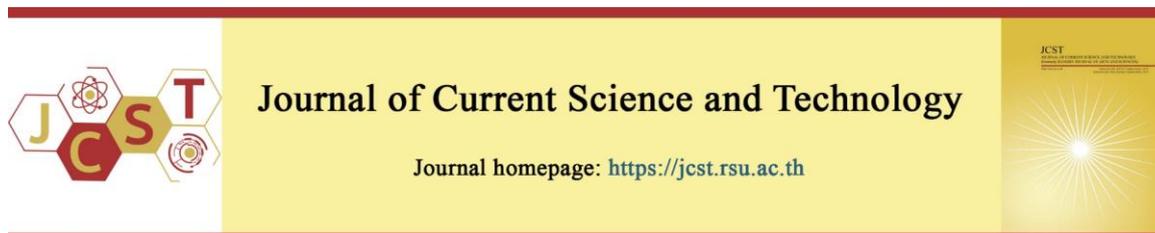


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Innovation for plant-based foods: Allergen-free vegan meat and egg products from rice processing by-products

Yupakanit Puangweraku^{1*} and Suvimol Soithongsuk²

^{1,2}Faculty of Food Technology, College of Agricultural and Food, Rangsit University, Patumthani 12000, Thailand

¹E-mail: lombiotec@yahoo.com; ²E-mail: suvimol.88@gmail.com

*Corresponding author

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Abstract

This study aimed to develop plant-based vegan meat and egg products that still contain high nutritional and functional components from the by-products of rice processing. The ingredients such as rice malt, red rice malt, and bio-cellulose from rice kombucha processing were prepared and mixed to produce vegan meat and egg products. The optimal ratio of a rice protein hydrolysate, as an alternative emulsifier to commercial food hydrocolloid, was also investigated. The results showed that the vegan meat contained comparable content of protein and minerals, with 16 times lower fat and 3 times higher dietary fiber than the commercial vegan meat. Similarly, the vegan egg product contained the same content of protein and minerals, but with 14 times lower fat and 4 times higher fiber than the product from a real egg. The vegan meat and egg products contained higher polyphenol, antioxidant, ACE inhibitory peptide, D-saccharic acid 1,4-lactone (DSL), and gamma-aminobutyric acid (GABA) than the real meat, real egg, and commercial vegan products. The commercial hydrocolloid can be substituted by the natural rice protein hydrolysate at 70% and 100% for vegan meat and egg products, respectively. Both products exhibited comparable properties to the commercial products in terms of color, firmness, oil holding capacity, size expansion, as well as sensory attributes (color and taste). The shelf-life study indicated that the vegan meat and vegan egg powder could be stored at -18°C and at ambient temperature for 18 and 16 months, respectively. Product cost and pricing calculations showed that the products were highly competitive. This innovative research presented a high potential for further commercialization for allergen-free plant-based proteins.

Keywords: allergy free; bioactive compound; nutritional value; plant-based food; rice protein hydrolysate; vegan egg; vegan meat.

1. Introduction

Today, consumer awareness is changing, resulting in a shift in dietary choices. Over time, a surging world population and industrial development have created a massive demand for animal protein and food production expansion. However, animal-derived products, particularly meat, have a significant impact on the environment via greenhouse gas production, land-water usage, and extensive energy consumption (Steinfeld, et al, 2006). The United Nations Framework Convention

on Climate Change (UNFCCC) revealed that the greenhouse gases increased to 413.3 ppm in the year 2020. This increase affected the temperature increasing over 1.5 °C without a tendency to decrease. Greenhouse gas emissions from feed production represent 60-80% of the emission coming from chicken and pig, and 35-45% of the milk and beef sector (Grossi, Goglio, Vitali, & Williams, 2019). Xu et al. (2021) reported that global greenhouse gas emissions from animal-based foods are twice those of plant-based foods. In the

