Original Article

Indigenous *mucuna* tempe as functional food*

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The aim of the present study was to obtain the nutritive and bioactive compounds of *mucuna* tempe and the consumer preference of its formulated snack. The tempe was made traditionally from *mucuna* prurience seeds using 'ragi tempe' as inoculants. Proximate, vitamin, mineral and dietary fibre analysis was done using the methods of the Association of Official Analytical Chemists, isoflavone was analysed using high performance liquid chromatography (HPLC) with an RP-18 column and a UV detector. The preference test was done using scoring methods by trained panelists. The study demonstrates that compared with soybean tempe, *mucuna* tempe had a higher dietary fibre level, but lower vitamin E content. The *mucuna* tempe contains 31.5% protein, 7.3% fat, 3.0% ash, 58.1% carbohydrate and 9.1% fibre. It contains 0.551 mg/L isoflavone aglucone; daidzin is the highest, followed by Factor II (6,7,4 trihydroxy isoflavone) that is much higher than that of soybeans tempe. These are much higher isoflavone aglucone contents than found in soybeans tempe. The preference score for a 20% *mucuna* tempe-based snack was not significantly different compared with control or with the preference score for a 10% soybean tempe-based snack. This preliminary study needs to be followed by both *in vitro* and *in vivo* studies.

Key words: fibre, Indonesia, isoflavone aglucone, mineral, mucuna tempe, proximate, snack preference, vitamin.

Introduction

Tempe is one of the Indonesian traditional foods made by a fermentation process using a pure culture of *Rhizopus* sp. (*Rhizopus oryzae* or *Rhizopus oligosporus*) or mixed culture in the form of 'ragi tempe' as inoculant. During fermentation, the fungi grows throughout dehulled cooked soybeans and forms a white compact cake. Nowadays, tempe is becoming a popular healthy food worldwide or super food because of its bioactive compounds and healthy properties.^{1–3} Tempe is usually made from soybean, but almost all legume seeds are suitable for tempe processing.

Mucuna prurience L.DC. var. *utilis*, grows well in marginal dry land areas in Indonesia, and is one of Indonesia's indigenous beans. The *mucuna* seeds contain high protein, carbohydrate and fibre.^{4,5} Non-nutritional factors such as cyanogenic glucoside in *mucuna* seeds could be eliminated by appropriate soaking and boiling. Although *mucuna* has potential for some food products, up to the present time its tempe has low social economic status.⁶

Great attention has been given to the development of a health food with expected health benefits.^{6,7} The bioactive compounds in the health foods such as dietary fibre, oligo-saccharides, glycoside, vitamins and minerals, have physiological functions to regulate specified metabolic processes.^{8–10}

Soybean tempe which contains bioactive compounds such as superoxide dismutase (SOD) enzyme, isoflavone aglucone, and dietary fibre^{1,2,11,12} has been known as a functional food. Similarly, *mucuna* tempe is also expected to contain certain beneficial components. As a result, *mucuna* tempebased snacks have been formulated and promoted as a functional food. The author proposes that *mucuna* tempe might have similar beneficial nutrition components to soybean tempe, thus *mucuna* tempe could be classed as a functional food. The aim of the present study is to:

- 1. Study the nutrition components of *mucuna* and its tempe.
- 2. Study the bioactive components of *mucuna* tempe.

3. Study the consumer preference of a *mucuna* tempe formulated snack.

Materials and methods

The sample of *mucuna* seeds was obtained from farmers in Central Java, Indonesia. *Mucuna* tempe was made with traditional methods as follows: the seeds were soaked in water 3×24 h to remove the toxic constituent and to facilitate water uptake. The swollen beans were boiled until sufficiently tender (approximately 30 min), drained, peeled and sliced. The sliced beans were blanched for approximately 1 h, cooled, and inoculated with a traditional inoculum 'ragi tempe'. 'Ragi tempe' is a kind of tempe starter in the form of a white powder of mixed culture of *Rhizopus* sp. The inoculated substrate was wrapped in plastic bags, incubated for 48 h at approximately 30 °C, until firmly bound together by mycelium of the mould, and forming a compact white cake.

Proximate analysis, vitamins, minerals and dietary fibre was done according to AOAC:¹³ moisture and ash content by the thermographimetry method, protein by the micro-Kjeldahl method, fat by soxhlet extraction, fibre by hydrolysis, vitamin E by spectrophotometry, minerals using atomic absorption speectrophotometry (AAS) and dietary fibre by the detergent method.

Isoflavone aglucone was assayed using HPLC with RP-18 column and UV detector, after extraction with absolute

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^{*}This paper has been published in the Proceeding of the International Tempe Symposium in Bali, Indonesia 1997.

