

Physicochemical properties and sensory acceptance of *Canavalia ensiformis* tempeh energy bar

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Abstract

The food industry needs a creative approach to innovation in order to produce revolutionary materials, innovations and fresh, nutritious, sustainable food products. Tempeh is a traditional meal prepared using *Rhizopus oligosporus* to ferment dehulled and cooked soybeans to a compact and sliceable cake. Because of their high content of proteins, carbohydrates, lipids, and minerals, energy bars are snacks offering good sensory and nutritional properties. *Canavalia ensiformis* (Kacang Koro), is an underutilised legume that contains up to 32% of protein. To our knowledge, no prior studies have studied regarding tempeh and energy bars, especially in Malaysia. This research aimed to evaluate both the physicochemical properties and sensory acceptance of the *C. ensiformis* tempeh energy bar. Energy bars of six formulations (Formulation A – E) were produced using different percentages of *C. ensiformis* tempeh namely 0%, 4.8%, 9.5%, 14.3%, 19.0% and 23.8%. The physical characteristics of the energy bar were analysed based on colour profile analysis, texture analysis and also the pH value, while the chemical characteristics were analysed based on proximate analysis, calories, mineral content, and toxicity analysis. The results showed that the energy bars consist of moisture content of 9.29-13.09%, ash of 0.99-1.56%, crude fibre of 1.82-7.27%, protein of 4.93-10.34%, crude fat of 12.36-15.97%, carbohydrate of 58.91-64.94%, and calorie content of 4627.55 - 5267.80 cal. Energy bars with 23.8% of *C. ensiformis* tempeh exhibited highest in moisture and protein, moderate in ash and fibre and showed no significance in carbohydrate and fat contents. The taste and overall acceptability indicate that formulation A showed the best acceptance among the prepared formulations. The utilization *C. ensiformis* tempeh could diversify the usage of *C. ensiformis* in the food industry, hence promoting their application.

1. Introduction

Tempeh is a traditional fermented soy food, prepared from soaked and cooked soybeans by salt-free aerobic fermentation using *Rhizopus oligosporus* (Krisnawati and Adie, 2015). Freshly prepared tempeh is a cake-like substance that is fully coated and filled with white mycelium, with a clean, yeasty odour (Xiao, 2011). Tempeh is usually cooked before it is eaten, and preparation usually involves frying, deep-fat frying, or baking the product (Wilson, 1995). Tempeh is considered as a good source of protein, vitamins, antioxidants, phytochemicals, and other bioactive

beneficial substances (Mani and Ming, 2017).

Canavalia ensiformis or Kacang Koro contain more than 32% protein, yet they also produce somewhat mild antinutrient content (protease inhibitors, lectins, saponins and tannins) in the form of anti-nutritional factors (Eke *et al.*, 2007) which makes them not popular as human food. Therefore, few treatments have been conducted to render *C. ensiformis* edible to humans, such as heating, fermentation and extrusion, though obtaining favourable results (Tepal *et al.*, 1994).

The concept behind energy bars is to provide anyone on the go with a quick snack or breakfast (Zhu and

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Labuza, 2010). The energy bar is a convenient and healthy ready to eat food that supplies balanced nutrients which contain protein, fat, minerals, vitamins and carbohydrates (Ho *et al.*, 2016). Chitkara *et al.* (2017) reported that the primary purpose of energy bars is to satisfy hunger, replace a meal, and provide essential nutrition. The current market's energy bars are filled with a lot of fruit, nuts, granola, and sugar combinations (Norajit *et al.*, 2011) to increase the product's sugar, protein, and fibre content. The consumers are concerned about getting healthier foods, and this has changed their eating habits which promoted growth in the energy bar market (Silva *et al.*, 2016). Energy bar provides an effective sensory and nutritious snack because of their high carbohydrate, protein, lipid, and mineral content (Nadeem *et al.*, 2012). Athletes and other physically active people also use energy bars to fulfil their nutritional needs, since they can have high amounts of protein, fats and carbohydrates (Norajit *et al.*, 2011).

The protein content in energy bars was usually derived from soybeans and milk products as well as from other protein sources, such as peanut butter, nuts, to increase the protein quality (Aldrich, 2015). The development of the new product based on a 'modified version' of underutilised legumes such as *C. ensiformis* could produce a better form of energy bar hence increasing the usage of these underutilized legumes. The purpose of this research was therefore to evaluate the effect of *C. ensiformis* tempeh on the physicochemical properties and the sensory acceptability of the *C. ensiformis* energy bar.

