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Research Article
Chemical, Nutritional, Physical and Sensory Characterization of Tempe Made from Various Underutilized Legumes

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Abstract

Background and Objective: The exploration of a new food resource is important with the inevitably fast-growing global population. This study aimed to characterize the chemical, nutritional, physical and sensory properties of tempe made from various underutilized legumes. **Materials and Methods:** The chemical and nutritional characterization included proximate analysis and antinutrient compound determination, amino acid profiles and micronutrient analysis. Moreover, the sensory evaluation included that of appearance, texture, aroma, taste and overall acceptance and the physical characterization included texture and color analyses. The underutilized legumes used in this study were mung bean, cowpea, black soybean, kidney bean, groundnut and velvet bean. Tempe made from yellow soybeans, which is commonly consumed, was used as a control. **Results:** In terms of nutrition and chemical composition, black soybean was superior to the others and exhibited better properties than those of the control. On the other hand, the overall panelist acceptability of tempe made from underutilized legumes was lower than that of the control. **Conclusion:** This study found that underutilized legumes were potential raw material for making tempe. However, education, promotion and habituation will be needed to increase their acceptability by consumers.

Key words: Tempe, underutilized legumes, yellow soybeans, black soybeans, vegetable protein, phytate contents

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

As a result of population growth and changes in diets, demands of various food items are continuously rising. By 2050, the global population is estimated to reach 9 billion[1]. On the other hand, agriculture faces multiple challenges, such as unpredictable weather patterns and the continuing decline of arable land, leading to an imbalance in food demand and supply. This situation is worsening, particularly in developing countries with a high population, such as Indonesia. Indonesia experiences one of the largest proportion of growth stunting in the world and one-third of Indonesian children suffer from growth stunting[2]. On the other hand, Indonesia is known as a mega-diverse country and is rich in natural resources, including a variety of legumes that are valuable protein sources[3]. However, only a few legumes have been fully utilized. The use of underutilized legumes is restricted by the presence of antinutrient compounds that inhibit the absorption of nutrients in the body[4]. In addition, nutritional information of the underutilized legumes is still limited. One way to reduce the content of antinutrients in foods is by fermentation[5]; therefore, local Indonesian legumes can be used as raw material in tempe production. Tempe is one of the most popular sources of protein in Indonesia, with a consumption level of 0.46 kg $week^{-1}$ $person^{-1}$, which was higher than that of beef (0.009 kg $week^{-1}$ $person^{-1}$) and chicken (0.121 kg $week^{-1}$ $person^{-1}$) in 2018[6]. Tempe is generally made from fermented yellow soybean; hence, the dependency on yellow soybean is very high.

Several studies related to the use of alternative legumes to yellow soybean as raw material for making tempe have been carried out. Some of them have investigated the isoflavone content in mung bean tempe[7] and kidney bean[8], the vitamin E content and antioxidant activity in mung bean tempe[9] and the use of cowpea and gude as yellow soybean substitutes in tempeproduction[10]. However, these studies used different analytical methods; therefore, the results cannot be compared. In addition, these studies only focused on one or two aspects, such as chemical composition, functionality, nutrition, or sensory properties. Hence, the current study aimed to characterize the chemical, nutritional, physical and sensory properties of tempe made from six different underutilized legumes. The results of this study are expected to provide comprehensive information about the potential tempe made from underutilized legumes that could be needed to promote underutilized legumes as local food resources.

MATERIALS AND METHODS

Materials: Local varieties of yellow soybean (*Glycine max*), mung bean (*Vigna radiata*), cowpea (*Vigna unguiculata*), black soybean (*Glycine max*), kidney bean (*Phaseolus vulgaris*), groundnut (*Arachis hypogae*) and velvet bean (*Mucuna pruriens*) were obtained from a local market in Yogyakarta, Indonesia.

Tempe making: The processes of making tempe from various underutilized legumes were adopted from those utilized for making yellow soybean tempe. The first step was washing the beans twice, followed by dehulling, soaking overnight with a bean to water ratio of 1-3, boiling for 10-60 min steaming for 10-90 min, inoculating and fermenting. The cooking time varied (Table 1) for each legume because of their different characteristics. The cooked beans were then inoculated with 0.02% of a commercial starter of *Rhizopus oligosporus* (Raprima). Subsequently, the beans were packed in 10×15 cm^2 perforated plastic bags and incubated for 36 h at room temperature.

Sensory evaluation: Sensory evaluation was conducted by a hedonic test according to Meilgaard *et al.*[11]. The sensory evaluation was carried out with 34 untrained panelists. The scale used was a 1-7 scale (1 = dislike very much, 2 = dislike, 3 = quite dislike, 4 = neutral, 5 = quite like, 6 = like and 7 = like very much). The attributes tested for the fresh tempe were color, appearance, flavor, compactness and overall acceptance, whereas for fried tempe, the attributes included color, appearance, flavor, texture, taste and overall acceptance. For making fried tempe, fresh tempe was cut into

Table 1: Various conditions for tempe production with different legumes

Legume	Soaking time	Boiling time	Steam cooking time	References
Yellow soybean	24 h, twice	30 min	30 min	-
Mung bean	12 h, once	-	30 min	-
Velvet bean	24 h, once	60 min	15 min	-
Groundnut	24 h, once	-	90 min	6
Kidney bean	24 h, once	10 min	30 min	7
Cowpea	10 h, once	15 min	15 min	8
Black soybean	36 h, once	30 min	60 min	9

$2\times2\times2$ cm^3 pieces and soaked in 10% NaCl for 10 min. The soaked tempe was then fried in a deep fryer (Kenwood) using palm oil for 10 min at 155°C.