case of Denmark, the transition from animal-based food to plant-based food production can reduce greenhouse gas emissions in the range of 58.2-86.5%. The emission reductions will be possible with the full global implementation of the Planetary Health Diet in a scenario where domestic cattle, pig, and chicken production is downscaled to 76.2%, 21.1%, and 88.2% of current production (Prag & Henriksen, 2020). Plant-based diets focus on foods primarily from plants. Many studies have shown that these diets have low fat and no cholesterol. They are not only full of nutrients but also contain dietary fiber that can reduce the risk of NCDs (non-communicable diseases) such as obesity, diabetes, heart disease, vascular disease, and cancer (Li, 2014; Lin, Chiu, & Chang, 2019). Due to the nutritional benefits of plant-based foods, their market is growing continuously. Forbes, an American business magazine reported that the world's plant-based meat market in 2025 will cost 27.9 billion US dollars or grow 15% per year while the world's plant-based egg products in 2027 will cost 1.72 billion US dollars or grow 6.2% per year and up to 140 billion US dollars in 2029 (Franck, 2019). Plant-based food will be the group that drives the market to grow significantly, and this is an opportunity for new manufacturers (Nuntramas, Suprasert, & Akegachan, 2020). Many reports mentioned that consumers are shifting to eat a plant-based substitute for meat. 62% of Chinese consumers are likely to buy plant-based meat regularly (Allen, 2019) and 63% of Indian consumers were willing to turn to plant-based meat to replace eating meat (Bhatia, 2019). More than 23% of the US consumers turned to consume more plant-based food during the COVID-19 pandemic because it is a necessary product that has longer life storage and is often not in short supply in the market compared with animal-based products (Danziger, 2020). 1 of 4 European consumers is willing to reduce eating meat in 5 years, especially the elderly group who give the importance of health as the top priority (Geijer, 2017). In Thailand, 53% of Thai consumers are interested to reduce meat consumption while another 45% are willing to change to eating vegetarian food, vegan and plant-based food (Mintel Press Team, 2018).

The most important nutrient in plant-based food is protein. Almost all plant proteins can serve as raw material for meat analogs, but there are

several factors that determine their suitability for this purpose such as the availability and cost of the raw material and technological availability. The most widely used proteins for meat analogs include soybean, pea, and wheat proteins (Choudhury, Singh, Seah, Yeo, & Tan, 2020). However, soy protein has sensory limitations associated with beany, grassy, and bitter flavors when used in the products (Chamba, Hua, & Simwaka, 2014). Moreover, soybean is the most frequently genetically modified plant, which might not be accepted by consumers who are concerned over food naturalness, while wheat protein contains gluten that can cause celiac disease or wheat allergy for some consumers. Celiac disease is a form of gluten intolerance that involves the immune system attacking the gluten and the lining of the gut, leading to gut wall damage and may cause nutrient deficiencies (Kreutz, Adriaanse, van der Ploeg, & Vreugdenhil, 2020). Many attempts have been made to develop allergen-free food ingredients such as rice malt, red rice malt (angkak), bio-cellulose from rice kombucha, and rice protein hydrolysate. Previous research analyzed the important components of these ingredients, namely (1) rice malt that contains vitamin B1, B2, B3, B5, B6, GABA, fiber, antioxidants, and enzymes that help to digest starch and protein; (2) red rice malt that contains monacolin K and mevinolin that have many benefits such as anti-inflammatory, anti-bacteria and antioxidant activity; (3) rice protein hydrolysate that contains branched-chain amino acid (BCAA), which functions as brain and muscle booster, and ACE inhibitory peptide, which helps to reduce blood pressure; (4) rice protein isolate gave 2-methyl-3-furyl-disulfide, 2-methyl-3-furanthiol and 2, 6-dimethyl-pyrazine that are egg flavor compound; (5) dried yeast that contains vitamin B12, iron, zinc, and beta-glucan; and (6) bio-cellulose that contains fiber and DSL, which help to inhibit cancer (Puangwerakul & Soithongsuk, 2020; Puangwerakul & Soithongsuk, 2021).

Plant-based foods are not only one of the main trends for "Foods Today" and "future Foods", but they also present health benefits for consumers. Moreover, rice processing by-products contain various types of functional components suitable as raw materials for different value-added food applications. Therefore, the objective of this innovative research is to develop allergen-free

vegan meat and egg products using raw materials from by-products and waste of rice processing as ingredients.

2. Objectives

The goal of this innovative research was to develop allergen-free vegan meat and egg products by evaluating the substitution effect of rice protein hydrolysate on physicochemical properties and shelf life of developed vegan meat and egg. The effects of cooking on physical changes and sensory evaluation of vegan meat and vegan egg products were also compared with the commercial vegan meat and vegan egg.