2. Material and methods

2.1 Raw materials

C. ensiformis was harvested from Kuala Berang, Terengganu. The starter tempeh was obtained from Teluk Panglima Garang, Selangor. Other ingredients used in the production of *C. ensiformis* tempeh energy bar include butter, raisins, honey, and brown sugar which were purchased in Kuala Terengganu, Terengganu, Malaysia.

2.2 Preparation of *Canavalia ensiformis* tempeh

C. ensiformis tempeh was prepared according to the procedure described by Kustyawati *et al.* (2017) with some modification. *C. ensiformis* beans were washed and soaked in clean tap water at room temperature (28°C) for 48 hrs and the soaking water was changed 5 times, and then boiled for 30 mins (the ratio of water to *C. ensiformis* was 3:1). This was followed by dehulling to remove *C. ensiformis* skin from the cotyledon manually, and another boiling for 30 mins (Wan Mohamad Din *et al.*, 2020). Subsequently, *C. ensiformis* was boiled,

drained and air-dried. Then, 0.02 g of *Rhizopus* mold was inoculated for every 100 g cooked *C. ensiformis*, and packed into plastic bags, then incubated for 36 hrs at 32°C. The prepared tempeh was then kept in 4°C prior to further usage.

2.3 Preparation of *Canavalia ensiformis* tempeh energy bar

All the ingredients were weighed based on the formulations presented in Table 1. The sliced '*C. ensiformis* tempeh' was then roasted in the oven for 1 hr at 100°C. The *C. ensiformis* tempeh was then grind to small size using mortar and pestle. Prior to the mixing process, almonds were roasted for 10 mins in the oven, while oats also were baked for 10 mins. All the ingredients were cut to small sizes before mixing in a bowl. Brown sugar, honey, and butter were heated at low temperature (40°C) and then mixed with roasted ingredients. The prepared mixture was then pressed to produce a rectangular shape in a mould. The energy bar was then kept in the chiller (4°C) prior to further analysis (Wan Mohamad Din *et al.*, 2020).

Table 1. The formulation and ingredients of *C. ensiformis* tempeh energy bar

Ingredients	Formulation					
	Control	A	B	C	D	E
Honey (g)	15	15	15	15	15	15
Almonds (g)	25	20	15	10	5	0
<i>C. ensiformis</i> tempeh (g)	0	5	10	15	20	25
Raisin (g)	10	10	10	10	10	10
Brown sugar (g)	20	20	20	20	20	20
Instant oat (g)	10	10	10	10	10	10
Butter (g)	15	15	15	15	15	15
Total	105	105	105	105	105	105

2.4 Colour profile analysis

The instrument that was used to measure the colour was Konica Minolta colourimeter (Konica Minolta, Tokyo, Japan) based on L*a*b* colour system, where L (lightness), a (redness) and b (yellowness). The colourimeter was calibrated by using a white calibration plate before analysis. The instrument was placed on the energy bar formulation and the values of L* (lightness), a* (redness) and b* (yellowness) for each energy bar was recorded and analysed (Zainol *et al.*, 2020).

2.5 Texture profile analysis

The analysis was conducted using a TA.XT.Plus texture analyzer (Stable Microsystems, UK) to determine the hardness, and factorability of the energy bar based on

the mechanical characteristics where material was subjected to a controlled force from a deformation curve of response. The sample was placed centrally under the 3-point bend rig probe until the probe came to contact with the sample. Then, the deformation curves were recorded (Mamat *et al.*, 2018).

2.6 pH analysis

The acidity and alkalinity of the energy bar samples were determined using a pH meter (Mettler Toledo, Ohio, USA). The samples (5 g) were finely mixed and homogenized with 20 mL of distilled water, and measured the pH value.

2.7 Toxicity analysis

2.7.1 Determination of oxalate content

Pulverized sample (1 g) was weighed into a conical flask, and added to 75 mL of 3 M sulphuric acid was added and stirred with a magnetic stirrer for an hour. This was filtered and 25 mL aliquot of the filtrate was collected and heated to 80-90°C. This filtrate was kept above 70°C at all times. Next, the hot aliquot was titrated against 0.05 M of potassium permanganate oxide (KMnO₄) until an extremely faint pale pink colour persisted for 15-30 s (Agbaire, 2011). The oxalate content was calculated as followed: 1 mL of 0.05 M of KMnO₄ = 2.2 mg oxalate.

