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Research Article

Commercial development of red kidney bean tempeh

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Abstract

The objectives of this research were to produce red kidney bean tempeh and to investigate suitable packaging for the finished product. This work was undertaken to develop a tempeh based product that would be available for the snack foods market. Red kidney bean tempeh was developed and the physical characteristics and chemical composition were investigated. The finished tempeh was packed in three different types of packaging; nylon A, nylon B and metal foil then stored at room temperature for 3 months. Finally, the red kidney bean tempeh from three different packaging materials was subjected to sensory evaluation at 1 month intervals. From this experiment, it is apparent that red kidney bean makes a good material for tempeh. It looks like the original tempeh, but is darker in colour. Fried red kidney bean tempeh, packed in three different materials, changed in physical and chemical properties during storage. However, the property changes in fried tempeh kept in the nylon B container occurred slowly, especially acid value and crispness. It is concluded that nylon B is the more suitable material for packaging fried red kidney bean tempeh.

Keywords: snack food, packaging, inoculums, *Rhizopus oligosporus, Phaseolus vulgaris,* Thailand

Introduction

Tempeh is a traditional Indonesian fermented food made from soybeans through fermentation with *Rhizopus oligosporus*. When thinly sliced and deep fried in oil, tempeh obtains a crispy golden crust making it suitable for marinades, as a snack or staple food [1]. The fermentation

process of tempeh completely transforms the soybeans to produce a new flavour, aroma, texture and also increases the nutritional value and digestibility [2]. Tempeh also contains strong antioxidants which are highly reactive free radicals with oxygen [3]. There are more creative ways of cooking tempeh. Cooked tempeh can be eaten alone, or used in chili, stir fries, soups, salads, sandwiches and stews [4]. Recently, tempeh has attracted increasing interest for researchers and work has been undertaken on the development of seed inoculums for tempeh-based snack food [4] and growth of lactic acid bacteria and *Rhizopus oligosporus* during barley tempeh production [5]. Traditional tempeh is usually made from soybean but it can also be made from other substrates. Chompreeda, *et al* [6], studied a fermentation process for making tempeh from groundnuts and evaluated the acceptability of the final product. Many kinds of beans are extensively cultivated in Thailand, but, apart from soy, none of them have yet been used for tempeh production. The objectives of this research were to produce red kidney bean tempeh and to investigate the suitable packaging.

Materials and Methods

Rhizopus oligosporus MSCMU 2001 was obtained from the Faculty of Agro-Industry at Chiang Mai University, Thailand. Red kidney beans (*Phaseolus vulgaris* L.) cv. Kampangsaen 60 were purchased from a local market in Phitsanuloke province. Sticky rice was also purchased from a local market. Bacto peptone (Merck), sodium hydroxide (Ajax Finechem), methyl red (Fluka), Sulphuric acid (Merck) and potassium hydroxide (Ajax Finechem) were purchased from the companies indicated.

Preparation of tempeh starter

The inoculums were prepared from pure culture of *Rhizopus oligosporus* MSCMU 2001 grown on potato dextrose agar (PDA) plates. The pure culture was sub-cultured periodically on PDA slant as stock cultures. The tempeh starter was prepared by the process shown in Figure 1.

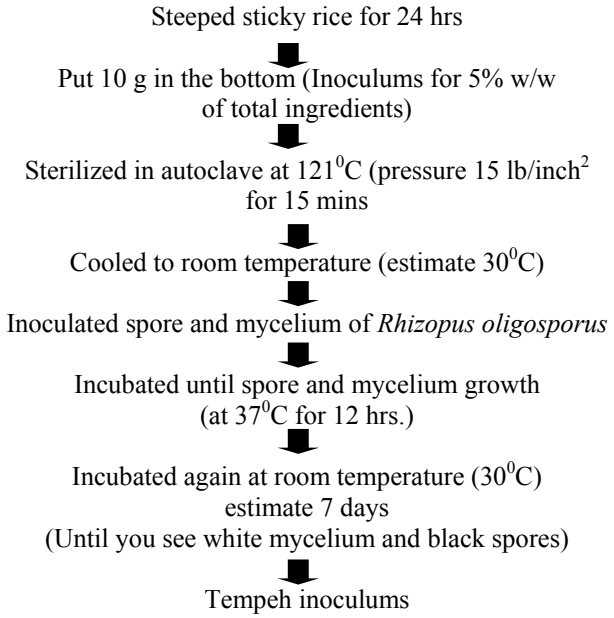

Figure 1. The process of tempeh starter production (adapted from Krusong [2])**.**

Tempeh process

Red kidney bean was used as substrate for tempeh production as shown in Figure 2.

