

Nutritional and Physicochemical Properties of Thai Traditional Fermented Soybeans (Thua Nao) Produced by Ethnic's Conventional Method in Northern Thailand

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ABSTRACT:

The content of total phenolics, DPPH-free radical scavenging effect, total antioxidant activities, and nutritional properties of Thua Nao (TN), a Thai traditional fermented soybean, collected from four ethnic's conventional methods in Northern Thailand were investigated. In general, the samples contained 6.18–7.04% moisture, 3.24–3.78% ash, 47.43–49.50% protein, and 4.16–4.88% fat. The total phenolic contents of methanol extracts of the various TN, varying on the source of sampling, range between 31.45 and 39.54 (mg/L of gallic acid). For antioxidant activity, the IC₅₀ values as determined by the DPPH method ranged from 22.52 to 2.81 mg/mL of sample extract. There may be variations between these properties. Because of the variety of soybeans, the fermentation method, and starter microorganism species. Our study provides detailed information regarding the nutritive quality of TN. The commodity tends to be a decent source of protein-based on protein content and a potent in the antioxidant diet with high phytochemical antioxidant content and powerful antioxidant activity.

Keywords: Fermented Soybeans, Nutritional Properties, Physicochemical Properties

INTRODUCTION

Thuanao(TN) is a popularly eaten traditional fermented soybean in northern Thailand. For decades, the pride of culinary cultures has been. TN, conceived by local wisdom from generation to generation, is considered a low-cost meat substitute and can be applied in various uses.¹In many local dishes, TN can be eaten directly as a staple food and/or as a taste enhancer. Traditional TN fermentation relies on microbial activity, especially, that, of, the, Bacillus, group.^{2,3}Proteolytic activity is a key, step in the fermenting process. During, the, fermentation, soy proteins are hydrolyzed into peptides, amino acids, and, ammonia, resulting, in, an alkaline, condition, that, would, help, prevent contamination of spoilage microbes.³At present, most alkaline fermented foods are made, using, mixed, microbial, cultures, at, the, household, stage. No specific inoculum is required.³During, TN, fermentation, several, biochemical changes occur, as stated. It is hypothesized that a major activity is probably protein hydrolysis. Microbial

proteases help speed up the breakdown of proteins into different peptides and amino acids. Moreover, there is a conversion by these microflora of isoflavones abundant in soybeans. The biochemical mechanisms of these fermented soybean products are identical, although it should be noted that, due to: (1) soybean variety; (2) fermentation conditions; and (3) indigenous microflora, the difference in nutritional quality can be observed.³

A number of amino acids tend to include in TN products.⁴ Accessible data indicate that all amino acids with large quantities of total free amino acids ranging from 11.03 g/kg to 61.23 g/kg are found in TN, as dry basis. The major amino acids are tryptophan, followed by glutamic acid, cysteine, lysine, and leucine.⁴ In addition, when using a pure starter culture of *B. subtilis* TN51, an increase of total free amino acids, including essential amino acids, are found.⁴ It is also possible to detect a large increase in hydrophobic amino acids and a decrease in the amount of charged amino acids, which is possible through the process of bacterial biotransformation.^{3, 4} TN products have been assessed obtained in aglycone isoflavone.^{5,6} Isoflavone compounds are interested in Aglycone because clinical trials indicate that they can be absorbed more easily and in higher concentrations than glucosylated equivalents.⁵

The purpose of this study was to evaluate the nutritional and physicochemical properties of Thai traditional fermented soybeans (Thuanao: TN) produced by ethnics' conventional method in Northern Thailand.

MATERIALS AND METHODS

Raw materials and sample preparation

Fresh TN products used in this study were collected from 4 local ethnics villages, two villages from Mae Cham district and another 2 from Hod district. Ching Mai Province, Thailand. The samples once collected were transported to the laboratory in portable coolers and stored at -20°C until used.

Samples extraction

According to the method of Dajantaet al.⁴, briefly, the ground powder of the lyophilized sample (30 g) was extracted with 300 ml of 80% (v/v) methanol for 24 h in room temperature with continuous shaking. The extracts were filtered through Whatman No.1 paper, concentrated under vacuum at 40°C and freeze-dried. The lyophilized extracts were stored at -20°C, and before measuring the content of total phenolic compounds, antioxidant and antimicrobial activities, the extracts were dissolved in methanol.

Proximate analysis

The proximate analysis was determined according to AOAC official methods.⁷ The crude protein content ($N \times 6.25$) was estimated using the macro Kjeldahl method, the crude fat was determined by extraction using Soxhlet apparatus, moisture contents were analyzed by hot air

oven method at 100°C and the ash content was analyzed by weight before and after incineration at 600 ± 15 for 24 hours.

Total Phenolics Content

Total phenolic content (TPC) of the CPEs were assessed using the Folin–Ciocalteu procedure as modified by Kahkonen et al.⁸ Briefly, 200L of crude extract solution was mixed with 1mL of Folin–Ciocalteu reagent, and then 0.8mL sodium carbonate (7.5%) was added. The mixture was incubated at room temperature for 30 minutes and was measured at 750 nm using a spectrophotometer. The results were expressed as mg/L of gallic acid.

Determination of antioxidant activity: DPPH assay

Scavenging activity on DPPH was assessed according to the method as modified by Blois⁹. Briefly, the mixture of 10L of different concentrations of the extracts and 190L of 80M DPPH in methanol was shaken and incubated at room temperature in the dark for 30 minutes. The control was prepared without extract. Absorbance at 517nm was measured with a spectrophotometer. The experiment was performed three times, each time in triplicate. The percent inhibition was calculated from control using the following equation:

$$\% \text{ inhibition} = [(A_{517} \text{ control} - A_{517} \text{ sample}) / A_{517} \text{ control}] \times 100$$

where A₅₁₇ control and A₅₁₇ sample were the absorbance values at 517 nm of DPPH with sample and DPPH of control, respectively.