2.7.2 Determination of phytic acid content

Sample (2 g) was weighed into a 250 mL conical flask. Approximately 100 mL of 2% HCl was used to soak the sample for 3 hrs and then filtered through filter paper. A 25 mL aliquot of the filtrate was placed in a separate 250 mL conical flask and 5 mL of 0.3% ammonium thiocyanate solution was added. Approximately 53.5 mL of distilled water was added and this will be then titrated with standard iron (III) chloride solution which contained 1.95 mg of iron per mL until a brownish yellow colour persisted for 5 min (Unuofin *et al.*, 2017). Phytic acid was calculated as follow: Phytic acid (%) = Titre value × 0.00195 × 1.19 × 100.

2.7.3 Determination of tannins content

Tannin content was determined using the Folin-Ciocalteu method. One millilitre of energy bar sample was mixed with 7.5 mL of distilled water and 0.5 mL of Folin-Ciocalteu phenol reagent. Then, 1 mL of 35% sodium carbonate solution was added to the mixture and diluted to 10 mL with distilled water. The mixture was vigorously shaken and let it stand for 30 min at room temperature (25°C). Absorbance for test and standard solutions were measured against blank at 725 nm with a UV/Visible spectrophotometer. The tannin content was

expressed in terms of mg of GAE/L of extract (Wan Mohamad Din *et al.*, 2020).

2.8 Proximate analysis

The moisture, ash, fat, fibre and crude protein content of the *C. ensiformis* tempeh energy bar were determined according to AOAC International (2007) standard procedures. All analyses were carried out in triplicates.

2.9 Calorie content analysis

The calorie content was determined using a bomb calorimeter (LECO, USA). The sample was placed in the crucible in the combustion chamber for the combustion process. Once the sample is ignited, a thermometer which is partially submerged in the water records the temperature changes that occur. The heat of combustion (cal/g), the change in temperature was recorded as the calories content of the sample. (Wan Mohamad Din *et al.*, 2020).

2.10 Mineral content analysis

The determination of mineral nutrient was carried out using an Inductively Coupled-Plasma-Mass Spectrophotometry (ICP-MS) (Agilent Technologies, U.S.A). Dried sample (2 g) was placed into the crucible, burned in a furnace at 500°C for 24 hrs. The crucible was then cooled at room temperature and 2 mL of concentrated HCl was added and was let to evaporate to dryness on a hot plate. Then, 10 mL of 20% HNO₃ was added and the crucible then placed in the water bath for 1 hr. Finally, the solution was analysed using ICP-Mass Spectrophotometer.

2.11 Sensory analysis

The sensory evaluation session was performed based on a 7-point hedonic scale (higher score indicates better quality attributes (1, dislike very much and 7, like very much)) (Mamat *et al.*, 2018). The colour, aroma, appearance, crispiness and flavour of the energy bars were evaluated. All the attributes were independently judged by forty untrained panels based on their likeness. The sample was packed and coded with a 3-digit code. The mean score for each attribute was reported.

2.12 Statistical analysis

All data were analysed statistically using SPSS (Version 20) statistical software package. The results were expressed as mean ± standard deviation. The significant difference at (p < 0.05) was performed by one-way analysis of variance (ANOVA) and Fisher's Least Significant Difference (LSD) test (Hau *et al.*, 2018).

3. Results and discussion

3.1 Colour profile analysis

Table 2 reveals that the L* value of the formulations was also increased with the increasing amount of *C. ensiformis* tempeh, suggesting the influence of tempeh on the lightness of the energy bar of colour. The findings also showed that formulation A had the highest a* rating, which shows the energy bar's redness. Formulation E, on the other hand, has the lowest value of a* (2.67). The energy bar's red colour may be attributed to the amounts of almond in the formulation as well as the presence of other almond ingredients. The formulation D's b* value (yellowness) was found to be the highest (25.23), while formulation A was found to be the lowest (20.62). For the other tests, the b* value was identical, suggesting that the butter mixture might be other ingredients that could have led to an increased value in yellowness of energy bar. The energy bar colours are quite similar to each formulation due to the colour of its ingredients, while the amount of *C. ensiformis* tempeh and the amount of almonds gives a small effect to the bars.