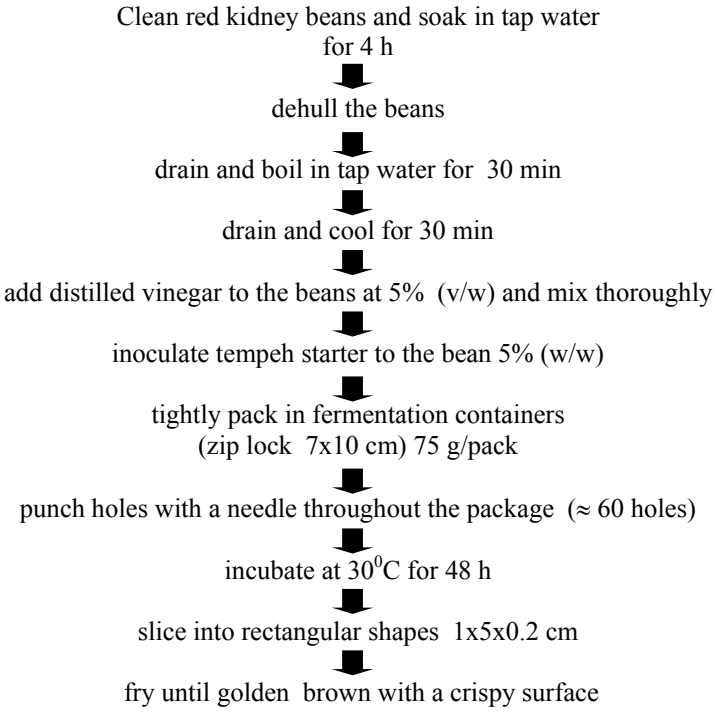

Figure 2. Process of red kidney bean tempeh production (adapted from Chompreeda [6]).

Proximate analysis of fried red kidney bean tempeh

The following AOAC [7] methods were used to determine proximate composition: drying at 105^{0}C for 24 h for moisture (method 925.098); incineration at 550^{0}C for ash (method 923.03); defatting in a Soxhlet apparatus with petroleum ether for crude fat (method 920.39C); microkjeldahl for crude protein (N× 6.25) (method 960.52); and determined gravimetrically after chemical digestion and solubilization of other materials present. The fibre residue weight is then corrected for ash content after ignition carbohydrate content was estimated by difference.

Colour and texture analysis

The surface colour of samples was measured as colour values, (L*,a*,b*) using a Hunter Lab Colorflex® 4510 (Hunter Association Laboratory, Inc. USA).

The crispiness of samples was measured using a texture analyzer, TA.XT. plus (CHAPA TECHCENTER, Stable Micro Systems Ltd. UK).

Water activity (a_w)

5 g of ground samples was measured for a_w using a hygrometer Aqua Lab model CX3TE Series 3 (CHAPA TECHCENTER Stable Micro Systems Ltd. UK).

Determination of optimal packaging for storage

In the experimental design, fried red kidney bean samples were packaged in centre-seal pillow preformed bags made from three different materials, being nylon A, nylon B and metal foil. The shelf life was determined by monitoring colour values (L*,a*,b*), textural characteristics, peroxide value [7] and acid value (AV) [7] in fried red kidney bean tempeh samples stored at room temperatures for 0, 1, 3, 5, 7, 9, 11, 14, 21 and 28 days for the first months storage, 7, 14, 21 and 28 days for the second months storage and 10, 20 and 30 days for the third months storage.