Chemical characterization

Amino acid composition: Amino acid analysis was conducted in the accredited and certified laboratory of Saraswanti Indo Genetech (SIG), Bogor, West Java, Indonesia. The method used refers to that of Abdul Rohman and Ibnu Gholib[12] and the Bio Amino Acid Analysis System Guide[13]. A sample was weighed (0.1 g) and added to 5 mL 6 N HCl. The samples were then hydrolyzed for 22 h at 110°C. After hydrolysis, the mixtures were cooled and transferred into 50 mL volumetric flasks and diluted with pure water. The solutions were filtered through a 0.20 µm pore size filter. The filtrate (500 µL) was pipetted and 40 µL AABA+460 µL distilled water was added. A 40 µL standard of mixed amino acids was pipetted and 40 µL of the internal standard AABA and 920 µL distilled water were added and then homogenized. The mixture or standard (10 µL) was added to 70 µL AccQ-Fluor borate buffer and vortexed. Then, 20 µL of fluor A reagent was added and mixed. After 1 min, the sample or standard was incubated at 55°C for 10 min. An aliquot (1 µL) was injected into an Ultra Performance Liquid Chromatography (Waters, Milford, USA) system and separated using AccQ. Tag Ultra C18 column (1.7 µm particle size, 2.1 mm$\times$100 mm) at 49°C prior to detection with a photo diode array detector at λ = 260 nm. The separation was performed at a flow rate of 0.5 mL min^{-1}.

Nutrition characterization

Proximate analysis: Samples were prepared in uniform sizes and then analyzed for moisture content by thermogravimetry; the protein content was determined by the micro-Kjeldahl method, the fat content by the Soxhlet method, the ash content by thermogravimetry and the crude fiber carbohydrate contents by difference[14].

HCN content analysis: HCN content was determined by spectrophotometry as described by Nwokoro and Anya with modification[15]. The samples were extracted with distilled water for 2 h at room temperature. One milliliter of the extract was then added to 1 mL0.1 N NaOH and 5 mL alkaline picrate and then diluted with 3 mL distilled water. The solution was then incubated in a 100°C water bath for 30 min. The aliquot was then analyzed in a UV spectrophotometer (Genysys 10S UV-Vis, Thermo Scientific, USA) at a wavelength of 520 nm. KCN (240 mg L^{-1}) was used as a standard.

Phytate content analysis: The phytate content was determined according to Davies and Reid's method[16]. The sample was first dried in a cabinet dryer at 60°C for 10 h and ground until it passed through a 60 mesh sieve. Then, 0.05 g of sample was extracted with 20 mL HNO_3 for 4 h in a shaker incubator at room temperature. The filtrate was then diluted with 0.4 mL distilled water and added to 1 mL $FeCl_3$. Subsequently, the sample was incubated at 100°C in a water bath for 20 min. Then, 5 mL n-amyl alcohol was added and the sample was analyzed in a UV spectrophotometer (Genesys 10S UV-Vis, Thermo Scientific, USA) at λ = 495 nm. Na-phytate was used to generate a standard curve with concentrations of 0.0000, 0.0056, 0.0112, 0.0168 and 0.0224 mg mL^{-1}.

Fe and Zn content analysis: The analysis of the Fe and Zn contents was conducted at SIG with an inductively coupled plasma-optical emission spectrometry (ICP-OES) method using an ICP-OES instrument (Agilent Technologies Type 720, USA). A sample was weighed (0.5-1.0 g) into a vessel, combined with 5-10 mL HNO_3 and then sealed and subjected to microwave digestion. The digested sample was then put into a volumetric flask and diluted with distilled water. The mixture was filtered and analyzed in the ICP-OES system with a plasma gas flow, an auxiliary gas flow and a nebulizer flow of 10 L min^{-1}, 0.5 L min^{-1} and 0.6 L min^{-1}, respectively.

Vitamin B9: The analysis of vitamin B9 was conducted at SIG. Three grams of each sample was dissolved with 0.05 M NaH_2PO_4(pH 6.30) and sonicated for 10-15 min. The mixture was then added to NaH_2PO_4 0.05 M at pH 6.30. An aliquot was then put into a 2 mL tube, centrifuged at 14000 rpm for 25 min and filtered with a Minisart RC 0.20 µm filter. An aliquot (5 µL) was injected into the UPLC-PDA system (Waters Acquity H-Class, USA) and separated using an Acquity BEH C18 column (1.7 µm particle size, 2.1$\times$50 mm) at 40°C. The mobile phase contained 0.1% H_3PO_4 (A) and pure acetonitrile (B) and had a flow rate of 0.2 mL min^{-1}.

Physical characterization

Color and texture analysis: The colors of the fresh and fried tempe samples were analyzed with a chromameter (Konica Minolta, Japan) to determine their L*, a* and b* values. The texture of the fresh tempe was analyzed with a texture analysis machine (TA1, Lloyd Instrument, UK), which determined the hardness, cohesiveness, resilience and springiness index. The samples were cut into $2\times2\times2$ cm^3 samples. The texture analysis machine was operated with the

NEXYGEN Plus 3.0 program, which generated a graph with two curves. The results of the analysis were determined by the relationship of the force and time curves.