3.2 Texture profile analysis

Table 2 shows that the hardness of the *C. ensiformis* energy bar (formulation D) was found to be significantly the highest (7.71±0.37 N) while the control formulation had the lowest hardness value (6.44±0.73 N). In general, the low hardness of the formulation was observed when a low amount of tempeh was incorporated into the formulation. This could mean that when the proteins are used, the bar may have a better or worse texture than expected (Imtiaz et al., 2012). In addition, all formulations showed a somewhat similar fracturability value indicating that the tempeh did not have an analytical effect on the texture profile value. However, due to the low amount of tempeh incorporated in the formulations, low fracturability was observed in the analysis. When proteins are blended together, they may have synergistic or antagonistic effects. Moisture migration, phase separation (Hogan et al., 2012) and fat oxidation could be the main reason for the texture value recorded in this study (Dan and Labuza, 2010). Mridula et al. (2013) stated that the hardness of the energy bar is significantly affected by the level of sweeteners and

flaxseed.

3.3 pH value Analysis

Table 3 shows the pH value of *C. ensiformis* tempeh energy bar ranges between 5.30 to 4.82 and the control energy bar was 5.44. The data reveal that the acidity of all formulation of energy bar was significantly different ($p < 0.05$) between control, formulation A, B and C except for formulation D and E. Control sample, illustrated the highest pH value compared to other formulations (5.44±0.01), while formulation E exhibited the lowest pH value (4.82). According to Silva et al. (2016), the pH value of the energy bar incorporated with jeriva flour ranged from 6.78 to 6.92, where the pH of jeriva flour itself is 4.96. Compared to *C. ensiformis* tempeh energy bar, the pH value obtained was acidic. The ingredient that probably caused the pH above 5 is honey. This could be because the average pH of honey is 3.9, with a typical range of 3.4 to 6.1 (White et al., 1962).

3.4 Toxicity analysis

3.4.1 Oxalate content

Table 3 shows the oxalate content of *C. ensiformis* tempeh energy bar formulation and control. The results showed that there was a significant difference ($p < 0.05$) between formulation D and E except for formulation control A, B and C which showed no significant difference. The range of oxalate content of *C. ensiformis* tempeh energy bar was from 45.43 mg to 54.90 mg while the formulation control was 45.10 mg. The formulation E has the highest oxalate content which was 59.4 mg, meanwhile formulation A has the lowest which was 45.43 mg. Such a finding could be explained by the increasing amount of *C. ensiformis* tempeh in the energy bar which increased in the oxalate content. Fermentation also caused the changes of some organic acids such as acetic acid, oxalic acid, citric acid and succinic acid, which eventually influenced the pH. This finding was in agreement with previous studies on soybean fermentation using *R. oligosporus* starter (Vong et al., 2018) or *Rhizopus* starter (Moa et al., 2013). A possible explanation for these results may be the fermentation of *C. ensiformis* helps to reduce the amount of oxalate in the legume. Besides, fermentation helps to reduce the

Table 2. The colour profile analysis for (L* a* b*), hardness and fracturability of the *C. ensiformis* tempeh energy bars

Formulation	L*	a*	b*	Hardness (N)	Fracturability (mm)
Control	49.42±1.05 ^{bc}	5.33±0.41 ^a	24.18±0.24 ^{ab}	6.44±1.73 ^b	44.66±1.49 ^b
A	47.52±4.00 ^c	5.37±0.39 ^a	20.62±4.06 ^a	6.50±0.94 ^b	45.80±0.27 ^{ab}
B	50.70±3.20 ^{bc}	4.75±0.16 ^{ab}	24.52±1.99 ^{ab}	6.55±0.78 ^b	45.28±0.73 ^{ab}
C	52.94±0.43 ^{ab}	4.53±0.16 ^b	25.18±1.00 ^a	6.83±0.99 ^b	46.82±0.65 ^a
D	55.44±1.15 ^a	3.45±0.67 ^c	25.23±1.28 ^a	7.71±0.37 ^a	45.14±1.06 ^{ab}
E	52.22±4.79 ^{bc}	2.67±0.58 ^c	21.38±2.37 ^{ab}	7.47±0.01 ^{ab}	44.96±0.03 ^{ab}

Mean±standard deviation values with different superscript within the column are significant different at $p < 0.05$