Sensory evaluation

Four pieces of fried tempeh in three different packaging materials were served to 30 panelists along with a questionnaire for evaluation. The sensory evaluation was carried out by using a 9 point hedonic scale [8], given numerical values of 1 (dislike extremely) through to 9 (like extremely). The parameters assessed included the flavour, colour, texture and overall acceptability.

Results and Discussion

Tempeh process

Fresh red kidney bean tempeh after 48 h fermentation looked much like soy bean tempeh (Fig. 3). It was considered to be a good tempeh because the beans were knitted together to form a firm, dense, chewy cake by a mat of white mycelia with a mild ammonia smell and no grey or black patches of spores forming on the surface (Fig 4A). The appearance of spores is not harmful but gives an unacceptable appearance [4]. Tempeh was easily sliced to thin pieces because of the firm texture. After deep frying, the red kidney bean tempeh obtained a crispy golden crust (Fig. 4B).

Figure 3. Growth of *Rhizopus oligosporus* on red kidney bean tempeh.

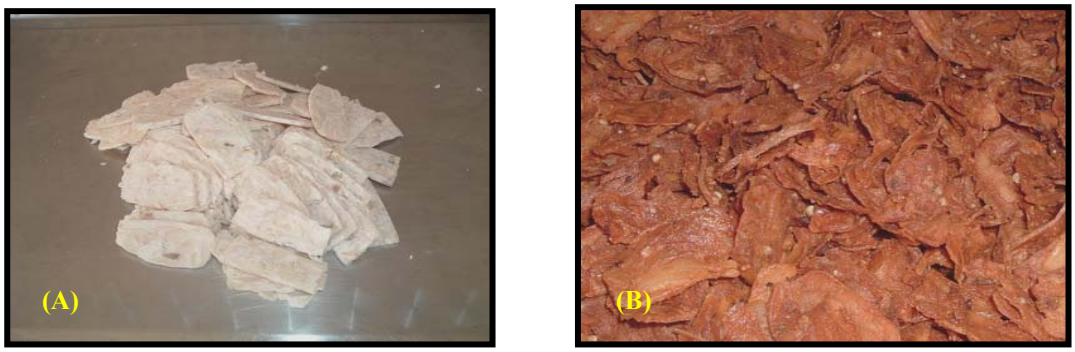

Figure 4. Fresh red kidney bean tempeh (A) and (B) fried red kidney bean tempeh.

Physical properties and chemical composition

The results of examining some physical properties and the chemical composition of red kidney bean tempeh are shown in Tables 1 and 2 respectively.

Table 1. Physical properties of red kidney bean tempeh.

Sample	Colour			Texture (gramforce)
	L*	a*	b*	
Fried red kidney bean tempeh	23.90±1.29	7.65±0.83	7.80±0.59	299.50±1.45

Table 2. Chemical composition of red kidney bean tempeh.

Chemical composition	Fried red kidney bean tempeh
MC (%)	5.02 ± 0.00
Protein (%)	16.61 ± 1.11
Fibre (%)	0.21 ± 0.03
Ash (%)	7.10 ± 0.17
Fat (%)	21.32 ± 1.56
CHO (%)	49.74 ± 0.00
a_w	0.30 ± 0.00

Following frying, the colour of tempeh as indicated by L*, a* and b* (Table 1) became a darker brown and its water activity was 0.3 (Table2). Frying is a unit operation which is mainly used to alter the eating quality of a food. A secondary consideration is the preservative effect that results from thermal destruction of microorganisms and enzymes, and a reduction in water activity at the surface of the food (or throughout the food, if it is fried in thin slices). When food is placed in hot oil, the surface temperature rises rapidly and water is vaporised as steam and a crust is formed. The surface crust has a porous structure, consisting of different-sized capillaries. During frying, both water and water vapour are removed from the larger capillaries first and replaced by hot oil. This explains why fried tempeh has a high fat content. Moisture content, protein, fat and carbohydrate of dried red kidney bean were 12.1%, 20.3%, 1.2% and 62.7%, respectively [9] and were compared to the chemical composition of fried red kidney bean tempeh. It was observed that there was no large difference in protein content between fried tempeh and dried red kidney

