

PHYSICO-CHEMICAL CHARACTERISTICS OF TRIBAL BEAN (*Canavalia virosa*) AND ITS ALTERNATIVE TOFU AND TEMPEH FOOD PRODUCTS

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Submitted 22 March 2010; Accepted 6 August 2010

ABSTRACT

Increasing price of soybean becomes a serious problem for producers of traditional foods such as tempeh and tofu. These traditional foods are important protein sources for many Indonesian people. Tribal bean (*Canavalia virosa*) could be used as a substitution of soybean for tempeh and tofu processing. This study aimed to determine physico-chemical characteristics of tribal bean and its products such as tofu and tempeh. Tribal bean old pods were peeled manually in the Postharvest and Agricultural Machinery Laboratory of the Yogyakarta AIAT. The peeled seeds were dried until 10% water content and their epidermis were removed mechanically by using an abrasive peeler to produce yellowish clean peeled beans. The beans were analyzed physically and chemically using the standard procedure. Since the tribal bean seeds contained high HCN, to minimize HCN content the beans were presoaked for 48 hours in water. The beans were then mixed with soybean at a ratio of 50:50 or 25:75 and processed for making tempeh and tofu using traditional method. Physico-chemical and organoleptic characteristics of the tribal bean tempeh and tofu were analysed, involving organoleptic test with hedonic method, texture, as well as water, ash, protein and crude fiber contents. The results showed that tribal bean contained protein (37.30%), essential amino acids, minerals and fiber (3.1%), and a toxic substance HCN. Presoaking the beans in water for 48 hours significantly reduced HCN content by 98.51%, from 1334 ppm. Tofu made of a mixture of tribal bean and soybean at a ratio of 25:75 plus 2% rice vinegar as a coagulant has a white color and normal flavor appearances, and was accepted by panelists. The tribal bean tempeh contained 78.1% water, 1.21% ash, 8.14% protein, 3.1% crude fiber, and 44 ppm HCN. Tempeh made of a mixture of tribal bean and soybean at ratios of 50:50 and 25:75 showed good characters (flavor, taste, color, and texture) and panelist acceptance, as well as nutrition values (76% water, 2.71% ash, 14% protein, 0.25% crude fiber, and 14% lipid). However, HCN content in the tofu was still higher (85 ppm HCN) than the recommended maximum value of 50 ppm. This study suggests that tribal bean is more suitable for tempeh than for tofu based on its HCN content.

[**Keywords:** *Canavalia virosa*, tempeh, tofu, physico-chemical characteristics]

INTRODUCTION

Protein deficiency is a major problems in poor nutrition status of Indonesian population. Animal protein

sources such as meat, eggs, milk, and fish are relatively expensive, therefore their consumption were generally low (Setiawan 2005). Traditional foods made of soybean, such as tempeh and tofu, become an important source of proteins for low income Indonesian people. However, price of soybean is high recently, therefore it cannot be afforded by many. On the other hand, Indonesia is rich in protein source crops such as tribal bean (*Canavalia ensiformis*) that can be made to various traditional rich in protein foods, such as tempeh and tofu.

Tribal bean is wildy grown along the coastal areas of about 3000 ha in Bantul, Kulon Progo, and Gunungkidul Districts of Yogyakarta Province. The plant is locally known as *kerandang*. The plant produces pods and seeds. The green young pod is usually consumed by local communities as a vegetable, while the old bean is used for making tempeh (Djaafar and Purwaningsih 2009).

Tribal bean belongs to a Leguminosae family, having 10-15 cm long pods, containing 4-8 brown bean seeds per pod (Mukhopadhyay *et al.* 1985). The tribal bean plant has good adaptation in poor soils, such as coastal sand. The plant is flowering throughout the year. Tribal bean roots are rich in root nodules which bind nitrogen from the air so it can nourish the soil. The plant is likely adaptive to high temperature of the tropic, grows well in the dry season, and apparently resistant to diseases and rodents.