3. Materials and methods

3.1. Materials

All ingredients used in vegan meat and egg are % weight/ weight as follows:

3.1.1 Vegan meat

100g/ one serving size contains 25% fresh bio-cellulose from kombucha, 15% rice protein hydrolysate, 15% rice malt, 9% angkak red rice malt, 4% red bean and 3% cockle mushroom, 2% commercial hydrocolloid (Benecel™ A4M modified cellulose), 1% rice bran oil and 1.5% dried yeast.

3.1.2 Vegan egg powder

24g/one serving size contains 33% rice protein hydrolysate, 34% rice malt powder, 17% rice malt bran, 5% dried bio-cellulose powder, 4% rice protein isolate, 3% dried yeast, 2% angkak red rice malt powder and 2% commercial hydrocolloid (Benecel™ A4M modified cellulose).

All ingredients used in 3.1.1 and 3.1.2 were kindly supplied by the Community Enterprise of Nong Sarai Farmers Group, and Innovative Research and Incubation of Entrepreneur Center, Rangsit University.

3.1.3 Commercial vegan meat were purchased from USA

260g/one serving size contains 41% pea protein, 12% canola oil, 5% coconut oil, 5% potato starch, 0.37% yeast extract and yeast powder, 0.25% maltodextrin, 0.2% salt, 0.15% sunflower oil, 0.02% color from beet juice extract, methyl cellulose stabilizing agent and flavor (INS460, INS461, INS422, INS414, INS300, INS363, INS260).

3.1.4 Commercial vegan eggs were purchased from Australia

50g/one serving size contains 41.7% pregelatinized chickpea flour, 32% corn flour, 8% plant protein, 3% dextrose, 1% salt, 1% pigment from turmeric, stabilizing agent, and foaming agent (INS407, INS461, INS170).

3.2. Method

3.2.1 Preparation of vegan meat and egg and chemical analysis

The main ingredients for the preparation of vegan meat and egg products were mixed following the recipe from previous research (Puangwerakul & Soithongsuk, 2020; Puangwerakul & Soithongsuk, 2021). The nutritional compositions of the products were analyzed by proximate analysis (AOAC, & Horwitz, 2000). Besides, biologically active compounds and activities were analyzed as follows: total phenolic compounds (Singleton & Rossi, 1965); antioxidant activity (Brand-Williams, Cuvelier, & Berset, 1995); ACE inhibitory activity (Li, Liu, Shi, & Le, 2005); DSL (Wang, Gan, Tang, Wang, & Tan, 2009); and GABA (Kitaoka and Nakano, 1969). To compare the quality of the developed products, fresh meat and egg (size number 0) and the commercial plant-based meat and egg used as the benchmark brands were also analyzed in the same procedures.

3.2.2 Use of protein hydrolysate from rice as an emulsifier and a foaming agent

Protein hydrolysate can function as both an emulsifier and a foaming agent. Therefore, to reduce the amount of modified cellulose; commercial hydrocolloid in the recipe of vegan meat and egg, the hydrocolloid was substituted with rice protein hydrolysate. The ratios of hydrocolloid: rice protein hydrolysate were varied at 100: 0 (control), 70: 30, 50: 50, 30: 70, and 0: 100, respectively. The products from 5 treatments were sensory evaluated by 15 trained panelists using a 5-point hedonic scale to select the best recipe.

3.2.3 Quality of the products before and after frying

The best formula for vegan meat and egg that were chosen in 3.2.2 were evaluated for color and texture before and after frying. To prepare fried vegan egg, 24 g of vegan egg powder was mixed with 70 g of water until homogeneous. The mixture was fried under medium heat until both sides were cooked. To prepare fried vegan meat, the frozen vegan meat was

thawed for 30 minutes and fried the product until fully cooked. The color (L^* , a^* , b^* value) of the products were measured with a Chroma meter (Minolta model CR-10, JP). The Texture Profile Analysis (TPA) of vegan meat before and after frying were measured with a Texture Analyzer (model LRX, Lloyd Instrument, Hampshire, UK) by measuring cutting force as newton (N). The viscosity of vegan egg before frying was measured by Brook Field Viscometer in Centipoise (cP) and its TPA after frying was measured by the same procedure for vegan meat. After frying, the products were also measured for oil holding capacity and expansion by the modified method of Ang (1991).