The half-maximal inhibitory concentration (IC₅₀) was then calculated from the equation analyzed from the logarithmic regression curve between soybean extract concentration (mg/ml) and scavenging activity¹⁰

Statistical analysis

Data were expressed as means ± standard deviation of triplicate observations. The data were also subjected to analysis of variance (ANOVA) and Duncan's multiple range tests. P < 0.05 was considered statistically significant.

RESULTS AND DISCUSSION

Proximate analysis

It was found that fresh TN contained moisture, ash, fat, and protein at ~6.18–7.04%, 3.24–3.78%, 4.16–4.88%, and 47.43–49.50%, respectively, as shown in Table 1.

Table 1 Nutritional Properties of TN.

	Nutrients			
	Moisture (%)	Ash (%)	Fat (%)	Protein (%)
TN1	6.41	3.24	4.88	47.43

TN2	7.04	3.45	4.52	48.27
TN3	6.18	3.78	4.16	49.50
TN4	6.52	3.62	4.63	49.44

TN1 = Thuanuo from villages 1 in Mae Cham district, TN 2 = Thuanuo from villages 2 in Mae Cham district, TN 3 = Thuanuo from villages 3 in Hod district, TN 4 = Thuanuo from villages 4 in Hod district.

Total phenolics content and antioxidant activity

The total phenolic contents of methanol extracts of the various TN, varying on the source of sampling, range between 31.45 and 39.54 (mg/L of gallic acid). TN1 displayed significantly the highest total phenolic contents among the TN collected.

Based on the DPPH radical scavenging assay. The wide variations of inhibiting free radicals were found in different fresh TN. The IC₅₀ of fresh TN extracts ranged from 2.52 to 2.81 mg/ml of sample extract. TN3 displayed significantly the highest inhibition among the TN collected as shown in Table 2.

Table 2Total phenolics content and antioxidant activity of TN

	Total phenolic contents (mg/L of gallic acid)	Antioxidant activity DPPH (IC ₅₀ g/ml)
TN1	39.54 ± 1.06	2.77 ± 0.04
TN2	31.45 ± 1.96	2.52 ± 0.09
TN3	38.55 ± 1.12	2.81 ± 0.02
TN4	33.01 ± 1.76	2.74 ± 0.07

TN1 = Thuanuo from villages 1 in Mae Cham district, TN 2 = Thuanuo from villages 2 in Mae Cham district, TN 3 = Thuanuo from villages 3 in Hod district, TN 4 = Thuanuo from villages 4 in Hod district, Data are mean ± standard deviation.

DISCUSSION

It should be noted that the characteristics of fresh TN were that only a slightly sticky material could be identified and that the visible dark brown color of the TN collected from 4 markets tended to vary widely on the same line with Dajanta et al.⁴ This may be due to the results of the disparity between processes of production, including a range of soybeans, boiling time of cooked soybean, fermentation conditions and period of soy incubation. In addition, fresh TN has been shown to have a significant difference in chemical compositions, including moisture, ash, fat, and protein. In general, the results obtained were similar to previous investigations.^{4, 11, 12} High protein and fat content has been found in fresh TN, protein, and fat are the main components of soybean.³ Therefore, the enzymatic degradation of soy proteins is definitely a key process in the fermentation of TN.³ Essential unsaturated fatty acids including linoleic (an omega-6 fatty acid) and linolenic (an omega-3 fatty acid) acids are also major components in soybean,⁴ abundance in essential unsaturated fatty acids cause

a low level of saturated fatty acids and absence of cholesterol,^{13, 14} soybean protein and fat can also be considered a healthy diet.

Phenolic compounds are one of the main classes of compounds that act as key antioxidants in soybeans.¹⁵ The total phenolic contents of fresh TN in this study, varying on the source of sampling, range between 31.45 and 39.54 mg/L of gallic acid. These values are relatively consistent with the findings of Lin et al.¹⁶ and Dajanta et al.⁴ which were ranged from 23.70 to 45.72 and 30.46 and 44.58 mg/L of gallic acid. The level of phenolic and isoflavone compounds associated significantly with higher antioxidant activity.¹⁷ The high phenolic content was expected to be responsible for the greater free radical scavenging effects and the overall antioxidant activity of TN in the current study. Besides antioxidant phytochemicals isoflavones and phenolics, other components including oligoproteins, free amino acids, and melanoidins involved in fermented soybeans have been reported to support the antioxidant effect.¹⁸

In conclusion, nutritional value, total polyphenol content, and antioxidant activity were calculated from 4 separate TN sources. The 4 extracts display slightly distinct compositions of nutrients. The highest TPC and antioxidant activity were at TN3. Finally, different sources and methods of preparation are essential for its composition and potential viability. TN includes many ingredients, TN is a strong source of protein with a high content of antioxidants phytochemicals, and antioxidant activities based on the protein content and potent to antioxidant. The data available from this study is important as standard values for further work on the development of TN nutritive quality.

CONCLUSIONS

It is important to understand the TN composition data from the point of view of a nutritionist. It offers useful data on the nutritional value of local food items. Moreover, these data can be used as a dietary standard or as the basic recommendation for the health policy of the Country. Earlier studies, including this study, have confirmed the chemical composition of the TN. The development of the product can also be improved in the expectation that the nutritional quality will be improved. TN appears to be a good protein source based on protein content and potent antioxidant diet food with high levels of antioxidants phytochemicals and strong antioxidant activity.

CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

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