bean. Actually, both chemical compositions should not be compared because one was from a frying process, while the other was not. Unfortunately, the chemical composition of fresh tempeh was not shown. However, Marata *et al* [10], studied the nutritional value of tempeh and this value and the values for unfermented soybean were compared. There were no large differences in protein and ash content between tempeh and unfermented soybeans, but free amino acids were increased during fermentation. The amount of different free amino acids in the palatable tempeh was from 1 to 8.5 times as much as that of unfermented soybeans. Fibre slightly increased during fermentation. The fat content of tempeh was slightly lower than that of soybeans, but the acid value was noticeably higher. Riboflavin, vitamin B6, nicotinic acid and pantothenic acid were increased during fermentation, although there was little alteration in thiamine.

The shelf life of fried food is mostly determined by the moisture content after frying. Food that is more thoroughly dried by frying, for example potato chips, maize and other potato snack food, have a shelf life of up to 12 months at ambient temperature. The quality is maintained by adequate barrier properties of packaging materials and correct storage conditions. The fried red kidney bean tempeh was also thoroughly dried and had low water activity and moisture content so it is expected to be a long shelf-life product from the next investigation.

Determination of optimal package for storage

Fried red kidney bean tempeh stored in three different packaging materials changed in physical and chemical properties during storage. Figures 5 and 6 show all of the samples slightly increased in L* value. However, the effect of packaging material on L* and a* values of all samples were not statistically significant, but b* values were significantly different (Fig. 7). Crispness of fried tempeh was determined as compressive force by texture analyzer. The compressive force of fried tempeh samples in nylon A, nylon B and metal foil bags after storage ranged from 203.89 to 602.26, 198.91 to 597.55 and 200.21 to 603.49 gramforce (gf) respectively. Figure 8 shows compressive force of all samples dramatically increased after storage. The three different packaging materials significantly affected compressive force of fried tempeh as well as acid values. Figure 9 shows acid value (AV) of all samples dramatically increased after storage. In this research, peroxide values (PV) were not detected in all samples, including newly fried samples.

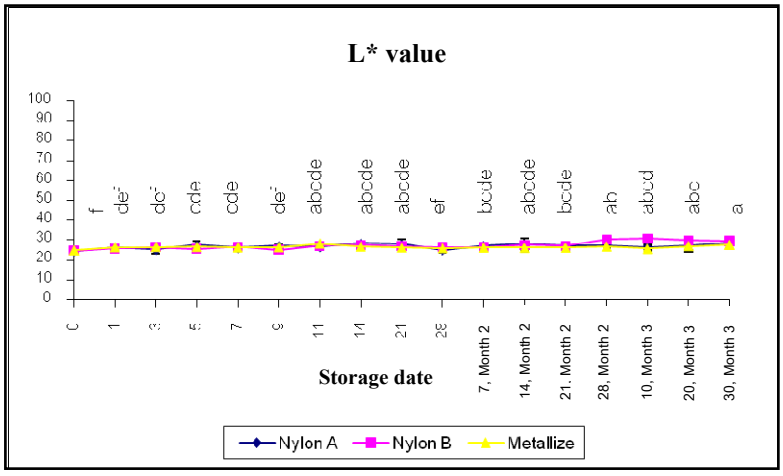

Figure 5. Changes in L* value of fried red kidney bean tempeh during storage in three different packaging materials at room temperature for 3 months.