3.2.4 Sensory evaluation of the products

The best formula for vegan meat and egg that were chosen in 3.2.2 were evaluated for consumer acceptance. Two groups of consumers: general and vegan consumers (100 persons/group) were requested to conduct a 5-point hedonic scale for the appearance, color, taste, texture, and overall acceptability of the products after frying.

3.2.5 Shelf-life study

The shelf-life of vegan meat was evaluated at -18°C for 18 months and the shelf-life of vegan egg powder was evaluated at accelerating temperatures for 12 months. The accelerated shelf-life study at 45°C and 55°C was performed by the method of Steele (2004). The temperature-accelerating factor or temperature quotient, Q_{10} , and Q_1 were calculated as follows.

$$Q_{10} = \frac{\text{shelf life at } 45^{\circ}\text{C (days)}}{\text{shelf life at } 55^{\circ}\text{C (days)}}$$
$$Q_1 = Q_{10}^{0.1}$$

During the shelf-life study, the products were taken every 3 months for microbial and chemical (rancidity) analysis, total microbial count by the method of Maturin and Peeler (2001), and 2-

thiobarbituric acid (TBA) as the method of Wrolstad et al. (2005).

4. Results and Discussion

4.1 Nutritional and chemical compositions

The chemical compositions of vegan meat were compared with real beef and commercial vegan meat as illustrated in Figure 1. The vegan meat had lower lipid content, but higher mineral and fiber content than the real beef. In comparison to commercial vegan meat, no difference in protein and mineral content was observed. However, fat content in the vegan meat was sixteen times lower, fiber content was three times higher, and carbohydrate content was five times higher than the real beef and commercial vegan meat due to the raw ingredient source.

The chemical compositions of the vegan egg compared with that of the real egg exhibited no difference in protein and mineral content. However, fat content in the vegan egg was fourteen times lower, fiber content was four times higher, and carbohydrate content was eighteen times higher than the real egg due to the raw ingredient source. The protein content of the vegan egg was found to be three times higher, while the fat content was found to be two times lower than the commercial vegan egg. No significant difference in mineral and fiber content was found, and the carbohydrate content of the vegan egg was one point four times higher than the commercial vegan egg.

The vegan meat and vegan egg, on the other hand, contained fewer calories than the real meat and real egg, suggesting that their consumption could help people to control their weight.

For bio-functional compounds analysis, vegan meat and vegan egg contained the highest 5 functional bioactive compounds: phenolic compounds, antioxidant, ACE inhibitory agent, DSL, and GABA when compared with meat, real egg, and commercial vegan products as shown in Figure 2.

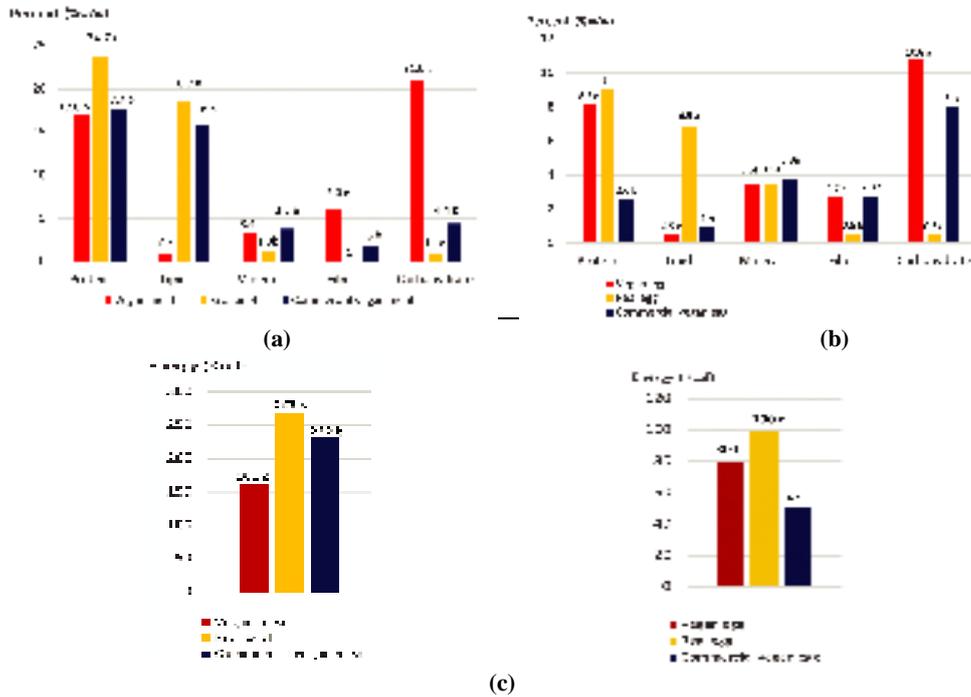


Figure 1 Proximate composition of vegan meat (a), vegan egg (b), and their energy values (c) compared with real beef, real egg, and commercial vegan products. Data with different letters ^{abc} represent a significant difference ($p \leq 0.05$).