	Μ	Soybeans	
	Ref 21	Ref 5	Ref 22
Protein	33.8	28.4-31.0	46.3
Fat	4.8	3.4-5.1	19.1
Ash	3.4	_	6.3
Carbohydrate	50.1	62.3-63.3	28.5
Fibre	7.3	15.5-16.6	3.7

Table 1. Proximate content of *mucuna* and soybean seeds (% db)

 Table 2. Mineral content of soybean and mucuna seeds (mg/100g)

Kinds of bean	K	Na	Ca	Mg	Cu	Fe	Mn	Zn
Soybean								
seeds	89	5	57	131	1.25	8.20	2.40	5.95
tempe	111	29	29					
Mucuna								
seeds	131	2	37	328	1.10	9.45	1.35	3.60
tempe	85	31	23					

K, potassium; Na, sodium; Ca, calcium; Mg, magnesium; Cu, copper;

Fe, iron; Mn, manganese; Zn, zinc.

methanol and purification with polyamide in methanol at a concentration of 25, 50, and 70%.

Preference test was done using scoring methods by some trained testers.¹⁴ Data were analysed with ANOVA and Duncan Mutiple's Range Test.

Results and discussion

The proximate and mineral contents of mucuna seeds

Table 1 shows the proximate content of *mucuna* compared with soybeans. The protein and fat contents of *mucuna* are lower than those of soybeans, but the carbohydrate content is approximately twofold higher, the fibre content is also higher. The variable proximate components of *mucuna* are probably due to genetics, growing area and methods of analysis.

Minerals including iron, calcium and magnesium are well recognized as essential components of nutrition; and have physiological functions as bioactive compounds.^{9,15} The claimed benefits of minerals include: increased levels of calcium, iron, bone and tooth growth, as well as prevention of osteoporosis and certain anaemias. Table 2 shows that some of the mineral contents of *mucuna* seeds are as high as those of soybeans; the levels of magnesium and iron are higher, but the calcium content is lower. In *mucuna* and soybean tempe, the magnesium and iron content is similar. The iron content of both seeds is lower compared to its tempe form, possibly due to the microbiological activities occurring during the fermentation process.

Chemical composition of mucuna seeds and its tempe

Mucuna seeds and its tempe contain a considerable amount of protein, although not as much as in soybean (Table 3). Carbohydrate and fibre contents are nearly twofold higher, while the fat in *mucuna* is low compared to soybean. Tempe manufacturing has caused these changes. *Mucuna* tempe has lower protein and fat content compared to soybean tempe, but its carbohydrate and fibre contents are higher.

Table 3. Chemical composition of soybean and *mucuna*tempe products (% db)

	Protein	Fat	Ash	Carbohydrate	Fibre	Ref
Soybean						
seeds	46.3	19.1	6.3	28.5	3.7	22
tempe	45.9	18.3	2.8	32.8	5.9	4
	46.5	19.7	3.6	30.2	3.1	22^{+}
Мисипа						
seeds	33.8	4.8	3.4	50.1	7.3	4
tempe	31.5	7.3	3.0	58.1	9.1	4

 $^{\dagger} \text{This}$ line of values represent that the component analysis sometimes varies with each other.

 Table 4. HCN content of mucuna seeds and its tempe (mg/100g)

	Whole beans	Peeled beans	
Fresh	11.050 a	10.070 b	
Soaked in water			
1×24 h	9.922 c	5.568 d	
2×24 h	2.348 e	1.452 f	
3×24 h	0.310 g	0.265 g	
Tempe	-		0.000 h

Data in column followed by the same letter are not significantly different at $\alpha 5\%.$

 Table 5. Vitamin E, dietary fibre and oligosaccharide of *mucuna* and soybean tempe

Bioactive compounds	Mucuna tempe	Soybean tempe
Vit. E (mg/100g) Dietary fibre (g/100g):	46.587	101.760
NDF	66	44
ADF	28	38
Oligosaccharide	Stachyose	Stachyose

Cyanogenic glucoside of mucuna seeds and its tempe

The cyanic acid (HCN) content of *mucuna* that is released by the cyanogenic glucoside is shown in Table 4. It is shown that soaking has significantly reduced the HCN content in fresh beans, particularly in peeled beans. After 3×24 h soaking, the HCN content is not significantly different in the whole or peeled beans. However, there is no HCN left in tempe form, thus it is safe to be consumed.

Vitamin, dietary fibre and oligosaccharides of mucuna tempe

Dietary fibre has long been recognized to have beneficial effects. Hanny reported that foods with a high content of dietary fibre can reduce the energy intake.⁷ Furthermore, Kritchevsky¹⁶ stated that different fibre sources vary in their effects of reducing the risk of colon cancer. Dietary fibre slows digestion and absorption of lipid.¹⁷

As seen in Table 5, the neutral dietary fibre (NDF) of *mucuna* tempe is higher than that of soybean tempe, but the acid dietary fibre (ADF) is lower. Oligosaccharides are low in calorie content, preventing tooth decay, and controlling bifidobacterium activation. The kind of oligosaccharide in *mucuna* tempe as well as in soybean tempe is stachyose.