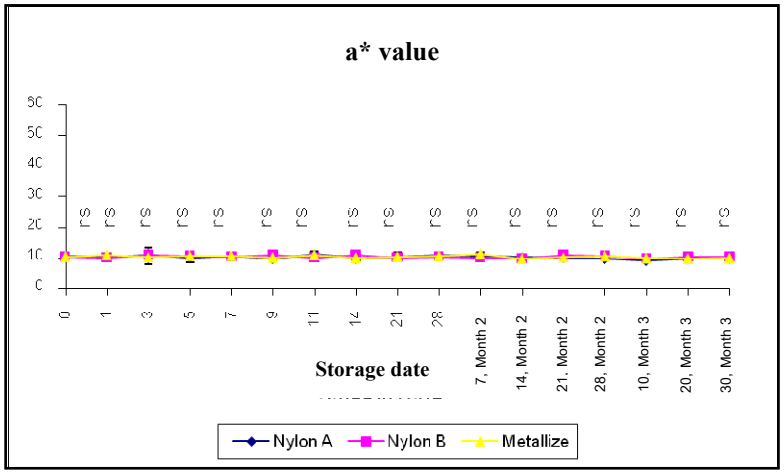

Figure 6. Changes in a* value of fried red kidney bean tempeh during storage in three different packaging materials at room temperature for 3 months.

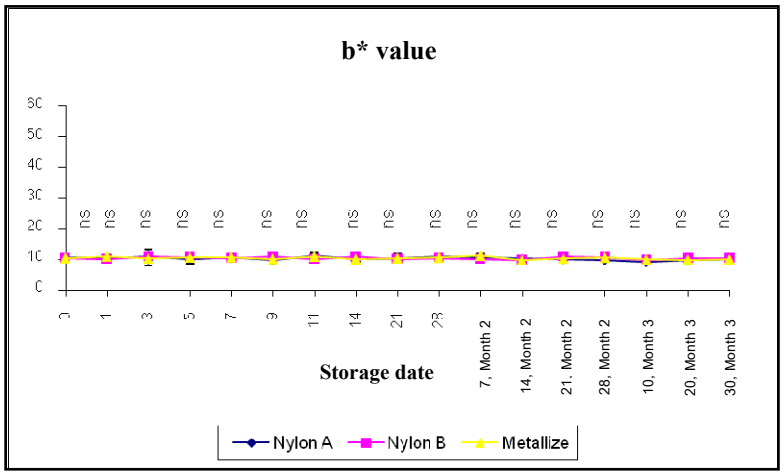

Figure 7. Changes in b* value of fried red kidney bean tempeh during storage in three different packaging materials at room temperature for 3 months.

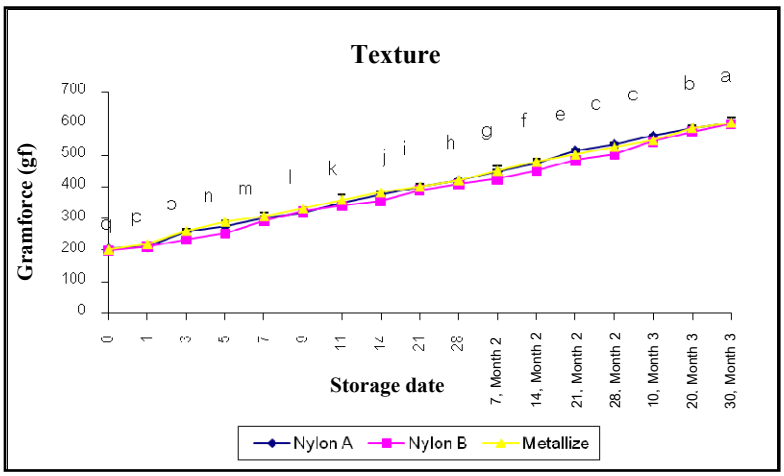

Figure 8. Changes in crispness (compressive force) of fried red kidney bean tempeh during storage in three different packaging materials at room temperature for 3 months.

