THE EFFECT OF A SPICE ON THE NUTRITIONAL CHARACTERISTICS AND MICROBIAL POPULATION OF STORED SOYBEAN FLOUR

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Abstract

Objective: To study the effect of ethanolic extract of a spice Aframomum meleguata on the nutritional quality and microbial population of stored soybean flour have been carried out.

Method: The preliminary phytochemical analysis of the extract of A. meleguata shows the presence of tannins, polyphenols and saponins. The pH, moisture, fat, crude protein and microbial load were evaluated in soybean flour treated with extract of A. meleguata at different concentrations of 1.0mg/10gm w/w, 0.5mg/10gm w/w and 0.1mg/10gm w/w for a period of 60 days. All the parameters listed above were investigated on weekly basis.

Results: Results showed that the pH values of the control and treated samples changed significantly at different concentrations (P<0.05), for bacterial population there was insignificant effect of the extract (P>0.05) while that of fungi was significant (P<0.05). The moisture and fat content occurred within the same range for both treated and the control as they changed significantly with concentrations (P>0.05). Variation in crude protein with concentrations was significant (P<0.05).

Conclusion: Comparing the results of the extract with the commercial preservative, sodium benzoate, for all the parameters tested, extract of A. meleguata performed better in the keeping quality of stored soybean flour and should be preferred than commercial preservative. However, this is a preliminary investigation, more work should be done to isolate pure component from this spice and compared with any known commercial preservative.

Keywords: Aframomum meleguata, Microbial population, Soybean flour.

Introduction

Soybean has been consumed in several ways in various parts of the world. There is a growing awareness in the production and utilization of soybean in Nigeria within the last decade. An expanding market is due largely to the popularity of soybean as a miracle crop to address problems of malnutrition. It is widely used for the preparation of “dawadawa”– a fermented paste used as a food condiment. There is also growing interest in the preparation of soy moi-moi, soy akara, soy cheese, and incorporation of soybean in soups and stews1. Soybean has been incorporated into local Nigerian dishes in order to improve their nutritional value 2,3. Furthermore, the utilization of
soymilk in the treatment of infant protein-
malnutrition (kwashiorkor) has been
reported\(^1\). Soymilk has been the most
largely consumed soy product, and in
Nigeria, it is currently being produced at
household level. Militating against these
promising usages of the miracle crop is the
keeping quality of the products, which
need to be quarantined in order to ascertain
success in the struggle to alleviate problem
of malnutrition. Local methods are in use
through spice application.

Spices have been extensively used
in the medical field. They have been used
in curing diseases and healing of wounds.
Some work has been done using different
spices to inhibit the growth of bacteria.
Cumin inhibited bacterial growth\(^4\).
Components of \textit{A. melegueta} have been
found to be antimicrobial\(^5,6\).

This work therefore aims at
investigating the shelf-life of treated
soybean flour, since there has been
growing concern over the use of
commercial preservatives because of their
potential carcinogenic properties; the use
of natural preservative is therefore
receiving increasing attention.

Materials and Methods

Collection of Materials

Five hundred grams of soybean
seeds (\textit{Glycine max} (L) marr) (TGX 536-
02D) a variety from IITA, Ibadan, were
collected from the crop production
department in the School of Agricultural
Engineering, Federal University of
Technology, Minna. The seeds were kept
under dry conditions in Laboratory cabinet
for further use.

The spice used for preservation
\textit{Aframomum melegueta} “grains of
paradise” was purchased in dried form
from Minna main market and authenticated by Dr. Tsado of crop
production Department, Federal University
of Technology, Minna.

Preparation of soybean flour

This was carried out following the
method of konan and Agbo\(^7\). Two litres of
distilled water was added to 500g of
cleaned soybean seeds in a 5 liter bowl and
then the water was drained off and washed
seeds were oven dried at 55\(^0\)C overnight.
After drying, these were ground into fine
powder using a cleaned and dried local
milling machine. The powdered soybean
was sieved using a home sieve and kept in
a sterile container for subsequent use
(Flow chart 1).

Cleaned soybean

i. Addition of water

ii. Drainage of water

Washed, cleaned soybean seeds.

i. Oven-dried at 53\(^0\)C

overnight

Dried soybean seeds

i. Milling

ii. Sieving

Soybean flour

Flow chart 1: Preparation of Soybean
flour.

Preparation of Extract of Spice

The dried spice \textit{A. melegueta} was
crushed to powdered form by the use of an
electric blender (Mx-391N). Ethanol
(95\%) was employed for the purpose of
extraction. 400ml of ethanol was added to
100g of the powdered spice in a two liter
conical flask and kept in the refrigerator
4\(^{0}\)C for 7 days with occasional shaking. The mixture was then filtered using Whatman filter paper No.1. A rotary evaporator was used in Vacuum at 40\(^{0}\)C to concentrate the extract. A brown semi-solid substance of “grains of paradise” were obtained and kept in MaCartney bottles for further use.