Tribal bean contains 31.3% protein and 4.9% fat with a caloric value of 1512.4 kJ/100 g. Tribal bean also contains various essential amino acids, including isoleucine, lysine, histidine, sistine, methionine and threonine, as well as antioxidant especially isoflavon that can be useful for humans (Snyder and Kwon 1987; Siddhuraju and Becker 2001). Tribal bean also contains anti-nutritional substances such as tannins, HCN, and phenolic compounds (Sridhar and Seena 2005). The presence of anti-nutrition compounds in tribal beans is not a real problem because these com-

pounds can be reduced even be lost during processing such as washing, soaking, and heating.

The protein content of tribal bean is equivalent to soybean, therefore tribal bean can be used as a substitution to soybean in making tempeh and tofu. The study aimed to determine the physico-chemical characteristics of tribal bean and its products, like tempeh and tofu as an alternative food.

MATERIALS AND METHODS

Materials

Tribal bean pods were harvested from wildy grown plants in the coastal areas of Bugel Village, Panjatan District, Kulon Progo Regency, Yogyakarta. The other materials were yeast for making tempeh and coagulants for making tofu, such as rice vinegar (from market), lactic acid 98% (technical brand), and pineapple extracts. Soybean of Wilis variety was obtained from market. Rice vinegar is a solution of fermented rice.

Methods

The research consisted of four activities, such as tribal bean preparation and its physico-chemical analysis, reducing HCN content in the seeds, processing of tribal bean seeds for tempeh and its physico-chemical analysis, and processing of tribal bean seeds for tofu and its physico-chemical analysis.

Tribal bean preparation and its physico-chemical properties

The old brown pods of tribal bean were peeled manually in the Postharvest and Agricultural Machinery Laboratory of the Yogyakarta Assessment Institute for Agricultural Technology (Yogyakarta AIAT). The peeled beans were dried until 10% water content and their epidermis were further removed mechanically by using an abrasive peeler to produce yellowish clean peeled beans (Fig. 1). Physico-chemical characteristics of the beans were analyzed, including physical length and width, 100 grain weight, density and color of the seeds, water content (gravimetry), ash (Muffle furnace), protein (Kjeldhal), lipid (Soxhlet), minerals (HPLC), and amino acids (HPLC) (AOAC 1990). The analysis was conducted in the Postharvest and Agricultural Machinery Laboratory of Yogyakarta



Fig. 1. Tribal bean seeds as raw materials for making tempeh and tofu: (a) old pods, (b) tribal bean seeds, (c) peeling the seed epidermis, (d) peeled tribal beans.

AIAT and Chemical Laboratory of Agricultural Technology Faculty, Gadjah Mada University, Yogyakarta. The data were statistically analyzed using one-way ANOVA (Steel and Torrie 1993).

Reducing HCN content in the seeds

The tribal bean contained ample HCN of about 1134 ppm (Djaafar *et al.* 2009), therefore the beans were processed to minimize HCN content before processed for making tempeh and tofu. Since there is no standard protocol for minimizing the tribal bean yet, various treatments were applied, such as soaking the beans in water for 18 hour; soaking the beans for 18 hours followed with drying seed until bean water content of about 10%; soaking the beans for 24 hours followed with boiling for 25 minutes; and soaking the beans for 48 hours. Analysis of HCN content in the treated beans was conducted using a spectrophotometry method.

The tempeh processing

Yellow, clean, peeled tribal bean seeds were obtained as described previously. The beans were pre soaked

to minimize HCN content as described above. The presoaked beans were then processed for tempeh using a traditional method (Purwaningsih 2008). The peeled beans were washed, soaked in water for 24 hours and boiled for 45 minutes. The boiled beans were then washed and soaked again for 24 hours. Finally, the beans were washed and steamed for 45 minutes. After chilling, the beans were inoculated with tempeh yeast, packed in plastic bags and fermented for one day at room temperature (27-30°C).

To obtain the optimal ratio of tribal bean and soybean, the tribal beans were mixed with soybeans at different ratios such as 25:75, 50:50, 75:25, and 100:0. The experiment was designed in a complete random with four replications (Scheaffer *et al.* 1990). Soybean var. Wilis was obtained from a local market.