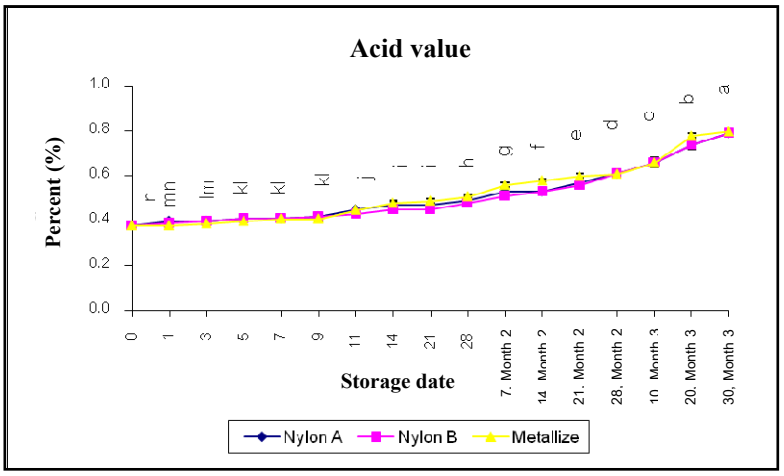

Figure 9. Changes in acid value (% free fatty acid) of fried red kidney bean tempeh during storage in three different packaging materials at room temperature for 3 months.

Sensory evaluation

The results of sensory evaluation of fried red kidney bean tempeh during storage in three different packaging materials at room temperature for 3 months are shown in Tables 3, 4 and 5. The variation in time affected sensory evaluation. According to Tables 3, 4 and 5, when time storage time increased from 0 to 3 months, the colour, flavour, taste, crispness and overall acceptability scores were reduced. This was due to rancidity and the loss of crispness.

Table 3. Effect of time on sensory evaluation (9-point hedonic scale test) of fried red kidney bean tempeh in Nylon A.

Treatment	Attributes				
	colour	flavour	taste	crispness	overall
1	6.13^{ab}±1.92	6.43^{a}±1.77	6.73^{a}±1.85	7.26^{a}±1.65	7.00^{a}±1.55
2	6.56^{a}±2.04	6.53^{a}±1.65	6.73^{a}±1.91	7.30^{a}±1.64	7.26^{a}±1.48
3	5.70^{b}±1.70	5.40^{b}±1.88	5.93^{a}b±1.91	5.93^{b}±1.96	6.00^{b}±1.78
4	5.46^{b}±2.08	5.43^{b}±2.04	5.43^{b}±2.02	6.06^{b}±1.77	5.96^{b}±1.71

* Means within the same column with different letter are significantly different ($p \leq 0.05$)
^{ns} are not significantly different at $p > 0.05$

Table 4. Effect of time on sensory evaluation (9-point hedonic scale test) of fried red kidney bean tempeh in Nylon B.

Treatment	Attributes				
	colour	flavour	taste	crispness	overall
1	6.50^{ab}±1.75	6.56^{a}±1.61	5.90^{b}±2.04	6.86^{ab}±1.45	6.66^{a}±1.77
2	6.96^{a}±1.60	6.86^{a}±1.69	7.16^{a}±1.58	7.30^{a}±1.17	7.13^{a}±1.31
3	4.86^{c}±1.85	4.63^{b}±2.28	5.20^{b}±2.15	5.10^{c}±1.80	5.20^{b}±1.82
4	5.76^{b}±1.77	5.33^{b}±2.17	5.46^{b}±1.84	6.16^{b}±2.03	5.45^{b}±1.86

* Means within the same column with different letter are significantly different ($p \leq 0.05$)
^{ns} are not significantly different at $p > 0.05$

Table 5. Effect of time on sensory evaluation (9-point hedonic scale test) of fried red kidney bean tempeh in metal foil package.

Treatment	Attributes				
	colour	flavour	taste	crispness	overall
1	5.83^{bc}±1.51	5.90^{b}±1.47	6.40^{a}±1.03	6.80^{a}±1.32	6.63^{ab}±1.32
2	6.96^{a}±1.32	6.56^{a}±1.47	6.66^{a}±1.72	7.20^{a}±1.24	7.13^{a}±1.22
3	5.26^{c}±1.68	5.16^{c}±1.51	5.26^{b}±1.77	5.63^{b}±1.49	5.63^{c}±1.40
4	6.30^{ab}±1.78	5.16^{c}±1.51	5.50^{b}±1.59	6.10^{b}±1.58	6.16^{bc}±1.44

* Means within the same column with different letter are significantly different ($p \leq 0.05$)
^{ns} are not significantly different at $p > 0.05$

Treatment 1: Newly fried red kidney bean tempeh
Treatment 2: Fried red kidney bean tempeh after storage for a month
Treatment 3: Fried red kidney bean tempeh after storage for 2 months
Treatment 4: Fried red kidney bean tempeh after storage for 3 months

The results of sensory evaluation of fried tempeh in three different packaging materials for equal storage time are shown in Tables 6, 7 and 8. The hedonic score of all attributes ranged from neither like nor dislike, to like moderately. The scores of each attribute were not significantly different ($P > 0.05$).