**Phytochemical analysis of “grains of paradise”:**

Preliminary phytochemical analysis was carried out using the method described by Sofowora\(^{6}\) and Fadeyi and Akpan\(^{8}\). The spice was screened for the presence of sesquiterpenes, saponins, phenolic compounds or tannins and anthraquinones glycosides.

**Treatment of Soybean flour with extract of A. meleguata:**

30mg of a spice A. melegueta was dissolved in 3ml of acetone to obtain a stock solution of concentration 10mg/ml. To 100g of soy flour 1ml of this stock solution was added to get a concentration of 15g/10gm soy flour and mixed vigorously using a sterile spoon. The treated soy flour was dispensed in polythene bags (triplicates) each containing 10g and sealed using an electrical impulse sealer. Similarly, 2ml of acetone was added to the remaining 2ml of extract stock solution (Solution A; conc. 5mg/ml). 1ml of this solution A was added to another 100g of soy flour (concentration 0.5mg/10mg) and was mixed with a sterile spoon. The treated samples were distributed into polythene bags and sealed as explained above. 0.2ml of stock Solution A was added to 100g of soy flour (concentration 0.1mg/10mg and treated the same way as described above. Sealed samples were stored at room temperature (28±2\(^{0}\)C) for 60 days. Control samples were also set up. On weekly basis triplicate samples were analysed for Microbiological and Nutritrional qualities.

**pH Determination**

A pH meter (MicropH 3310 Crison) was used to determine the pH of treated and untreated samples. 1g of each of treated and untreated samples was added to 10ml of distilled water and after vigorous shaking the pH was measured.

**Isolation, Identification and Enumeration of Bacterial and Fungal Population:**

The morphological characteristics of bacterial isolates were studied by growing the cultures on nutrient agar. Identification was based on colonial morphology and biochemical tests as described by Collins and Patricia\(^{9}\). For fungal isolates morphological characteristics of the isolates were studied by growing the cultures on potatoes dextrose agar as described by Smith\(^{10}\). For enumeration, serial dilution up to 10\(^{-8}\) were made following the method of Fawole and Oso\(^{1}\).

**Determination of (%) Moisture:**

Moisture content (%) was determined using an electric protimeter grain master 2000 following the instructions in the operating manual.

**Fat/Lipid Determination:**

The fat content (%) of treated and control samples were determined by direct soxhlet extraction using petroleum ether as solvent.

**Determination of Crude Protein:**
The protein determination was carried out using the micro kjeldahl method Bradstreet.  

**Result**

The preliminary phytochemical analysis of extracts of *A. melegueta* shows the presence of polyphenols, tannins and saponins. A pH change of within a narrow range was observed with the addition of the extract, as the control changes between 6-7 that of treated samples remains within a range of 7 (Fig.1). The moisture content was low in the treated soybean flour, the level of moisture dropped over time in the treated samples (Fig.2). In fig. 3, the fat content of extract treated samples showed no significant variation (P>0.05) as it ranged mostly between a narrow limit of 10-20%. The crude protein of treated samples declined over time (Fig.4). The microbial load in terms of bacteria population was highest in the control and significantly (P<0.05) lowest in the treated samples (Fig. 5).

**Discussion**

A pH change of within a narrow range was observed with the addition of the extract, this may be attributed to the deterioration of the control as a result of enzymatic activities of microorganisms. The higher concentrations of 0.5mg/10gm and 1.0mg/10gm showed no significant decline. This showed the stability of the soybean flour at this concentration. The microbial load in terms of bacteria population was highest in the control and significantly (P<0.05) lowest in the treated samples. This probably may be to the bioactive components of the natural preservative which is known to contain series of pungent compounds that are bactericidal Oloke and Kolawole.  

Similarly, microbial load in terms of fungal population shows no significant variation (P<0.05) with respect to extract treated samples. This was in agreement with Oloke and Kolawole who observed the antifungal effectiveness of the components of the spice.

**Conclusion**

From the results obtained, it is concluded that soybean flour can be stored for a period of two months or longer at room temperature using 1.0mg/10gmw/w concentration of the extract of *A. melegueta*. However, more research need to be carried out to isolate the bioactive component of the spice and this should be compared with any known commercial preservative.

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Fig. 2: Moisture content (%) of soybean flour treated with extract of *A. melegueta*

Fig. 3: Fat (%) of soybean flour treated with extract of *A. melegueta*
Fig. 4: Crude Protein (%) of soybean flour treated with extract of *A. melegueta*

Fig. 5: Bacterial Population of soybean flour treated with extract of *A. melegueta*