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Effect of *Bacillus sphaericus* on Proximate Composition of Soybean (*Glycine max*) for the Production of Soy Iru

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Abstracts: *Bacillus sphaericus* was isolated from naturally fermented soybeans (*Glycine max*) and used as a single starter for controlled laboratory fermentation for soy iru production at 35°C for 72 hours and the result of proximate analysis obtained before and after fermentation showed that, the moisture content of the boiled unfermented was 59.16% and increased slightly to 59.40% in naturally fermented soybeans and reduced to 49.99% in soybeans fermented with *B. sphaericus*. There was increase in percentage protein content from 24.50% to 31.24% after natural fermentation and 42.39% in control fermentation. The fat content increased from 17.12% to 19.90% after natural fermentation and 24.68% in controlled fermentation. There was increase in ash content from 0.97% to 1.72% and 9.58% of boiled unfermented, naturally fermented and controlled fermented sample. The fiber content reduced in fermented sample from 3.85% to 3.47%, and increased to 9.20% in controlled fermented sample while there was reduction in carbohydrate content from 26.30% to 22.86% in fermented and 18.12% in controlled fermented sample.

Key words: *Bacillus sphaericus*, *Glycine max*, proximate nutrient composition, soy iru

Introduction

Soybean (*Glycine max*) seed is rich in plant protein. Fermented soybean foods have been an intricate part of the oriental diet for millennia. The preparation of many indigenous or traditional fermented foods remains today a house hold art. Fermented soybeans food and condiments such as soysauce, miso and tempeh are manufactured on a commercial scale in the ancient as well as many Western countries (Larry, 1984)

In Northern Thailand, fermented soybeans, called "thua nao" have been produced and consumed locally for several decades which is produced by fermenting cooked soybeans naturally. Similar fermented soybean products have been described in several countries i.e. "Kinema" in India, "Schudouch" in China and "natto" in Japan. The best characterized fermented soybean product is probably natto" - The Japanese styled fermented soybeans. (Larry, 1984). Natto is commercially prepared using a pure starter culture of *B. substilis* natto strain.

The laboratory fermentation of soybeans for soy iru production using *Bacillus sphaericus* as a single starter culture needed further investigations on in knowing the differences between the proximate nutrient composition of soybean fermented with *B. sphaericus* and soybeans fermented naturally. Therefore, this present study aimed at fulfilling this objective.

Materials and Methods

Collection and preparation of material: Soybean seeds were purchased from Oja Oba main market Akure, Ondo State,. Nigeria. The seeds were boiled in water for 2 hours until very soft, it was then dehulled and boiled again for another 1 hour. 30g of soybean was then wrapped in plantain leaves and foil paper and was sterilized by autoclaving at 121°C for 15 minutes according to Larry 1984 technique.

Pure culture of Bacillus sphaericus was collected from microbiology laboratory of Food Science and Technology Department of Federal University of Technology, Akure, Nigeria. The starter cultures was isolated from naturally fermented soybeans (after fermenting the soybeans by boiling, dehulling and wrapping soybeans in plantain leaf without autoclaving and allowed to ferment naturally for 72 hours at 35°C).

Three ml - of cell suspension of *Bacillus sphaericus* was then added separately to the sterilized soybeans and were allowed to ferment at 35°C for 72 hours

Proximate analysis: Proximate analysis was carried out on the unfermented soybean, naturally fermented soybeans and soybeans fermented with the starter culture according to AOAC (1990) .

Results and Discussion

The result of soybeans obtained before fermentation (boiled, unfermented and soybeans fermented with *B. sphaericus*) and after fermenting naturally according to Table I showed that there was a slight increase in moisture content of naturally fermented sample from 59.16% in unfermented to 59.40% in naturally fermented soybean. There was reduction in the moisture content of soybeans fermented with *B. sphaericus* shown in Table 2. According to Njoku and Okemadu, 1989 who recorded high moisture content in his research and discovered that the high moisture content may be due to the fact that

Table 1: Proximate composition of boiled unfermented soybean and naturally fermented soybeans

Parameters	Unfermented Soybean (%)	Naturally Fermented Soybeans (%)
Moisture Content (wet basis)	59.16	59.40
Ash Content	0.97	1.72
Fat Content	17.12	19.90
Crude Protein Content	24.50	31.25
Crude Fiber Content	3.85	3.47
Carbohydrate Content	26.30	22.86

* mean of three determination

Table 2: Proximate Composition of Unfermented Soybean and Soybean Fermented with *Bacillus sphaericus*

Parameters	Unfermented Soybean (%)	Soybean Fermented with *B. sphaericus.*
Moisture Content (wet basis)	59.16	49.99
Ash Content	0.97	9.58
Fat Content	17.12	24.68
Crude Protein Content	24.50	42.39
Crude Fiber Content	3.85	9.20
Carbohydrate Content	26.30	18.12

* mean of three determination

seeds were mashed because during mashing, some little quality of water was added and that the result of moisture content obtained after fermenting with *B. sphaericus* compared favorably with the findings of Onifade and Jeff-Agboola (2003) who recorded reduction in moisture content of coconut fruit after fermenting with organisms.

According to result on Table 1 and 2. There was increase in percentage of Ash, fat, protein and fiber content after fermenting naturally and with starter culture. Ash content was highest in fermented sample with *B. spaericus* (9.58%) followed by naturally fermented sample (1.72%) and was lowest in boiled unfermented (0.97%). This result agrees with the result of the fermented sample of Mbajunwa (1995) and Enujiugha and Olagundoye (2001).

The fat content of the boiled unfermented soybeans (17.12%) falls within the range recorded by other workers (Achinewhy, 1983, Jonnes *et al.*, 1987. The increase in the percentage oil content of naturally fermented sample (19.90%) followed by increased in the sample fermented with *B. sphaericus* which is the highest (24.68%) agrees with the result obtained by Mbajunwa (1995) and Achinewhy (1983) who found the same thing and suggested that fermentation probably enhances oil extraction. And according to Akindumila and Glatz. 1998 who reported that certain microorganism could produce microbial oil during growth on substrate.

Protein content was highest in fermented sample with *B. sphaericus* (42.39%) followed by naturally fermented sample (31.25%) and the least recorded was in unfermented soybeans (24.50%). Microorganisms have been reported to increase the protein content of samples on which they grow (Raimbault, 1998 and Rodolfo *et al.*, 2000). Also, according to Popenoe 1969, protein increase could result from slight protein synthesis by proliferation of the microorganisms and a synthesis of enzyme proteins or from a rearrangement of the composition following the degradation of other constituents.

Crude fiber was high in soybeans fermented with *B. sphaericus* (9.2%) followed by unfermented soybeans (3.85%) and natural fermented soybean (3.47%) which agrees with the result of fermented sample from Mbajunwa (1995) and Enujiugha and Olagundoye (2001). Unfermented soybeans showed a carbohydrate content of 26.30%, followed by reduction in naturally fermented soybeans (22.86%) and soybeans fermented with *B. sphaericus* (18.12%). This may be due to possible transformation of carbohydrate to fat as reported by Akindumila and Glatz, 1998 which may explain the reason for the reduction of the carbohydrate content in the fermented sample.

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