Table 6. Sensory evaluation (9-point hedonic scale test) of fried red kidney bean tempeh in three different packaging materials for a month.

Treatment	Attributes				
	colour	flavour	taste	crispness	overall
1	$5.53^{ns}\pm2.01$	$6.06^{ns}\pm1.72$	$5.96^{ns}\pm2.09$	$6.66^{ns}\pm1.47$	$6.26^{ns}\pm1.70$
2	$5.90^{ns}\pm1.58$	$5.76^{ns}\pm1.65$	$5.80^{ns}\pm1.80$	$6.93^{ns}\pm1.11$	$6.30^{ns}\pm1.20$
3	$5.50^{ns}\pm1.81$	$5.36^{ns}\pm1.65$	$5.66^{ns}\pm1.66$	$6.50^{ns}\pm1.40$	$5.96^{ns}\pm1.56$

* Means within the same column with different letter are significantly different ($p\leq0.05$)
^{ns} are not significantly different at $p>0.05$

Table 7. Sensory evaluation (9-point hedonic scale test) of fried red kidney bean tempeh in three different packaging materials for 2 months.

Treatment	Attributes				
	colour	flavour	taste	crispness	overall
1	$5.20^{ns}\pm1.93$	$5.63^{a}\pm1.56$	$5.53^{a}\pm1.94$	$6.03^{a}\pm1.51$	$5.66^{a}\pm1.91$
2	$5.06^{ns}\pm1.76$	$4.56^{b}\pm1.88$	$4.36^{b}\pm2.10$	$5.10^{b}\pm2.04$	$4.96^{b}\pm2.05$
3	$4.86^{ns}\pm1.99$	$5.26^{ab}\pm1.59$	$5.10^{ab}\pm2.04$	$5.93^{a}\pm1.91$	$5.36^{ab}\pm1.79$

* Means within the same column with different letter are significantly different ($p\leq0.05$)
^{ns} are not significantly different at $p>0.05$

Table 8. Sensory evaluation (9-point hedonic scale test) of fried red kidney bean tempeh in three different packaging materials for 3 months.

Treatment	Attributes				
	colour	flavour	taste	crispness	overall
1	$6.53^{a}\pm1.38$	$5.56^{ns}\pm1.73$	$5.66^{ns}\pm1.88$	$6.00^{ns}\pm1.91$	$6.10^{ns}\pm1.62$
2	$5.36^{b}\pm1.62$	$5.56^{ns}\pm1.50$	$5.50^{ns}\pm2.04$	$6.23^{ns}\pm1.69$	$5.86^{ns}\pm1.47$
3	$6.86^{a}\pm1.25$	$5.80^{ns}\pm1.24$	$5.86^{ns}\pm1.81$	$6.36^{ns}\pm1.65$	$6.36^{ns}\pm1.42$

* Means within the same column with different letter are significantly different ($p\leq0.05$)
^{ns} are not significantly different at $p>0.05$

Treatment 1: Fried red kidney bean tempeh in Nylon A
Treatment 2: Fried red kidney bean tempeh in Nylon B
Treatment 3: Fried red kidney bean tempeh in Metalize

Conclusions

From the results of this research, it is feasible to use red kidney bean as a good material for tempeh. Red kidney bean tempeh looks like the original tempeh but is darker in colour. Fried red kidney bean tempeh in three different types of packaging changed in physical and chemical properties during storage at room temperature due to oxygen head space in the packaging. However, the property changes in fried tempeh kept in the nylon B container occurred more slowly, especially acid value and crispness. It is concluded that nylon B is the more suitable material for packaging fried red kidney bean tempeh.

Acknowledgement

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