Table 3. The pH values and anti-nutrients content in different formulation of *C. ensiformis* tempeh energy bars

Formulations	pH	Oxalate (mg)	Tanins (%)	Phytic Acid (%)
Control	5.44±0.02 ^a	45.10±0.45 ^d	0.01±0.001 ^d	1.93±0.10 ^b
A	5.30±0.07 ^b	45.43±0.25 ^d	0.01±0.001 ^d	2.50±0.05 ^a
B	5.22±0.06 ^c	45.75±0.29 ^{cd}	0.001±0.0001 ^c	2.37±0.12 ^a
C	4.85±0.07 ^d	48.73±0.42 ^c	0.02±0.001 ^c	2.00±0.15 ^b
D	4.93±0.09 ^c	54.66±0.32 ^b	0.02±0.001 ^b	1.53±0.13 ^c
E	4.832±0.09 ^c	59.41±0.38 ^a	0.03±0.001 ^a	1.17±0.07 ^d

Mean±standard deviation values with different superscript within the column are significant different at $p < 0.05$

anti-nutritional and toxic factors in the raw materials by making the proteins and minerals which are complex with these phytochemicals readily available (Adegbehingbe, 2015).

3.4.2 Phytic acid content

The results showed that there was a significant difference ($p > 0.05$) between formulation except for formulation control and C which has no significant difference similar to formulation A and B which also showed no significant difference (Table 3). The range of phytic acid content *C. ensiformis* tempeh energy bar was from 2.50% to 1.17% while the formulation control was 1.93%. The formulation A contained the highest tannin level which was 2.50%, meanwhile formulation E contained the lowest which was 1.17%. This may be due to the fact that the increased percentage of *C. ensiformis* tempeh in the energy bar reduced the amount of phytic acid content in the energy bar (Sridhar and Seena, 2006).

3.4.3 Determination of tannins content

Table 3 shows that there was a significant difference ($p < 0.05$) between all formulations except for control and formulation A. Formulation E has the highest tannins content (0.03%), while formulation C has the lowest (0.00%). The highest content of tannins in formulation E still considers a safe percentage of tannins to be consumed. This is because high dietary levels (approximately 5%) can cause death (Sridhar and Seena, 2006). The level of content of tannins in the *C. ensiformis* tempeh energy bar is, however, below consumable level and can still be eaten.

3.5 Chemical composition of *Canavalia ensiformis* tempeh energy bar

Table 4 indicates the moisture content of the energy bar *C. ensiformis* tempeh varying from 9.37% to 13.09% where the maximum moisture content was 13.09% and the lowest moisture content of the energy bar *C. ensiformis* tempeh was 9.37%, with just 9.29% moisture content in the control energy bar. Increasing the amount of *C. ensiformis* in the formulation thereafter increases the amount of moisture content in the energy bar of *C. ensiformis*. Similar findings were reported by Arbaje et al. (2016), who stated that granola bars moisture content was affected by the amount of granola in the formulations. The highest ash content of *C. ensiformis* tempeh energy bar was recorded in formulation A (1.48%) while the lowest percentage of ash content was found in the control sample. This shows that the addition of *C. ensiformis* tempeh in the formulation reduced the ash content in *C. ensiformis* tempeh energy bar. This may be due to the usage of the nuts, where the almonds itself contains higher ash content, which was 3.13% (National Germplasm Resources Laboratory, 2009). The ash content in this study could be traced to the nuts used, which could be considered as origin when minerals, such as calcium, iron and magnesium, are used (Fernandes et al., 2010).

The highest protein content was found in formula E (10.34%), while the lowest protein content was found in formula B (4.93%). There was no significant difference between formulations A, C and D (5.34%), (5.25%) and (5.95), respectively. The higher value of protein content

Table 4. The chemical composition of *C. ensiformis* tempeh energy bar was analysed in term of moisture, ash, protein, crude fat, crude fibre and carbohydrate.