Statistical analysis: One-way ANOVA (p<0.05) was used to determine significant effects on different parameters of chemical characteristics on the materials using IBM SPSS Statistics 24. The least significant differences between sample means were determined by Duncan multiple range tests (p<0.05). Kruskal-Wallis and Dunn's multiple comparison tests (p<0.05) were used for physical and sensory characteristics.

RESULTS AND DISCUSSION

Chemical characteristics

Amino acid profile: Amino acids, particularly, essential amino acids, are crucial in our diet since human bodies are not able to synthesize them. The essential amino acids analyzed in this study were phenylalanine, isoleucine, valine, leucine, threonine and histidine. In addition to having nutritional benefit, amino acids also contribute to the taste of food. Figure 1 shows the amino acid profiles for all tempe samples. The yellow soybean tempe had the highest essential amino acid content, up to 89,620.76 mg kg^{-1}, followed by that of the groundnut tempe (88,375.96 mg kg^{-1}) and the lowest was observed in the mung bean tempe (38,566.33 mg kg^{-1}). In addition, the groundnut tempe had the highest leucine and valine levels. The yellow soybean tempe was superior in terms of threonine, histidine and phenylalanine levels, while the black soybean tempe had the highest lysine content.

In general, the most dominant amino acid in all fresh tempe samples was glutamic acid (14,241.77 to 53,370.99 mg kg^{-1}), while the least dominant amino acid in all samples was tyrosine (2,301.13-9,590.11 mg kg^{-1}). As mentioned above, amino acids are responsible for the sweetness, umami, neutral and bitter tastes of food. Consumers usually prefer tempe with an umami taste and no bitter aftertaste. Therefore, glutamic and aspartic acids are desirable because they give an umami taste[17], while hydrophobic amino acids, such as L-phenylalanine, L-tyrosine, L-tryptophan, L-leucine, L-valine and L-isoleucine elicit a bitter taste, which is not favored[18]. Figure 2 shows that the tempe with the highest amino acids giving an umami taste was the groundnut tempe but it also had the highest bitter-tasting amino acid contents. Among the samples, the groundnut tempe had the highest total amino acids associated with

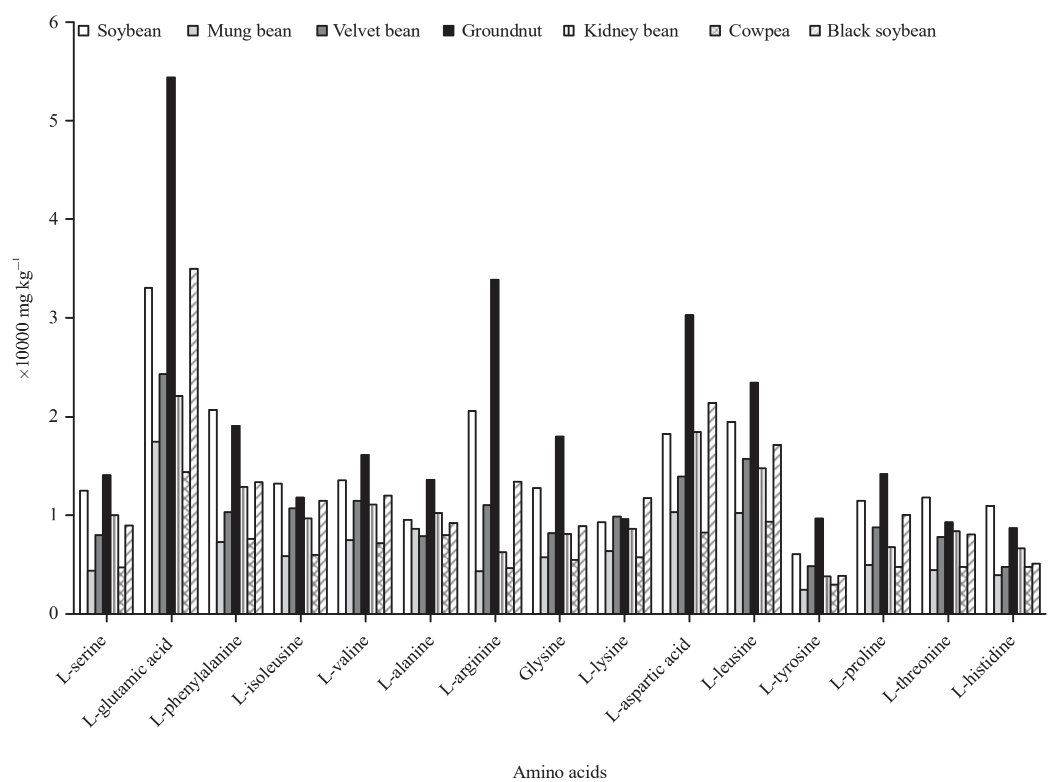

Fig. 1: Profile of amino acids in fresh tempe made from different legumes

Table 2: Proximate content of various raw legumes

Raw legume	Water (%)	Ash (%)*	Proteins (%)*	Lipids (%)*	Crude fiber (%)*	Carbohydrates (%)*
Yellow soybean	10.88^{c}	5.59^{b}	34.15^{b}	19.21^{b}	11.19^{b}	29.82^{e}
Mung bean	11.60^{b}	4.08^{e}	15.78^{g}	3.70^{e}	6.59^{e}	69.82^{b}
Velvet bean	10.81^{c}	4.52^{d}	23.21^{e}	6.07^{d}	18.14^{a}	48.05^{d}
Groundnut	9.11^{d}	2.41^{f}	33.35^{c}	46.73^{a}	8.65^{d}	8.81^{f}
Kidney bean	12.62^{a}	4.70^{d}	24.35^{d}	6.31^{d}	6.79^{e}	57.85^{c}
Cowpea	11.50^{b}	5.23^{c}	19.75^{f}	1.85^{f}	1.69^{f}	71.47^{a}
Black soybean	9.21^{d}	6.65^{a}	39.28^{a}	13.66^{c}	10.39^{c}	30.01^{e}

