77-0.98 and 0.92-0.98g/ml obtained when using *Rhizopus nigricans*, *Rhizopus oligosporus* and *Rhizopus oryzae*, respectively. The highest value was obtained in samples C, F and I while the least value obtained in sample A. Fermenting

Table 1. Functional properties of fermented pigeon pea

Sample	Water Absorption Capacity (%)	Swelling Capacity (g/ml)	Oil Absorption Capacity (%)	Bulk Density (g/ml)
A	189.00±4.16 ^a	8.27±0.500 ^c	172.00±7.211 ^d	0.74±0.000 ^d
B	194.00±5.774 ^a	8.19±0.388 ^c	162.00±1528 ^e	0.94±0.025 ^b
C	74.67±1.528 ^e	6.81±0.799 ^e	183.00±8.622 ^c	0.98±0.027 ^a
D	179.00±5.686 ^a	10.93±0.397 ^a	199.00±8.083 ^a	0.77±0.023 ^d
E	170.00±1.824 ^c	8.94±1.101 ^b	208.00±1.000 ^a	0.93±0.025 ^b
F	71.67±2.517 ^e	7.11±0.549 ^e	194.00±9.292 ^a	0.98±0.027 ^a
G	174.00±2.539 ^b	10.27±0.450 ^a	205.00±10.066 ^a	0.92±0.122 ^c
H	154.00±2.000 ^d	10.74±0.214 ^a	184.00±15.044 ^c	0.95±0.000 ^a
I	73.33±3.786 ^e	7.23±0.387 ^d	120.00±7.506 ^f	0.98±0.027 ^a
J	66.33±0.577 ^f	11.93±0.809 ^a	185.00±4.993 ^{bcd}	0.90±0.023 ^b

Sample A, B and C: Pigeon pea fermented with *Rhizopus nigricans* at 24, 48 and 72 h, respectively
 Sample D, E and F: Pigeon pea fermented with *Rhizopus oligosporus* at 24, 48 and 72 h, respectively
 Sample G, H and I: Pigeon pea fermented with *Rhizopus oryzae* at 24, 48 and 72 h, respectively
 Sample J: Control sample

pigeon pea using *Rhizopus nigricans* at 24 h gave the least value compared to the highest values obtained at 72 h using *Rhizopus nigricans*, *Rhizopus oligosporus* and *Rhizopus oryzae*, respectively. The bulk density values obtained in this study were higher than those of Edema et al. [26] obtained for soybean flour (0.38g/ml). However, the result of the findings is in agreement with the reports of Onimawo et al. [27] which had the values that ranged from 0.06 and 0.60-0.75g/ml for cowpea and bambara groundnut, respectively.

3.2 Proximate Composition of Fermented Pigeon Pea

The result of proximate composition of fermented pigeon pea flour using *Rhizopus nigricans*, *Rhizopus oligosporus* and *Rhizopus oryzae* is presented in Table 2. The level of protein content increased from 17.10 – 22.09, 18.27 -20.13 and 16.71 -18.49% using *Rhizopus nigricans*, *Rhizopus oligosporus* and *Rhizopus oryzae*, respectively. Fermenting pigeon pea with *Rhizopus nigricans* gave the highest value of 22.09% while the least value of 16.09% was obtained in an unfermented sample. The increase in protein content is in agreement with the results obtained during fermentation of beniseed flour and pearl millet according to Oshodi et al. [28]; Fasasi [29].

The ash content of the fermented pigeon pea showed decreasing trend from 3.17 -3.00, 3.67 – 2.67 and 3.50-2.83% using *Rhizopus nigricans*, *Rhizopus oligosporus* and *Rhizopus oryzae*, respectively. Fermenting pigeon pea with *Rhizopus oligosporus* gave the highest value of 4.00% whereas sample F fermented using *Rhizopus oligosporus* at 72 h gave the least value of 2.67%. Increment in the ash content of fermented pigeon pea was in accordance with the reports of SefaDedeh et al. [30]; Adebawale and Maliki [25] on the fermentation of cowpea and fortification for the production of traditional foods, respectively. This similar finding was reported by Emmanuel et al. [31] for locust bean (*Parkia biglobosa*).

The moisture content of pigeon pea fermented using *R. nigricans* increased from 6.90-7.20 % whereas others slightly decreased from 7.20-7.13% and 7.30 -6.69 % using *Rhizopus oligosporus* and *Rhizopus oryzae*, respectively. Fermenting pigeon pea using *Rhizopus oligosporus* and *Rhizopus oryzae* which

indicated decreasing trend is good for enhanced keeping quality of product. Similar trend has been observed on complementary food formulated from fermented maize, soybean and carrot water as reported by Lucretia et al. [32].