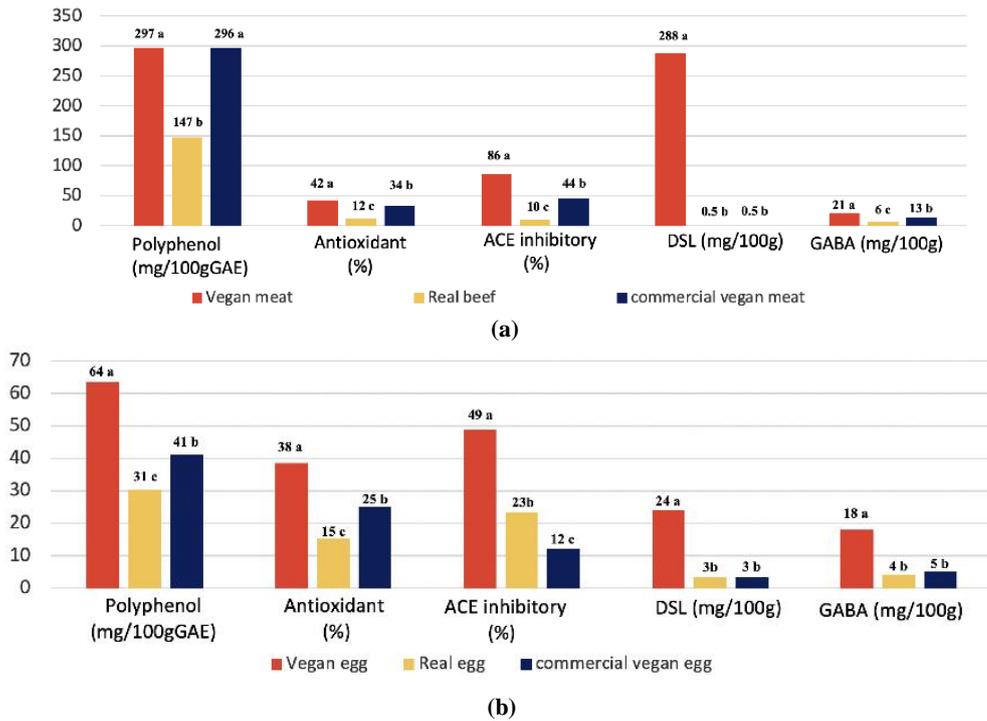


Figure 2 Bio-functional compounds of vegan meat (a) and vegan egg (b) compared with real beef, real egg, and commercial vegan products. Data with different letters ^{abc} represent a significant difference ($p \leq 0.05$).

4.2 Use of rice protein hydrolysate as a substitution for commercial hydrocolloids

Vegan meat products prepared from a mixture of commercial hydrocolloids and protein hydrolysate at different ratios of 100:0, 70:30, 50:50, 30:70, and 0:100 were compared with real beef and commercial vegan meat. Sensory evaluation with trained 15 panelists evaluated the texture, color, taste, and overall liking of the vegan meat to determine the highest sensory quality of the vegan meat formula. The result showed that the real meat received the highest overall liking score. However, the product with hydrocolloid and protein hydrolysate at a ratio of 30:70 was comparable to

the commercial vegan egg in all attributes, thus this formula was chosen for the production of vegan meat. Aside from the quality acceptability, another advantage of using this mixture of hydrocolloid and protein hydrolysate in the manufacturing of vegan meat was the low cost of the raw materials.

For the vegan egg, the use of hydrocolloid and protein hydrolysate in all ratios compared with the commercial vegan egg showed that the substitution of the commercial hydrocolloids by rice protein hydrolysate could be at 100% because of the functional property of rice protein hydrolysate as an emulsifier and a foaming agent on vegan egg product.