Different superscripts indicate significant differences ($p<0.05$) as determined by one-way ANOVA followed by Duncan's MRT. *of dry matter

Table 3: Proximate content of fresh tempe made from different legumes

Tempe	Water (%)	Ash (%)*	Proteins (%)*	Lipids (%)*	Crude fiber (%)*	Carbohydrates (%)*
Yellow soybean	61.79^{b}	1.88^{bc}	42.80^{b}	22.53^{b}	2.64^{bc}	21.83^{b}
Mung bean	70.60^{a}	6.23^{a}	36.33^{c}	1.73^{d}	2.79^{b}	23.25^{b}
Velvet bean	60.62^{b}	1.20^{d}	30.39^{d}	5.54^{c}	2.56^{bc}	31.43^{a}
Groundnut	40.32^{c}	2.49^{b}	27.71^{e}	48.66^{a}	2.04^{c}	19.63^{c}
Kidney bean	62.48^{b}	1.50^{cd}	29.21^{de}	1.79^{d}	3.76^{a}	30.64^{a}
Cowpea	68.36^{a}	2.10^{bc}	25.36^{f}	3.43^{d}	0.93^{d}	21.39^{b}
Black soybean	62.23^{b}	2.17^{bc}	51.37^{a}	23.37^{b}	3.03^{b}	6.30^{d}

Different superscripts indicate significant differences ($p<0.05$) as determined by one-way ANOVA followed by Duncan's MRT. *of dry matter

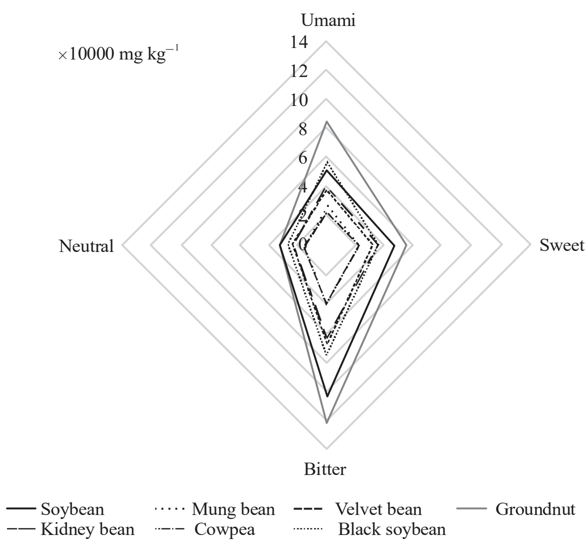

Fig. 2: Profile of amino acids based on their taste in fresh tempe made from different legumes

umami, sweet, bitter and neutral tastes compared to that of the other samples, indicating that groundnut tempe had the most diverse amino acid content.

Nutritional characteristics

Proximate content: The proximate analysis of all legumes is presented in Table 2. Protein and fat are the most important parameters for tempe. Tempe is expected to have high protein and low fat contents. Among the samples, black soybean had the highest protein content, followed by that of yellow soybean and groundnut, while the lowest was that of mung bean. In terms of fat content, cowpea had the lowest fat content, followed by that of mung bean and kidney bean.

Fermentation can change the proximate content. As shown in Table 3, the water content increased significantly in the tempe. This could have been caused by the soaking and boiling processes during tempe production. Soaking beans increases the water in the beans[19] as the water penetrates the seed coat into the bean center[20]. The mung bean tempe and cowpea tempe obtained the highest water contents. In addition, the protein content was also higher in tempe than in the raw legumes, which was due to the protease activity produced by *Rhizopus oligosporus* during fermentation[21]. The black soybean tempe had the highest protein content. Therefore, black soybean is a potential candidate for tempe production. On the other hand, the groundnut tempe had the highest fat content, as the fat content of this legume was greater than that of the other legumes.

Antinutrient compounds

HCN content: HCN is a toxic compound that is found in many plants, especially in legumes. In the 7 types of local legumes tested, velvet bean had the highest HCN content (Table 4). The decrease in the HCN content of each type of tempe also varied between 19.34% (mung bean) to 94.12% (velvet bean). In general, the HCN content decreased during the tempe-making process. Boiling, stripping, soaking with water and cooking are processes that can reduce the HCN content. Cyanogenic glycoside was extracted during soaking. Heat treatment can also reduce the cyanide content[22]. Similarly, Aman[23] reported that boiling legumes for 30 min can deactivate the linamarase