Organoleptic test of the tribal bean tempeh was carried out using a hedonic scale method by 20 semi-trained panelists. Parameters observed were texture, flavor, color, taste, and overall preference. Five levels of hedonic scale were used, namely 1 (strongly dislike), 2 (dislike), 3 (neutral), 4 (like), and 5 (strongly like) (Resurreccion 1998). Organoleptic test data were analyzed statistically by using one-way ANOVA (Steel and Torrie 1993). Chemical analysis of the tribal bean tempeh included water content (gravimetry), ash (Muffle furnace), protein (Kjeldhal), crude fiber (filtering method), and HCN (spectrophotometry) (AOAC 1990).

The tofu processing

Peeled tribal bean seeds used for making tofu were prepared as the same for tempeh as described above. The experiment was arranged in a completely randomized design with two factors and two replications. The first factor was ratio of the tribal bean and soybean, and the second factor was types of coagulant (rice vinegar, lactic acid, and pineapple extract). The ratio of tribal bean to soybean were: (1) 50:50 plus 2% rice vinegar; (2) 25:75 plus 2% rice vinegar; (3) 50:50 plus 2% lactic acid; (4) 25:75 plus 2% lactic acid; (5) 50:50 plus pineapple extract; and (6) 25:75 plus pineapple extract.

The peeled tribal bean seeds were washed and soaked in water for 48 hours. Volume of water used was ten times of the seed weight. Every 6 hours, the soaking water was replaced with new one. Soybean seeds were washed and soaked in water for 5 hours without any change of water replacement. The tribal bean and soybean seeds were mixed at the ratio as mentioned above, and blended for 2 minutes by using

blender. Water was added into the blender as much as eighth times of the seed weight. Blended tribal bean and soybean milk was filtered using filter cloth and boiled at a temperature of 90°C until completely boiled. Coagulant was mixed slowly with the milk at temperature of 80°C. After completely mixed, the milk-coagulant was slowly transformed into curds and whey. The whey was ladled out into a tank and the curds were ladled roughly into a cloth-lined perforated wooden box. The box was then closed with a wooden lid. The curds were pressed under a slatted lid with 20 kg stone for 15 minutes (Sulistiyani 2009).

Organoleptic tests of the tribal bean tofu was carried out using a hedonic scale method such as color, texture, flavor, taste, and overall preference by 20 semi-trained panelists. Five levels of hedonic scale (1 to 5) were used, namely: 1 (strongly dislike), 2 (dislike), 3 (neutral), 4 (like), and 5 (strongly like) (Resurreccion 1998). Organoleptic data were analyzed statistically by using one-way ANOVA (Steel and Torrie 1993).

Physical characteristics of the tribal bean tofu assessed were rendement, color, flavor, and plasticity. Color and smell were also observed by panelists. Plasticity of the tribal bean tofu was measured by using a penetrometer. Acidity of the whey was measured by using a pH-meter. Chemical characteristics of the tribal bean tofu included standard protocols such as water content (gravimetry method), ash (Muffle furnace), protein (Kjeldahl), lipid (Soxhlet), crude fiber (filtering method), and HCN (spectrophotometry) (AOAC 1990).

RESULTS AND DISCUSSION

Physico-chemical Characteristics of Tribal Bean

Physical and chemical characteristics of tribal bean seeds were presented in Table 1 and 2. The tribal bean seeds are rich in protein and minerals. However, the bean seeds also contain very high HCN (1134 ppm), above the tolerance threshold for human body of 50 ppm kg⁻¹ body weight (Buckle *et al.* 1987). Therefore, the seeds must be treated before processed to minimize its HCN content.

The tribal bean seeds contains 37% protein, equivalent to soybean protein of 39% (Sitompul 2004; Anonim 2009). Important minerals found in the tribal bean were calcium, manganese, sodium, potassium, and phosphorous. The tribal bean also contains various essential amino acids similar to those found in soybean, but in lower quantities (Table 2).

Table 1. Physycal and chemical characteristics of tribal bean.

Component	Total
Physical character	
Seed length (mm)	14.27
Seed width (mm)	9.81
Length-width ratio	1.46
100 grain weight (g)	45.95
Seed density (g l ⁻¹)	603.49
Seed coat color	Brown
Chemical character	
Water (%)	12.70
Ash (%)	2.84
Protein (%)	37.30
Lipid (%)	1.31
Crude fiber (%)	1.83
Calcium (ppm)	1819
Iron (ppm)	94
Manganese (ppm)	25
Sodium (ppm)	893
Potassium (ppm)	22,159
Phosphorus (ppm)	2,487
Zinc (ppm)	44
HCN (ppm)	1134

Table 2. Amino acid content of tribal bean and soybean.

