

## Phenomenological Model of Percentage Protein in fermented Iru (*Parkia biglobosa*)

<sup>1\*</sup>Modupe Elizabeth Ojewumi, <sup>2</sup>Abiodun James Omoleye and <sup>3</sup>Adesola Adetutu Ajayi

<sup>1\*,2</sup>Chemical Engineering Department, Covenant University, Km 10, Idiiroko Road, Canaan land, P.M.B. 1023, Sango Ota, Ogun state, Nigeria, <sup>3</sup>Augustine University, P.M.B 1010, Epe, Lagos.

Correspondence to: modupeojewumi@covenanataniversity.edu.ng

\*<sup>1</sup>Orcid: 0000-0002-9254-2450

### Abstract

The optimum conditions for the fermentation of African locust bean (*Parkia biglobosa*) into a vegetable protein based condiment (Iru) were developed using Levenberg-Marquardt (or Powell) method (using PSI software) with three (3) variables namely; inoculum concentration (*bacillus subtilis*), temperature and the fermentation duration. African locust bean seeds were fermented at various temperature of 40 - 70 °C for five days (120 hours) with different concentrations of Inoculum. The proximate analysis shows that fermentation increased the percentage protein. Protein had the highest composition with about 51 % after 72 hours at the lowest fermentation temperature of 40°C.

**Keywords:** *bacillus subtilis*, fermentation, inoculum, proximate analysis

### Introduction

*P. biglobosa* (African locust bean) tree is a perennial deciduous tree that grows from 7 to 20 meters high [1]. The tree was listed as one of the plants having real wound healing properties in South Western Nigeria [2]. The pods are flat and have irregular cluster of up to 30 seeds [1, 3-5]. African locust bean tree was named *P. biglobosa* by Robert Brown, a Scottish botanist in 1826 after Mongo Park, a Scottish surgeon who explored West Africa in 1790's. Mongo Park gave this tree a local name 'nitta' [3, 6, 7]. *P. biglobosa* seed is known as Iyere in Yoruba land while the fermented seed as Iru. Iru is one of the major sources of plant protein in African diet which is known as fermented vegetable protein [3, 8].

Fermentation is the chemical breakdown of substance by bacteria, yeast or other microorganism into alcohol, carbon dioxide or organic acids. A starter culture is a microbiological culture which performs fermentation [9].

Phenomenological model is regarded as a decision tool that assists decision makers in effectively dealing with complex issues such as fermentation and oil spillage on soil surfaces [10, 11].

Since fermented African locust bean seed is consumed for the high protein content and health benefits embedded in it. This study used the percentage protein composition which is the highest composition obtained when proximate analysis was carried out to discovered and optimize the number of days and the appropriate fermentation temperature.

## MATERIALS AND METHOD

**Source of Materials:** *Parkia biglobosa* seed were purchased from open market.

**Inoculum Preparation:** Inoculum used were freshly prepared in the Microbiology Laboratory Department in Covenant University using method [12-14].

**Preparation of Seed:** The raw African locust bean seed were processed using method [15]. Microbial fermentation 400 g of the processed seed were inoculated using freshly prepared *B. subtilis*. Fermentation was carried out for 5 days. Samples were taken every daily (24 hours) and kept in a freezer for further analysis.

**Proximate analysis:** The proximate analysis were evaluated by the method described by [16].

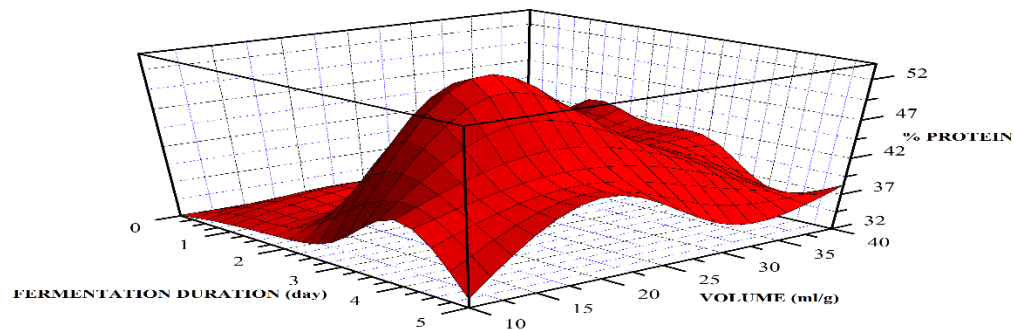
## RESULTS AND DISCUSSION

Assumption is that the experimentally measured % protein in 'Iru' is a complex function of the day, temperature and volume of inoculum used during fermentation i.e.:

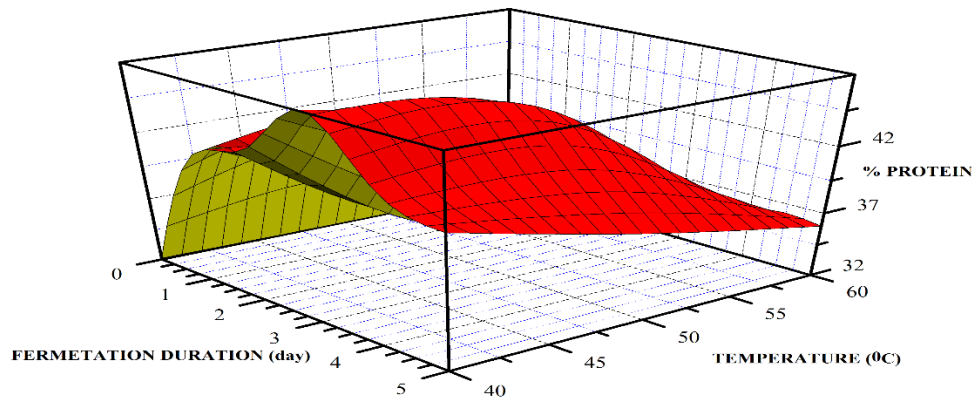
Let  $P$  = % protein, Day = fermentation duration (day); vol = volume of inoculum conc. (ml/g); temp = temperature ( $^{\circ}\text{C}$ )

$$P = f(\text{Day}, \text{vol}, \text{temp}) \quad [1]$$

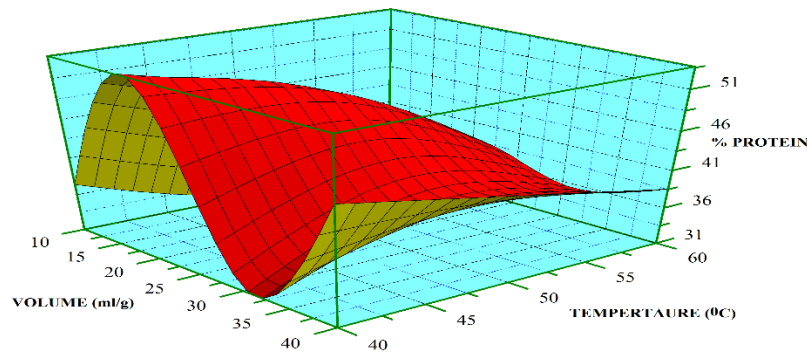
A 3-D plots of  $P$  vs. (Day, vol);  $P$  vs. (Day, temp) and  $P$  vs (vol, temp) are shown below:



**Fig. 1:** % Protein as function of Day and Volume



**Fig. 2:** % Protein as function of Day and Temperature



**Fig. 3:** % Protein as function of Volume and Temperature

Let us assume the model for equation [1] can be expressed in the form:

$$\begin{aligned}
 P = & P + A * Day + B * Day^2 + C * Day^3 + D * vol + E * vol^2 + F * vol^3 + G * temp + H \\
 & * Day * vol + K * Day * temp + M * vol * temp + N * Day * vol * temp + U \\
 & * temp^2
 \end{aligned}
 \quad [2]$$

Using equation [2] and the experimental data given, the unknown coefficients – P, A, B, C, D, E, F, G, H, K, M, N and U are correlated using Levenberg-Marquardt (or Powell) method ( using PSI software). The correlation coefficient was 0.885. The final values for the parameters are:

Variable Name	Initial Guess	Final Value
P	32.000	20.000
A	1.000	15.520
B	2.500	-2.649
C	4.500	0.197
D	5.000	2.593
E	713.000	-0.071
F	7.000	0.0007
G	-3.229	-0.323
H	-2.650	-0.165
K	-3.000	-0.110
M	4.000	-0.006
N	8.500	0.003
U	2.740	0.003

Note that from the results above, it is apparent that % protein is not dependent on vol<sup>3</sup> i.e. F = 0 is a reasonable assumption.

The equation generated above is now used in Excel Solver as an objective function to obtain values for the independent variables (Day, Vol, and Temp). In Excel, the objective function is:

$$= 20 + 15.52 * D2 - 2.649 * D2^2 + 0.197 * D2^3 + 2.593 * D3 - 0.0713 * D3^2 + 0.000667 * D3^3 - 0.3229 * D4 - 0.165 * D2 * D3 - 0.1103 * D2 * D4 - 0.00564 * D3 * D4 + 0.00284 * D2 * D3 * D4 + 0.00274 * D4^2$$

Several research works have been carried out on how fermentation enhances protein value of *Parkia biglobosa* seeds, this was established in figures 1-3 with increase in protein content at the third day of fermentation. The protein composition decreases as temperature increases. Figures 1-3 showed that at every fermentation temperature protein content increased from the first day up to the third day and declined from the fourth day. The % yield of protein from the fermentation of *P. biglobosa* at any given day of fermentation decreases with increase in fermentation temperature from 40 to 70 ° C. The protein yield at 40 ° C on the third day of fermentation is about 51%, it reduced to 32.6 % on the third day at a temperature of 70 ° C. This shows that *bacillus subtilis* functions well at lower temperature (40-50 ° C). Thus maximum yield of protein is achieved at the third day of fermentation with temperature 40 ° C.

The above 3-D plots shows the predicted effect of process variables on % Protein as the response. These plots represent graphically the regression coefficient in equation form in order to obtain the optimum conditions of the variables within the design region.

This work concluded that African locust bean should be fermented with *B. subtilis* for three days and at an optimum temperature of 40 °C. This is in agreement with the reports of [17, 18].

## Conclusion

This work clearly shows that a maximum yield of 51 % protein content was achieved through the fermentation of African locust bean using *B. Subtilis* at the optimum conditions of about 3 days of fermentation and 40 °C operating temperature using Levenberg-Marquardt correlation.

## Conflict of Interest

The authors declare no conflict of interest.

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