Table 4: HCN content of raw legumes and fresh tempe made from different legumes

Legume	HCN content in raw sample (ppm)	HCN content in tempe (ppm)	Reduction (%)
Yellow soybean	3.58±0.08[c]	0.51±0.06[e]	85.80±1.97[b]
Mung bean	1.35±0.01[e]	1.09±0.04[c]	19.34±2.27[e]
Velvet bean	45.48±0.46[a]	2.67±0.11[a]	94.12±0.22[a]
Groundnut	3.28±0.27[c]	1.00±0.12[c]	69.48±1.10[d]
Kidney bean	4.58±0.35[b]	1.48±0.27[b]	67.77±3.54[d]
Cowpea	1.87±0.03[d]	1.16±0.01[c]	37.61±1.23[e]
Black soybean	3.67±0.56[c]	0.74±0.09[d]	78.99±5.77[c]

Different superscripts indicate significant differences ($p<0.05$) as determined by one-way ANOVA followed by Duncan's MRT

Table 5: Phytate content of raw legumes and fresh tempe made from different legumes

Legume	Phytate content in raw sample (%)*	Phytate content in tempe (%)*	Reduction (%)
Yellow soybean	1.23±0.02[e]	0.32±0.01[cd]	74.18±1.15[b]
Mung bean	0.96±0.02[c]	0.38±0.03[d]	60.66±3.52[a]
Velvet bean	1.94±0.02[f]	0.14±0.01[b]	92.91±0.58[c]
Groundnut	0.65±0.02[b]	0.30±0.03[c]	53.67±3.98[a]
Kidney bean	0.55±0.01[a]	0.06±0.02[a]	88.96±2.46[c]
Cowpea	0.70±0.01[b]	0.32±0.02[cd]	55.33±3.06[a]
Black soybean	1.05±0.03[d]	0.46±0.03[e]	55.56±4.18[a]

Different superscripts indicate significant differences ($p<0.05$) as determined by one-way ANOVA followed by Duncan's MRT. *dry matter

and glucosidase enzymes, discontinuing the formation of HCN. The longer the duration of boiling, the lower the cyanide content because it dissolves easily in water and volatilizes due to heat[24].

The HCN content in all tempe samples varied between 0.51 and 2.67 ppm and the velvet bean tempe had the highest cyanide content. However, the HCN concentrations decreased significantly by up to 92.91%. The greatest decrease was probably because of an extended boiling process, while the smallest decrease in the HCN content in the mung bean and cowpea tempe was probably because their soaking and cooking times were shorter. The acceptable daily intake (ADI) of cyanide, according to the FAO, is 0.05 mg kg^{-1} body weight per day[25], which implies that velvet bean tempe is safe for consumption.

Phytate content: Phytic acid (myoinositol hexaphosphate) constitutes 1-3% of most plant seeds. It has a chelating potential and affects the absorption of polycationic neutralizers, such as Zn^{2+} and Fe^{3+}. The content of phytic acid in legumes ranges from 0.8-5.3% dry basis[26].

Among the samples, the velvet beans had the highest phytic acid content, followed by that of the yellow soybeans and black soybeans (Table 5). During the tempe-making process, the phytic acid of most legumes decreased. Phytic acid can dissolve in water; hence, soaking could reduce the phytic acid content. Soaking also increased the activity of phytase enzymes capable of hydrolyzing phytic acid. The optimal pH of the phytase enzyme activity is 5.0-5.2[27] and the growth of lactic acid bacteria during the soaking process also reduces the pH of the legumes to approximately 4.5-5.3, so phytase enzyme activity increases and the hydrolysis of phytic acid also increases[28,29]. In addition, *Rhizopus oligosporus* generates a phytase enzyme that hydrolyzes phytic acid into organic phosphate and inositol during fermentation[5,24]. Furthermore, the size of the legume used for making tempe can affect the phytic acid content. Rochmah *et al.*[30] showed that tempe made from velvet beans that underwent size reduction was easier to penetrate by the fungus mycelia, resulting in a greater phytic acid decrease. During the process of boiling, phytate will dissolve in the water, reducing the phytic acid content in the beans.

Of the tempe samples, the black soybean tempe had the highest phytic acid content, followed by that of the mung bean tempe, soybean tempe and cowpea tempe. However, the levels of phytic acid in all tempe samples were safe because the daily intake can be as high as 4500 mg[31]. The highest phytic acid reduction was observed in the velvet bean tempe. This antinutrient will become further reduced in tempe when fried before being consumed, as deep-fat frying of tempe in peanut oil halves the phytic acid content[5].

Micronutrient

Folate content: Folate (B9) is a vitamin that plays a critical role in the biosynthesis of nucleotides and in methylation reactions[32]. Table 6 shows that the mung bean tempe had the highest folate content, while the yellow soybean tempe had the lowest folic acid content. Several factors affecting the folic acid content of tempe are soaking, peeling, boiling and fermentation. Soaking could dissolve folate in water, thus decreasing the folate content. Boiling could degrade folate due to heat damage. On the other hand, fermentation

Table 6: Folate content of fresh tempe made from different legumes

Tempe	Folate (µg 100 g^{-1})
Yellow soybean	10,213.16
Mung bean	24,372.84
Velvet bean	11,307.43
Groundnut	11,806.97
Kidney bean	10,520.26
Cowpea	15,723.74
Black soybean	10,437.58

Table 7: Zinc and iron content of fresh tempe made from different legumes

Tempe	Zn (mg 100 g^{-1})	Fe (mg 100 g^{-1})
Yellow soybean	2.27	4.34
Mung bean	1.94	4.14
Velvet bean	2.44	4.11
Groundnut	3.14	4.16
Kidney bean	1.86	4.71
Cowpea	1.81	4.26
Black soybean	2.70	5.12

increases the folate content. During fermentation by Rhizopus oligosporus*, folic acid was synthesized; thus, folic acid increased by up to 4-5 times after 48 h*[33]*. Changes in folate content that occur during the fermentation process are likely related to the activity of the protease enzyme, which breaks down proteins into folate*[33]*.*