Amino acid component	Tribal bean (%)	Soybean ¹ (%)
Aspartate	1.17	4.10
Glutamate	2.75	4.40
Serine	0.70	1.40
Glycine	0.22	2.00
Histidine	0.27	0.90
Arginine	0.86	0.63
Threonine	0.31	2.00
Alanine	0.75	2.10
Proline	0.82	1.67
Tyrosine	0.35	1.75
Valine	0.73	3.10
Methionine	0.29	2.15
Cystine	0.13	0.49
Isoleucine	0.37	0.93
Leucine	1.78	2.93
Phenylalanine	0.29	2.10
Lysine	1.12	1.90

¹Source: Sitompul (2004).

Reduction of HCN Content in Tribal Bean

Presoaking tribal bean seeds in water reduced HCN content significantly. The best presoaking period was for 48 hours which very significantly (98.51%) decreased the HCN content from 1134 ppm to 16.84 ppm (Table 3).

The maximum threshold level of HCN tolerance for the human body is 50 ppm kg⁻¹ body weight (Buckle

et al. 1987); exceed this level, HCN will interfere oxygen transport in the blood tissue resulting in breathing difficulties. HCN can also poison body leading to death. HCN poisoning is characterized by symptoms, such as nausea, diarrhoea, shortness of breath, dizziness, weakness to coma (Manik 2003).

Organoleptic and Chemical Characteristic of Tribal Bean Tempeh

Panelists preferred more tribal bean tempeh made of a mixture of the tribal bean and soybean at a ratio of 75:25 which means that tribal bean can substitute soybean as many as 75% (Table 4). However, panelists dislike tribal bean tempeh from the other combination ratios due to its unpleasant flavor.

The tribal bean tempeh had 78.10% moisture content and is rich in important nutritions such as ash 1.21%, protein 8.14%, and crude fiber 3.10% (Table 5). HCN

Table 3. HCN content of tribal bean seeds trated with various soaking treatment.

Soaking treatment	HCN content (ppm)
Without soaking	1134
Soaking for 18 hours	378
Soaking for 18 hours and dried	394
Soaking for 24 hours and boiled for 25 minutes	33
Soaking for 48 hours	17

Table 4. Preference score of panelists on tempeh made of a mixture of tribal bean and soybean (1-5 scoring).

Ratio of tribal bean and soybean	Organoleptic characteristic				Overall preference
	Color	Texture	Flavor	Taste	
100:0	3.9	3.0	3.0	3.0	3.0
75:25	3.8	3.0	3.0	3.2	3.5
50:50	4.1	3.9	3.5	3.7	3.7
25:75	3.3	4.0	3.8	3.8	3.9

Table 5. Chemical composition of tempeh made of a mixture of tribal bean and soybean.

Ratio of tribal bean and soybean	Component				
	Water (%)	Ash (%)	Protein (%)	Crude fiber (%)	HCN (ppm)
100:0	78.10	1.21	8.14	3.10	44
75:25	72.71	0.45	9.50	5.32	27
50:50	67.71	0.79	14.15	8.73	18
25:75	58.50	1.15	20.58	10.53	8
0:100	55.30	1.60	20.80	4.20	0

content in the tribal bean tempeh ranged from 7.69 to 43.80 ppm, below the maximum safety level of 50 ppm (Buckle *et al.* 1987). The soaking and boiling processes seemed to be effective in reducing HCN content significantly. HCN content of the tribal bean tempeh was lower than that found in other traditional tempeh called mucuna tempeh (97 ppm) (Medikasari 2007). This shows that consumption of tribal bean tempeh will safe if it is consumed about 2.725 kg in a day by a normal adult body weight of 50 kg. The body has several mechanisms to effectively detoxify cyanide. Most of cyanide reacts with thiosulfate to produce thiocyanate in reaction processes catalyzed by sulphur tranferase enzymes such as rhodanese. The thiocyanate is then excreted in the urine over a period of days.