Table 1 Liking scores of vegan meat and egg prepared with different ratios of hydrocolloid to rice protein hydrolysate compared with real beef, real egg, and commercial product

Attribute	Scores					Real beef	Commercial vegan meat
	Ratios of hydrocolloid to rice protein hydrolysate in vegan meat						
	100:0	70:30	50:50	30:70	0:100		
Meat							
Texture	4.0±0.0 ^b	4.0±0.2 ^b	4.0±0.1 ^b	3.8±0.2 ^b	3.2±0.3 ^c	5.0±0.0 ^a	4.1±0.2 ^b
Color	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}
Taste	4.9±0.1 ^{ab}	4.8±0.0 ^b	4.9±0.1 ^{ab}	4.7±0.1 ^b	4.7±0.1 ^b	5.0±0.0 ^a	4.8±0.1 ^b
Overall liking	4.4±0.2 ^b	4.3±0.2 ^b	4.4±0.2 ^b	4.2±0.2 ^b	3.8±0.2 ^c	5.0±0.0 ^a	4.5±0.3 ^b
Egg							
Texture	3.8±0.1 ^b	3.7±0.1 ^b	3.7±0.1 ^b	3.7±0.1 ^b	3.7±0.1 ^b	5.0±0.0 ^a	3.8±0.1 ^b
Color	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}	5.0±0.0 ^{ns}
Taste	4.8±0.2 ^{ab}	4.7±0.1 ^b	4.8±0.1 ^b	4.6±0.3 ^b	4.8±0.1 ^b	5.0±0.0 ^a	4.7±0.2 ^b
Overall liking	4.4±0.2 ^b	4.3±0.4 ^b	4.3±0.4 ^b	4.3±0.3 ^b	4.3±0.4 ^b	5.0±0.0 ^a	4.3±0.3 ^b

Data are means ± SD

^{abc} Means in the same row with different superscripts are significantly different ($p \leq 0.05$)

ns: not significant

4.3 Effects of cooking on physical changes of vegan meat and egg

High temperature during the cooking affected the structure of red pigment in real beef and pork, resulting in a color change in the vegan meat as exhibited in Table 2. Hem protein had been completely destroyed while the red pigment in vegan meat was a group of substances polyketide with azaphilone group, which was not affected by heat (Dijksterhuis, & Samson, 2007). The red color of the vegan meat was rubropanthamine and monascorubramine, which are heat stable. However, the color value of the vegan meat before frying was closer to the color of raw pork than raw beef. After frying, the vegan meat's color was similar to cooked pork more than cooked beef.

The vegan egg before frying showed the highest brightness (L^*) and the lowest a^* (red) and b^* (yellow) value in contrast to the real egg and commercial vegan egg, which showed the highest a^* value and b^* value, respectively. The effect of heat on the color changing of vegan eggs resulted from caramelization and Maillard reaction in germinated rice malt flour, the main ingredient. Since rice malt powder contains sugar, reducing sugar and protein, including amino acids, resulted in a formation of melanoidins, the reddish-brown substance that decreased the brightness (L^*) (Kunze, 2004). For the real egg after frying, the color change resulting from heating changed the structure of lutein (orange-red color) and zeaxanthin (yellow color), which are carotenoid pigments. Their structures were changed from trans

(intense color) to cis (Packer, Hiramatsu, & Yoshikawa, 1999). The decrease in the red and yellow color of the commercial vegan egg after frying is related to the deterioration of curcumin (orange-red color) since curcumin is not stable at high temperatures of more than 50 °C (Chassagnez-Mendez, Correa, Franca, Machado, & Arujo, 2000). However, the L* and b* values of the vegan egg after frying were at the same level as the real egg.

For the cutting force value measured by texture analyzer compared with the real beef before frying, it was found that the raw beef and raw pork had a higher firmness than the vegan meat and commercial vegan meat, respectively. However, after frying, the firmness values of beef, pork, vegan meat, and commercial vegan meat were not different. After heating, the real beef shrank, squeezing meat fibers leading to a release of juices (containing water) and shrinkage.

The viscosity of the vegan egg before frying was the same as the real egg but lower than the commercial vegan egg. The commercial vegan egg had the highest viscosity since it used pregelatinized chickpea flour as raw material. The pregelatinized starch of chickpea flour could be dissolved in water at room temperature and gives instant viscosity (Swinkels, 1985). Rice malt flour was used in vegan eggs. It must be heated until completely cooked before its viscosity can be demonstrated. This is why its viscosity was lower than the commercial vegan egg. However, after

frying, both vegan egg products had the same firmness as the real egg.