Fe and Zn content: *Zinc has an important effect on homeostasis, immune function, oxidative stress, apoptosis and aging. In addition, many significant disorders of great* public health, such as atherosclerosis, neurological disorders, autoimmune diseases and age-related degenerative diseases, are associated with zinc de ciency[34]. Apart from zinc, another metal element essential for the body is iron. Iron carries oxygen from the lungs to body tissues by red blood cell hemoglobin, transports electrons within cells and acts as an integrated part of critical enzyme systems in various tissues[35]. The zinc and iron contents of all tempe samples are presented in Table 7. The results showed that the highest Zn content was found in the groundnut tempe, which was almost 1.5 times higher than that of the yellow soybean tempe. The next highest Zn content was observed in the black soybean tempe and velvet bean tempe, while the lowest zinc content was obtained for the cowpea tempe. For iron, the black soybean tempe had the highest among the samples, which was approximately 1.2 times higher than that of the yellow soybean tempe. The next highest iron content was observed in the kidney bean tempe, yellow soybean tempe and cowpea tempe, while velvet bean tempe contained the lowest amount of iron.

Physical characteristics

Texture: Texture profile analysis (TPA) imitates the process that occurs when food is bitten and then chewed by molars until it is ready to be swallowed[36]. The value of cohesiveness shows the ability of bonding in the product to resist deformation. The tempe compactness was shown through the cohesiveness value. The higher the value of cohesiveness, the more compact the tempe was. The cohesiveness of the tempe was influenced by the fungus mycelia's ability to penetrate the seeds. Moreover, compact tempe is a product determined to have good quality. The texture analysis of all tempe samples is presented in Fig. 3 and 4. The results showed that the yellow soybean and black soybean tempe samples had the highest compactness.

Springiness is the speed at which a compressed material returns to its previous condition, while the springiness index is the ratio of the sample's height with that after being decompressed[36]. A value of 1 means that the sample returned entirely to its initial height (elastic material) and a value of 0 indicates that the sample did not return to the initial height (viscous material). The springiness index values of the samples ranged from 0.840-0.893, which indicated that all of the tempe samples had fairly elastic properties. The cohesiveness value is directly proportional to the resilience and springiness index. The denser the mycelia, the more air cavities that form between mycelia; thus, the tempe becomes thicker. When pressure is applied, the air will be released from the cavities and when the pressure is released, the cavities between the mycelia fill up with air and the tempe returns to its original shape. Figure 3 shows that the yellow and black soybean tempe had the highest cohesiveness, resilience and springiness index. On the other hand, the groundnut and mung bean tempe had the lowest cohesiveness, resilience and springiness index values. This result was in accordance with the appearance of the mung bean tempe, which did not have thick mycelia.

Hardness is a mechanical parameter in the form of the force applied for deformation[36]. The higher the hardness value, the higher the force needed to deform the material. The mung bean tempe had the lowest hardness value compared to that of the other tempe samples. This could have been caused by the high water content of the mung bean tempe (70.60%). The texture of a material is affected by its water content[37]. A high water content causes tempe to become soft. It is known that the texture of tempe is affected by the growth of the fungus mycelia, water content, fat content and the type and amount of carbohydrates in the raw material[37].

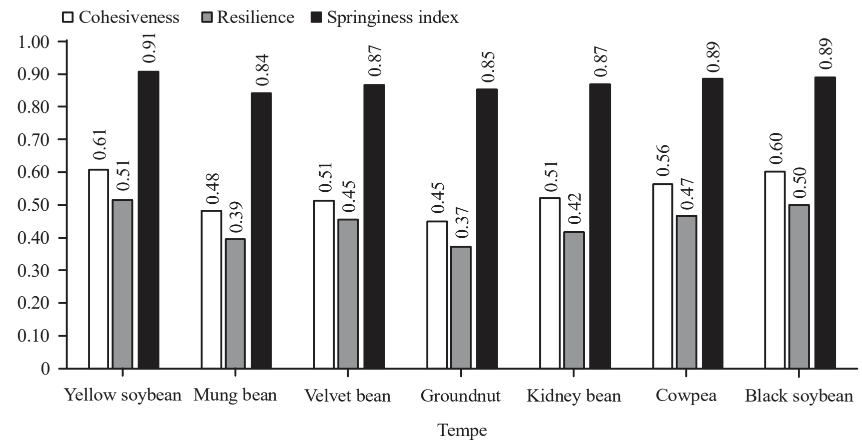

Fig. 3: Cohesiveness, resilience and springiness index of fresh tempe made from different legumes