Organoleptic and Physico-chemical Characteristics of Tribal Bean Tofu

The organoleptic test showed that panelists preferred the color of tribal bean tofu made of a mixture of tribal bean and soybean at a ratio of 50:50 plus 2% lactic acid as a coagulant (3.63) that was white. However, it was not significantly different with the tofu made of a mixture of tribal bean and soybean at a ratio of 50:50 plus 2% rice vinegar. The level of con-

sumers' acceptance of the tofu is the highest (78%). Tribal bean tofu made of a mixture of tribal bean and soybean at a ratio of 25:75 plus pineapple extract was mostly rejected by the panelists (72%), because the tofu had yellowish color and sour taste (Table 6 and 7).

Organoleptic characteristics of tofu made of a mixture of tribal bean and soybean plus 2% rice vinegar were not significantly different with that plus 2% lactic acid, but it was significantly different with that plus pineapple extract coagulant. Tofu made of a mixture of tribal bean and soybean at a ratio of 25:75 plus 2% rice vinegar had the highest preference on texture (3.55) and acceptance (78%). The lowest preference value on texture (1.65) was showed by tofu made of a mixture of tribal bean and soybean at a ratio of 25:75 plus pineapple extract and the level of consumer rejection of 98% (Table 6).

The flavor of tribal bean tofu plus 2% rice vinegar was not significantly different with that plus 2% lactic acid, but significantly different with that plus pineapple extract. Tofu made of a mixture of tribal bean and soybean at a ratio of 25:75 plus 2% rice vinegar had the highest preference score on flavor (3.30). The tofu made of a mixture of tribal bean and soybean (25:75) plus pineapple extract had the lowest preference score on flavor (2.43). Panelists mostly preferred the tofu taste made of tribal bean and

Table 6. Preference score of panelists on tofu made of a mixture of tribal bean and soybean with different coagulants.

Ratio of tribal bean and soybean	Coagulant	Organoleptic characteristic				
		Color	Texture	Flavor	Taste	Overall
50:50	Rice vinegar 2%	3.55b	3.50b	3.10b	2.64bc	3.25bc
25:75	Rice vinegar 2%	3.38b	3.55b	3.30b	3.49d	3.31c
50:50	Lactic acid 2%	3.63b	3.35b	3.25b	2.36b	2.92b
25:75	Lactic acid 2%	3.48b	3.38b	3.20b	2.94c	3.19bc
50:50	Pineapple extract	2.05a	1.96a	2.55a	1.84a	1.86a
25:75	Pineapple extract	2.15a	1.65a	2.43a	1.74a	1.79a

Numbers in the same column followed by the same letter are not significantly different at 5% DMRT.

Table 7. Rendement, color, and flavor of tofu made of a mixture of tribal bean and soybean with different coagulants.

Ratio of tribal bean and soybean	Coagulant	Rendement	Color	Flavor
50:50	Rice vinegar 2%	143.55	White	Normal
25:75	Rice vinegar 2%	152.98	White	Normal
50:50	Lactic acid 2%	148.91	White	Normal
25:75	Lactic acid 2%	145.67	White	Normal
50:50	Pineapple extract	107.46	Yellowish	Normal
25:75	Pineapple extract	131.81	Yellowish	Normal

soybean (25:75) plus 2% rice vinegar (3.49). The tofu made of a mixture of tribal bean and soybean (25:75) plus pineapple extract had the lowest value (1.74) because of the bitter taste.

Overall, the panelists mostly preferred the tofu made of tribal bean and soybean at a ratio of 25:75 plus 2% rice vinegar (3.31) with the acceptance level of 58%. The tofu made of tribal bean and soybean (25:75) plus pineapple extract had the lowest overall preferences value (1.79) and the highest rejection (93%).

Tofu made of a mixture of tribal bean and soybean at a ratio of 25:75 plus 2% rice vinegar had the highest rendement (152.98%) than the other ratios. While the tofu made of tribal bean and soybean at a ratio of 50:50 plus pineapple extract had the lowest rendement (107.46%) (Table 7).

The color of tofu made by adding rice vinegar and lactic acid as coagulants is white, while that added with pineapple extract had yellowish color. Overall, the tofu produced had a normal flavor. This is in conformity with the quality requirements of SNI 01-3142-1992 revised SII No. 0270-80 (Dewan Standardisasi Nasional 1992).