Factor II	Daidzin	Glycitein	Genistein	Aglucones total			
0.026	0.041	0.011	0.050	0.131			
0.060	0.304	0.086	0.101	0.551			
0.001	0.068	0.002	0.121	0.192			
0.002	0.060	0.035	0.003	0.100			
0.004	9.052	1.660	0.063	10.779			
	0.026 0.060 0.001 0.002	0.026 0.041 0.060 0.304 0.001 0.068 0.002 0.060	0.026 0.041 0.011 0.060 0.304 0.086 0.001 0.068 0.002 0.002 0.060 0.35	0.026 0.041 0.011 0.050 0.060 0.304 0.086 0.101 0.001 0.068 0.002 0.121 0.002 0.060 0.035 0.003			

Table 6. Isoflavones content of *mucuna* and its tempe (mg/L)

[†]Present study; traditional methods using 'ragi tempe' as inoculant. [‡]Ref 18. [§]Ref 12 lab. methods using *R. oligosporus*.

Table 7. Preference score of some snacks formulated with *mucuna* or soybean tempe

Materials	Appearance	Texture	Taste	Overall
Wheat flour (W)	74 b	78 b	84 d	82 b
W + soybean temp	e			
10%	68 b	78 b	74 cd	76 b
20%	72 b	58 a	54 ab	58 a
30%	62 b	64 ab	52 a	54 a
W + mucuna temp	e			
10%	64 b	64 ab	78 cd	68 ab
20%	56 ab	60 a	74 cd	68 ab
30%	40 a	52 a	68 bc	60 a

Data in column followed by the same letter are not significantly different at $\alpha 5\%.$

Table 5 also shows the vitamin E content of *mucuna* tempe. Vitamin E or α -tocopherol found in green vegetables, grains and meat inhibits carcinogenesis, such as UV-induced skin cancer, and also inhibits cell transformation.⁷ Van den Broek⁹ reported that vitamin E and isoflavones have physiological functions for intestinal and cholesterol control, metabolism improvement, hypotensive and anti-oxidative effects.

Isoflavones of mucuna and its tempe

Mucuna contains the isoflavones genistein and daidzin. As seen in Table 6, isoflavone levels in *mucuna* seeds are lower than in its tempe. The soybean isoflavones have been reported to have an oestrogenic effect as well as anti-oxidant and anticancer functions.⁷ However, as yet there are no data on *mucuna* and its tempe.

The raw beans contain a small amount of aglucone, as the isoflavones of the beans were in the form of a glucoside, a complex form with carbohydrate.¹⁸ The minor component in *mucuna* is glycitein, while in soybeans it is Factor II (6,7,4; trihydroxy isoflavone).

During tempe processing the aglucones increased because *R. oligosporus* in the fermentation hydrolysed the isoflavones. Wuryani¹⁹ concluded that during tempe fermentation the isoflavone glycosides (daidzin and genistin) were hydrolysed into their aglucones (daidzin and genistein). Wuryani¹⁹ stated that daidzin and genistein were the main isoflavones responsible for the anti-oxidative activity. Total daidzin and genistein in *mucuna* was higher than in soybeans, while it is the opposite in its tempe form. Factor II (6,7,4; trihydroxy isoflavone) and genistein in *mucuna* and its tempe were higher than in soybeans. *Mucuna* and its tempe contain higher Factor II (6,7,4; trihydroxy isoflavone)

and lower daidzin and glycitein than that of soybeans, possibly due to the bacterial activity during fermentation that produced Factor II (6,7,4; trihydroxy isoflavone) from daidzin and genistein.¹²

Mucuna tempe formulated snack

The nutritive and the bioactive compounds of mucuna tempe are considerably high and similar to those of soybeans, but up to now the development of *mucuna* tempe as a health food has been limited. In order to improve the development of mucuna tempe as functional food, it is important to produce some snacks formulated with mucuna tempe. The scoring preference of some snacks formulated with mucuna or soybeans tempe is seen in Table 7. Substitution of 10% soybean tempe into wheat flour products or with 20% of mucuna tempe into wheat flour products does not significantly decrease the preference score of the panelists. Substitution of 20% soybean tempe to the snack significantly decreased the taste preference score; the product has a bitter aftertaste and moderate beany flavour. Handajani et al.20 concluded that lipoxygenase activity is responsible for producing the beany flavour of mucuna. The higher fat content of soybeans probably affected the development of the stronger beany flavour of its product. Substitution of 20% mucuna tempe decreased the texture preference score; this is due to the hard texture of the product, while the appearance is not significantly different.

Conclusion

This study demonstrated that compared with soybean tempe *mucuna* tempe has a higher dietary fibre level, but lower vitamin E level. *Mucuna* tempe contains 0.551 mg/L isoflavone aglucones, with daidzin as the highest. It contains a higher level of Factor II (6,7,4; trihydroxy isoflavone), but lower daidzin and glycitein than soybeans. The preference score of a 20% *mucuna* tempe-based snack is not significantly different with either control or a 10% soybean tempe-based snack. This preliminary study needs to be followed by both *in vitro* and *in vivo* studies.

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