(%)	Control	A	B	C	D	E
Moisture	9.29±0.24 ^c	9.37±0.22 ^c	9.50±0.01 ^c	11.83±0.64 ^b	12.07±0.02 ^b	13.09±0.29 ^a
Ash	1.56±0.03 ^a	1.48±0.19 ^{ab}	1.36±0.03 ^b	1.29±0.01 ^b	0.99±0.02 ^c	1.00±0.01 ^c
Fat	15.03±0.65 ^a	12.64±0.08 ^a	15.9±5.88 ^a	12.36±1.70 ^a	13.41±0.26 ^a	12.63±0.6 ^a
Protein	6.26±0.69 ^b	5.34±0.88 ^{bc}	4.93±0.03 ^c	5.25±0.22 ^{bc}	5.95±0.33 ^{bc}	10.34±0.10 ^a
Fibre	7.27±2.78 ^a	7.60±1.76 ^a	9.33±1.27 ^a	6.90±2.39 ^{ab}	2.65 ^{bc} ±0.12	1.82±0.00 ^c
Carbohydrates	60.59±2.6 ^a	63.58±2.32 ^a	58.91±7.14 ^a	62.37±3.65 ^a	64.94±0.46 ^a	61.12±0.84 ^a

Mean±standard deviation values with different superscript within the row are significant different at $p < 0.05$

in energy bars may be due to the utilization of *C. ensiformis* tempeh in the energy bar production which contains higher protein compared to almonds, which was 34.47% and 21.06% respectively (Nimenibo-Uadia, 2017).

Table 4 also depicts that there was no significant difference ($p>0.05$) in the fibre content between the sample tested. The crude fibre in *C. ensiformis* recorded in this study was higher than that of soybean (4.28%) and kidney beans (4.2%) (Ameta and Ologhobo, 1994). The observed increase in crude fibre could be attributed to the presence of tempeh of *C. ensiformis* which provides more fibre than the *C. ensiformis* itself. The results also showed that there was no significant difference ($P>0.05$) in carbohydrate content between all formulations. The range of *C. ensiformis* tempeh energy bar was from 58.91% to 64.94%, while the carbohydrate content of control energy bar formulation was 60.59%. The carbohydrate source might be from the usage of brown sugar and honey in the production of *C. ensiformis* tempeh energy bars (E'zzati, 2019).

3.6 Calorie content analysis

Table 5 shows that there was a significant difference in the total caloric value between all formulations. The total calorie content of the energy bars ranged from 4742.5 cal to 5267.80 cal. Control sample contained significantly the highest calorie content (5276.80 cal) while formulation E exhibited the lowest calorie value (4627.55 cal). The decreasing trend of calorie value of the energy bar could be due to the reduced amount of cereals and nuts used in the bars (Okoye and Eke-Ejiofor, 2018).

3.7 Mineral content analysis

Table 5 also shows the mineral content that presents in five formulations of *C. ensiformis* tempeh energy bars and control. The data showed that Iron (Fe), Magnesium (Mg), Calcium (Ca), Phosphorus (P), and Potassium (K). Mineral level range in *C. ensiformis* tempeh energy bar was 25.32 – 58.63 mg/kg for potassium, 9.26 – 13.89 mg/kg for calcium, 35.55 – 83.44 mg/kg and 8.91 – 18.24 mg/kg of magnesium. The iron level shows quite

similar between each formulation which was in the range 1.00 – 2.82 mg/kg in 10 mL solution of sample. This shows that iron levels are not affected by an increasing amount of *C. ensiformis* tempeh and a decreasing amount of almonds. The mineral content in the energy bars was typically lowered as the amount of *C. ensiformis* tempeh increased. This result is in line with the study by Ho *et al.* (2016), which stated that ash content of energy snack bars is reduced when glutinous rice content increases banana puree as the main ingredient.

3.8 Sensory analysis

Table 6 showed that there was some variation yet no significant difference ($p>0.05$) between all formulations. The highest value of sensory colour acceptance was formulation B and the lowest colour acceptance was exhibited in formulation E. This might be due to the ingredients such as the amount of brown sugar, almond and butter that provide colour to the energy bar used in the same amount. According to Khouryieh and Aramouni (2012), changes in flaxseed bar colour may be associated with the possibility of browning Maillard reactions between flaxseed protein and other ingredients such as honey and brown sugar. Ho *et al.* (2016) demonstrated that the bright brown colour of the energy snack bar may be attributed to caramelisation occurring with the presence of sugar during heating and boiling.