Fat content of the fermented pigeon pea flour showed a decreasing trend ranging from 1.10 – 0.80, 1.01-0.91 and 0.92- 0.84% using *Rhizopus nigricans*, *Rhizopus oligosporus*, *R.oryzae*, respectively. The control sample had the highest value of 1.23% while sample C having the lowest value of 0.80%. Slight differences were observed in all samples at $p<0.05$. The result obtained is in agreement with the findings of Adebawale et al.,[33]; Anthony and Babatunde [34]; Emmanuel et al. [31] on fermented legumes and cereals. The low-fat content observed has potential to improve shelf-life through rancidity reduction.

The carbohydrate content of fermented pigeon pea flour showed a slight decreasing trend from 67.78 -65.47, 67.86- 67.40 and 69.81-68.56% using *Rhizopus nigricans*, *Rhizopus oligosporus* and *Rhizopus oryzae*, respectively. Sample J had the highest value of 71.35 % and Sample C had the lowest values of 65.47%. Slight differences were observed in all the samples at $p<0.05$. This result obtained is in accordance with the findings of Emmamuel et al. [31]; Ahmed et al. [35] during fermentation of locust bean and guar seeds. The decreasing trend was attributed to increase activity of amylolytic enzymes which hydrolyses starch and other complex carbohydrate to simpler starch (Adebawale and Maliki, [25]; Ojokoh et al., [36].

Fiber content of the fermented pigeon pea flour showed decreasing values of 1.65 – 1.50, 2.00-1.33 and 1.75-1.42% using *Rhizopus nigricans*, *Rhizopus oligosporus* and *Rhizopus oryzae*, respectively. Sample D had the highest value of 2.00% and sample F had the lowest value of 1.33%. Generally, the decreasing trend in fiber content could be linked to the observation of Omafuvbe et al. [37] indicating that at the end of boiling process, the boiled water obtained from the beans was observed to be more viscous than it was at the beginning of the process. The result is also similar to the finding of Onweluzo [38] that reported decrease during the fermentation of millet and pigeon pea seed for flour production.

Table 2. Proximate composition of fermented pigeon pea flour

Sample	Moisture Content (%)	Protein %	Ash (%)	Fat (%)	Fibre (%)	Carbohydrate (%)
A	6.90±0.529 ^a	17.10±1.186 ^a	3.17±0.289 ^b	1.10±1.123 ^{ab}	1.65±0.132 ^{abc}	67.78±1.642 ^{ab}
B	7.13±0.100 ^a	21.51±1.800 ^a	3.17±0.289 ^b	1.08±0.090 ^{abc}	1.58±0.144 ^{bcd}	65.76±1.505 ^b
C	7.20±0.200 ^a	22.09±2.461 ^a	3.00±0.000 ^b	0.80±0.108 ^d	1.50±0.000 ^{cd}	65.47±2.829 ^b
D	7.20±0.200 ^c	18.27±2.117 ^{ab}	3.67±0.289 ^a	1.01±0.045 ^{bcd}	2.00±0.000 ^d	67.86±2.305 ^{ab}
E	7.47±0.577 ^c	19.87±3.778 ^{ab}	4.00±0.000 ^a	0.99±0.189 ^{bcd}	1.42±0.144 ^{cd}	67.52±4.216 ^{ab}
F	7.13±0.500 ^a	20.13±1.048 ^{ab}	2.67±0.289 ^d	0.91±0.106 ^{bcd}	1.33±0.144 ^d	67.40±0.980 ^{ab}
G	7.30±0.321 ^a	16.71±0.567 ^b	3.50±1.000 ^{ab}	0.92±0.134 ^{cd}	1.75±0.500 ^{abc}	69.81±2.317 ^{ab}
H	7.87±0.416 ^a	17.33±1.444 ^b	3.50±0.500 ^{abc}	0.87±0.072 ^{cd}	1.75±0.250 ^{abc}	69.42±2.005 ^{ab}
I	6.67±1.102 ^a	18.49±2.677 ^{ab}	2.83±0.289 ^{cd}	0.84±0.283 ^d	1.42±0.144 ^{abc}	68.56±2.580 ^{ab}
J	6.67±1.102 ^{ab}	16.09±2.155 ^a	2.83±0.289 ^{ab}	1.23±0.178 ^d	1.83±0.144 ^{cd}	71.35±3.421 ^b

Sample A, B and C: Pigeon pea fermented with *Rhizopus nigricans* at 24, 48 and 72 h, respectively
Sample D, E and F: Pigeon pea fermented with *Rhizopus oligosporus* at 24, 48 and 72 h, respectively
Sample G, H and I: Pigeon pea fermented with *Rhizopus oryzae* at 24, 48 and 72 h, respectively
Sample J: Control sample

4. CONCLUSION

Pigeon pea (*Cajanus cajan*) fermented using different *Rhizopus* species has been investigated for its functional and proximate properties. Improvement in functional and proximate properties of fermented pigeon pea flour were observed. Therefore, the product has good potential to serve as functional ingredient and supplement in different food formulations

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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