Plasticity of tofu was stated with F-max value. The greater the F-max value, the tofu is more chewy. Tofu texture is influenced by coagulant used (Suhaidi 2003). The texture of tofu made of a mixture of tribal bean and soybean at a ratio of 25:75 plus 2% rice vinegar as a coagulant is chewy (58.62 N) (Table 8).

Tofu made of tribal bean and soybean (50:50) plus 2% rice vinegar had the highest HCN content (140 ppm), and the lowest HCN content (85 ppm) was found on tofu made of a mixture of tribal bean and soybean at a ratio of 25:75 plus 2% rice vinegar (Table 9).

The acidity (pH) includes coagulant pH and whey pH (Table 10). Protein coagulation during tofu processing changes the whey pH. The initial pH of tribal bean and soy milk that was 7 turned into 4.5 with addition of 2% rice vinegar. The pH decreased to 3 by addition of 2% lactic acid and turned into 5 with

Table 8. Texture of tofu made of a mixture of tribal bean and soybean with different coagulants.

Ratio of tribal bean and soybean	Coagulant	F-max (N)
50:50	Rice vinegar 2%	9.44
25:75	Rice vinegar 2%	58.62
50:50	Lactic acid 2%	11.89
25:75	Lactic acid 2%	52.42
50:50	Pineapple extract	4.40
25:75	Pineapple extract	3.60

pineapple extract coagulant. The whey could be used for tofu processing on the next day, but only for one use, because the whey had pH 6.

The chemical analysis showed that most panelists preferred the tofu made of a mixture of tribal bean and soybean at a ratio of 25:75 plus 2% rice vinegar as a coagulant (Table 11). The crude fiber of the tofu (0.25%) is higher than the quality standard (maximum 0.1) (Table 11). The high crude fiber content was due to the high crude fiber content of tribal bean (1.83%). The protein content of the tofu is qualified to SNI 01-3142-1992 revised by SII No. 0270-80.

Although tribal bean is rich in protein and minerals, it contains HCN which is harmful if it is consumed frequently. Therefore, further study is required to eliminate HCN content in the tribal bean.

Table 9. The HCN content of tofu made of a mixture of tribal bean and soybean with different coagulants.

Ratio of tribal bean and soybean	Coagulant	HCN (ppm)
50:50	Rice vinegar 2%	140
25:75	Rice vinegar 2%	85
50:50	Lactic acid 2%	88
25:75	Lactic acid 2%	115
50:50	Pineapple extract	98
25:75	Pineapple extract	109

Table 10. Acidity (pH) of coagulant and whey in processing tofu from a mixture of tribal bean and soybean.

Ratio of tribal bean and soybean	Coagulant	Milk pH	Coagulant pH	Whey pH
50:50	Rice vinegar 2%	7	3.0	4.5
25:75	Rice vinegar 2%	7	3.0	4.5
50:50	Lactic acid 2%	7	1.5	3.0
25:75	Lactic acid 2%	7	1.5	3.0
50:50	Pineapple extract	7	4.0	5.0
25:75	Pineapple extract	7	4.0	5.0

Table 11. Chemical content of tofu made of a mixture of tribal bean (75%) and soybean (25%) with 2% rice vinegar coagulant.

Component	Amount (%)	Quality standard of tofu ¹
Water	76	-
Ash	2.71	Maximum 1
Protein	14	Minimum 9
Crude fiber	0.25	Maximum 0.1
Lipid	14	-

¹SNI 01-3142-1992 revised SII No. 0270-80 (Dewan Standardisasi Nasional 1992)

CONCLUSION

This research has proven that tribal bean can be used to partially substitute the use of pricey soybean for making tempeh. Its high HCN content, requires careful pre-treatment by way of soaking for as long as 48 hours. Tribal bean use for tempeh of up to 50% mixture with soybean meets the standard of organoleptic content, flavor, taste, color, texture and HCN content. However, its use for tofu, even at a low portion of 25% of the mixture, failed to meet the quality standard as its HCN content was still higher than 50 ppm. Further study on the techniques for suppression of HCN content of this bean is needed

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