Formulation B exhibited the highest value for odour acceptance while the lowest was found in formulation E. The caramelization process that occurs on brown sugar may difficult the panel to distinguish the odour unless the smell of the *C. ensiformis* tempeh was strong. Ho *et al.* (2016) stated that the strong odour of the “energy” snack bar might be attributed by the caramelization that occurred during the heating and boiling process with the presence of sugar. Formulation A showed to the best hardness liked by the untrained panellist with a value of 5.30. Meanwhile, formulation E was not favoured by the panellist. Brahim *et al.* (2017) reported that the range of hardness for flaxseed energy bar was from 30 N to 40 N. Compared to *C. ensiformis* tempeh energy bar, the hardness for formulation A was not in the range. This

Table 5. Calories, mineral content of the different formulations of *C. ensiformis* tempeh energy bar

Formulations	Calorie content (Cal)	Fe	Ca	P	K	Mg
Control	5267.80±19.09 ^a	1.15±0.04 ^c	13.89±0.35 ^a	61.38±0.56 ^b	58.63±0.18 ^a	18.24±0.45 ^a
A	5058.30±114.2 ^b	2.45±0.09 ^b	10.57±0.52 ^{bc}	83.44±3.12 ^a	48.63±0.44 ^b	13.83±0.28 ^b
B	4985.95±82.8 ^b	1.00±0.03 ^d	10.30±0.20 ^c	46.76±0.08 ^c	49.07±0.22 ^b	11.39±0.08 ^c
C	5002.55±73.1 ^b	1.12±0.04 ^c	9.26±0.24 ^d	42.33±0.56 ^d	41.86±0.59 ^c	11.54±0.30 ^c
D	4742.5±48.79 ^c	2.82±0.02 ^a	11.10±0.51 ^b	83.16±0.14 ^a	33.46±0.11 ^d	13.92±0.14 ^b
E	4627.55±75.7 ^c	1.10±0.04 ^c	9.32±0.08 ^d	35.55±0.35 ^c	25.32±0.19 ^c	8.91±0.11 ^d

Mean±standard deviation values with different superscript within the column are significant different at $p<0.05$

Table 6. Sensory analysis of the different formulations of *C. ensiformis* tempeh energy bar

Formulations	Colour	Odour	Hardness	Fracturability	Flavour	Overall acceptance
Control	5.70±0.04 ^a	5.60±0.45 ^a	5.35±0.36 ^a	5.38±0.14 ^a	5.85±0.41 ^a	5.83±0.35 ^a
A	5.55±0.09 ^{ab}	5.45±0.25 ^a	5.30±0.18 ^a	5.28±0.42 ^a	5.68±0.16 ^{ab}	5.48±0.33 ^a
B	5.58±0.03 ^{ab}	5.48±0.29 ^a	5.18±0.38 ^a	5.03±0.22 ^{ab}	5.55±0.33 ^{ab}	5.48±0.23 ^a
C	5.00±0.04 ^c	5.15±0.42 ^{ab}	4.40±0.26 ^b	4.38±0.35 ^{bc}	5.13±0.23 ^{bc}	4.83±0.17 ^b
D	5.15±0.02 ^{bc}	4.73±0.32 ^{bc}	4.28±0.19 ^b	4.48±0.23 ^c	4.78±0.34 ^c	4.58±0.24 ^b
E	4.83±0.04 ^c	4.20±0.38 ^c	3.90±0.32 ^b	3.98±0.09 ^c	3.65±0.14 ^d	4.05±0.22 ^c

Mean±standard deviation values with different superscript within the column are significant different at $p < 0.05$

study also showed that the degree of likeness for *C. ensiformis* tempeh energy bar decreased as the hardness decreased.

The results also showed no significant difference ($p < 0.05$) in flavour acceptance between all samples except formulation E. The results was affected by the different amount of *C. ensiformis* tempeh incorporated in the product. However, formulation A showed the best formulation of *C. ensiformis* tempeh energy bar liked by the panellists while formulation E was the least.

The colour, odour, hardness, fracturability and taste, the panellist selected the most agreed formulation A compared to other formulations. This might be due to the different percentage of *C. ensiformis* in the formulation that affected the characteristics of the energy bar.

4. Conclusion

This study exhibited that *C. ensiformis* tempeh may serve as an excellent candidate for the development of an alternative energy bar using rich protein raw materials which have been under exploited. Formulation A was mostly accepted by the panellist. This product is safe to be consumed as the level of the toxicity is still within a safe range. This study shows that *C. ensiformis* tempeh can be used in energy bar production, thereby encouraging its use in the food industry.

Conflict of interest

The authors declare that there is no conflict of interest.

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