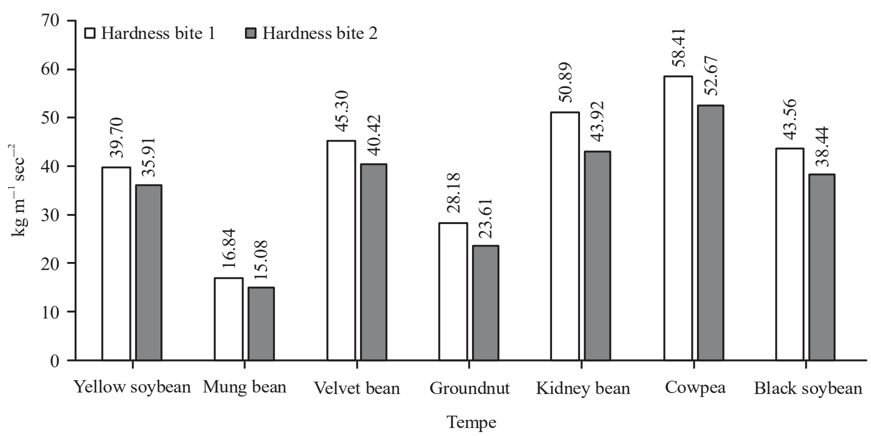

Fig. 4: The hardness of fresh tempe made from different legumes

Table 8: Color of fresh tempe made from different legumes

Tempe	L*	a*	b*
Yellow soybean	72.34 ± 2.75^{ab}	7.33 ± 0.47^{a}	20.86 ± 1.30^{a}
Mung bean	67.64 ± 1.71^{bc}	3.45 ± 2.37^{b}	18.63 ± 1.69^{ab}
Velvet bean	55.16 ± 3.21^{c}	6.31 ± 0.09^{ab}	10.72 ± 0.55^{bc}
Groundnut	70.20 ± 5.13^{abc}	5.99 ± 0.47^{ab}	15.90 ± 2.28^{abc}
Kidney bean	72.99 ± 1.50^{ab}	5.34 ± 0.43^{b}	8.37 ± 1.79^{c}
Cowpea	82.07 ± 3.57^{a}	5.72 ± 0.59^{ab}	9.67 ± 0.42^{bc}
Black soybean	66.25 ± 2.03^{bc}	6.31 ± 1.08^{ab}	18.31 ± 2.38^{ab}

Different superscripts indicate significant differences ($p<0.05$) as determined by a Kruskal-Wallis test followed by Dunn's test

Color: The color of the tempe was analyzed in fresh and fried tempe. The color of the fresh tempe is presented in Table 8. The color was evaluated as L*, a* and b* values, which correspond to brightness, green (negative) to red (positive) and blue (negative) to yellow (positive), respectively. The results showed that the cowpea tempe obtained the highest L* values, as this bean looked brighter than the others. The mung bean tempe had the lowest a* value because the mung beans had green skins that were not completely removed, while the yellow soybean tempe had the highest a* value. The yellow soybean tempe had the highest b* value, which was not much different from that of the mung bean tempe and black soybean tempe, which could have been because the soybeans, black soybeans and mung beans were more yellow than the other legumes.

The color values of all fried tempe samples are presented in Table 9. The results showed that the black soybean tempe had the highest a* value, implying that the fried black soybean tempe had a more reddish color, while the yellow soybean tempe had the largest b* value, which indicated that the yellow soybean tempe had a more intense yellow color than that of the other tempe samples. It can be seen that the L* value of all tempe samples decreased after frying due to the darker color caused by the Maillard reaction.

Sensory evaluation: Sensory evaluation is important for determining the acceptability of a product by panelists. The evaluation was conducted for both fresh and fried tempe, which are the two most common forms of tempe on the market. The overall acceptance of the fresh tempe produced from alternative legumes varied from 2.88-4.41 (Table 10). This indicated that most of the alternative soybean tempe samples were not favored by panelists. In addition, the yellow soybean tempe had the highest overall acceptability. Among the underutilized legumes, only the black soybean tempe obtained a similar score as that of the yellow soybean tempe. This was because yellow soybean tempe was more familiar to panelists compared to others. In fresh tempe, the attributes assessed were appearance and aroma. The most preferred appearance was that of the groundnut tempe because it had a compact texture, thick mycelium and fresh aroma, while the least preferred was that of the mung bean tempe because it had thin mycelium and alcohol-like aroma.

For fried tempe, the overall acceptance of the alternative soybean tempe samples was in the range of 3.03-3.93, except for that produced from black soybeans, which achieved an acceptance value of 5.13. This indicated that the panelists were between quite dislike and neutral for most of the samples. A slightly higher score than that of the fresh tempe samples was observed for the fried tempe samples, which could have been due to a decrease in the bean flavor, which is produced by lipoxygenase during the oxidation of polyunsaturated fatty acids[38]. The increase in temperature and the duration of heating have been correlated with a decrease in lipoxygenase activity[39]. Deep frying also caused the Maillard reaction, which generates products that greatly influence essential food quality attributes, such as flavor and aroma[40]. Similar to that of the fresh tempe, the highest overall acceptance value was obtained for the fried black soybean tempe. As expected, tempe made from yellow soybeans was the most preferred by panelists. For fried tempe, the attributes assessed were appearance, aroma, texture and taste. Similar to fresh tempe, the most preferred appearance was that of the groundnut and cowpea tempe samples. The most disliked appearance was that of the velvet bean tempe because it had a dark color. For aroma, the mung bean, cowpea and black soybean tempe samples had the same scores as that of the yellow soybean tempe, while the ground nut tempe had the lowest score. The low score of the groundnut tempe could be attributed to the rancid odor, as groundnut had the highest fat content among the alternative legumes. For the texture, that of the black soybean tempe was similar to that of the yellow soybean tempe. The similar texture could be explained by the size of the beans being the same; hence, they had similar density and compactness. In terms of taste, both the yellow and black soybean tempe samples obtained the highest scores. On the other hand, the cowpea and groundnut tempe samples had the lowest scores, which could be due to a lack of a tempe-like flavor in the cowpea tempe and the bitter taste of the groundnut tempe[41].

Table 9: Color of fried tempe made from different legumes

Tempe	L*	a*	b*
Yellow soybean	54.33 ± 1.42^{a}	12.91 ± 0.93^{a}	33.35 ± 1.32^{a}
Mung bean	44.22 ± 1.65^{abc}	10.04 ± 1.17^{ab}	27.92 ± 1.32^{ab}
Velvet bean	42.97 ± 1.65^{bc}	6.70 ± 1.32^{b}	16.34 ± 3.08^{c}
Groundnut	55.23 ± 3.96^{a}	9.81 ± 0.98^{ab}	28.88 ± 1.90^{ab}
Kidney bean	45.21 ± 0.91^{abc}	10.47 ± 0.57^{ab}	24.34 ± 0.79^{bc}
Cowpea	48.43 ± 3.53^{ab}	10.64 ± 1.41^{ab}	25.55 ± 3.34^{abc}
Black soybean	38.83 ± 0.64^{c}	13.75 ± 1.32^{a}	22.32 ± 1.86^{bc}

Different superscripts indicate significant differences different (p<0.05) as determined by a Kruskal-Wallis test followed by Dunn's test

Table 10: Sensory evaluation scores of fresh and fried tempe made from different legumes

	Fresh tempe			Fried tempe				
Tempe	Appearance	Aroma	Overall acceptance	Appearance	Aroma	Texture	Taste	Overall acceptance
Yellow soybean	6.13^{a}	4.97^{a}	5.53^{a}	6.02^{ab}	5.35^{a}	5.64^{a}	5.29^{a}	5.64^{a}
Mung bean	3.31^{c}	2.84^{c}	2.88^{c}	3.45^{c}	4.26^{a}	3.84^{c}	2.74^{bc}	3.16^{c}
Velvet bean	2.56^{c}	3.56^{bc}	2.94^{c}	2.26^{c}	3.80^{cd}	2.80^{c}	3.23^{bc}	3.03^{c}
Groundnut	4.88^{ab}	2.59^{c}	3.34^{bc}	5.80^{ab}	2.97^{d}	4.00^{bc}	2.39^{c}	3.19^{c}
Kidney bean	3.25^{c}	2.88^{c}	3.06^{c}	3.39^{c}	4.16^{bcd}	4.19^{bc}	3.93^{ab}	3.93^{bc}
Cowpea	3.75^{bc}	2.47^{c}	2.97^{c}	5.26^{ab}	4.68^{abc}	3.39^{c}	2.77^{bc}	3.39^{c}
Black soybean	4.63^{b}	4.84^{ab}	4.41^{ab}	4.26^{bc}	4.97^{ab}	5.26^{ab}	5.10^{a}	5.13^{ab}

Different superscripts indicate significant differences (p<0.05) as determined by a Kruskal-Wallis test followed by Dunn's test

Table 11: Various parameters determining the rank of tempe made from different legumes

Tempe	Amino acids	Essential amino acids	Umami/bitter amino acids	Protein content	Lipid content	Folate content	Fe content	Zn content	Overall acceptance	Appearance	Taste	Aroma	Texture	Average	Rank
Yellow soybean	2	1	7	2	3	7	3	4	1	1	1	1	1	2.62	2
Mung bean	6	7	3	3	1	1	7	5	3	2	2	1	3	3.38	5
Velvet bean	5	5	5	4	2	4	6	3	3	2	2	3	3	3.62	6
Groundnut	1	2	2	5	4	3	5	1	3	1	3	4	2	2.77	3
Kidney bean	4	4	4	4	1	3	2	6	2	3	1	2	2	2.92	4
Cowpea	7	6	6	6	1	5	4	7	3	1	2	1	3	4.00	7
Black soybean	3	3	1	1	3	6	1	2	1	1	1	1	1	1.92	1

Overall, the results of the chemical, nutritional, physical and sensory analyses showed that the highest score was obtained by the black soybean tempe, followed by that of the control, groundnut and kidney bean tempe samples (Table 11). The results of this study showed the superiority of underutilized legumes to yellow soybean in terms of chemical properties. However, the fate of the protein in the gastrointestinal tract has not been studied. Further studies of the *in vitro* and *in vivo* digestibility of tempe made from various underutilized legumes is needed to evaluate the protein quality. In addition, the effect of different cooking methods on the chemical, nutritional, physical and sensory properties of the tempe is of interest and should be investigated.

CONCLUSION

The results of this study suggested that, in terms of nutrition and chemical composition, the black soybean tempe was superior to that produced from other underutilized legumes and exhibited better properties than those of the yellow soybean tempe. However, the overall panelist acceptability of tempe produced from underutilized legumes was lower than that of the yellow soybean tempe. The results of this study indicate that several underutilized legumes are potential raw materials that could substitute yellow soybeans in the production of tempe. Moreover, the acceptability of these underutilized legume tempe products can be increased with education, promotion and habituation.

SIGNIFICANCE STATEMENT

This study is the first report on the characterization of seven different legumes for making tempe, including the chemical, nutritional, physical and sensory properties. The results of this study showed that underutilized legumes are a good alternative vegetable protein to yellow soybeans. This study will help the government promote the utilization of these legumes to combat growth stunting problems.

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