The vegan meat that was fried showed physical changes, unlike the real beef. The vegan meat showed an oil holding capacity 4 times higher than the real beef, resulting in their size expansion that is not different from the commercial vegan meat. The real beef, however, showed the opposite effect by shrinking down in size (Figure 3a).

From Figure 3b, the vegan egg had two times higher oil holding capacity but two times lower size expansion than the real egg. Size expansion is related to different chemical compositions in an egg. The real egg contains albumin protein that functions as the main structure for foaming stability, resulting in a layer between the interface of water and air (Liang & Kristinsson, 2005). In contrast, the vegan egg does not contain albumin but a higher amount of starch than the real eggs. Therefore, its expansion capacity was lower than the real egg. Lipid in starch granule (rice malt starch in vegan egg; chickpea starch and corn starch in commercial vegan egg) combine with amylose to form an inert amylose-lipid complex. The complex is a soft crystal structure that enhances starch granule strength causing low expansion capacity. Galliard, and Bowler (1987) and Bogracheva, Morris, Ring, and Hedley. (1998) indicated that the higher amount of lipid in starch granule, the lower the expansion capacity of the product. Therefore, the vegan egg that contained lower fat content showed higher size expansion than the commercial vegan egg.

Table 2 Physical properties of vegan meat and egg before and after frying compared with real meat, real egg, and commercial product

Sample		Before frying				After frying				
Meat	Appearance	L*	a*	b*	Cutting force (N)	Appearance	L*	a*	b*	Cutting force (N)
Vegan meat		60.90± 1.10 ^{aA}	6.37± 0.45 ^{nsB}	21.68± 0.71 ^{nsA}	5.02 ± 0.20 ^{bB}		57.30± 1.65 ^{bA}	5.90± 0.98 ^{nsB}	23.36 ± 2.57 ^{nsAB}	6.95 ± 0.30 ^{aNS}
Real beef		42.38± 1.82 ^{bC}	9.38± 0.30 ^{aA}	11.26± 1.32 ^{bB}	6.64 ± 0.30 ^{bA}		47.40± 2.11 ^{bB}	9.01 ± 1.62 ^{bA}	17.46 ± 1.15 ^{aB}	7.13 ± 0.20 ^{aNS}
Real pork		58.56± 1.65 ^{aA}	6.92± 0.56 ^{aB}	21.14± 2.78 ^{bA}	6.46 ± 0.20 ^{bA}		57.24± 2.82 ^{aA}	5.78 ± 0.01 ^{bB}	27.52 ± 2.48 ^{aA}	7.03 ± 0.30 ^{aNS}

Sample	Before frying					After frying					
	Meat	Appearance	L*	a*	b*	Cutting force (N)	Appearance	L*	a*	b*	Cutting force (N)
Commercial vegan meat		51.12±1.22 ^{ab}	9.02±0.33 ^{nsA}	12.88±1.28 ^{ab}	4.21±0.30 ^{bc}		47.35±1.38 ^{ab}	9.04±0.65 ^{nsA}	16.86±1.32 ^{ab}	6.98±0.40 ^{nsB}	
Egg	Appearance	L*	a*	b*	Viscosity (cP)	Appearance	L*	a*	b*	Cutting force (N)	
	Vegan egg		72.39±0.07 ^{AA}	4.34±0.84 ^{ac}	38.72±2.04 ^{ac}	6487±166 ^B		70.39±0.30 ^{BA}	4.00±0.18 ^{ab}	32.57±2.89 ^{BB}	2.76±0.22 ^{AB}
	Real egg		63.75±1.25 ^{BB}	7.43±0.92 ^{AA}	45.23±1.31 ^{ab}	6550±180 ^B		69.85±1.19 ^{AA}	1.20±0.92 ^{BA}	36.21±5.43 ^{BB}	3.02±0.20 ^A
Commercial vegan egg		58.06±0.06 ^{BC}	2.73±0.49 ^{AB}	47.06±0.17 ^{AA}	36960±357 ^A		60.64±0.16 ^{AB}	1.46±0.40 ^{BA}	40.81±2.70 ^{BA}	2.50±0.15 ^B	

^{abc} Means ± SD in the same row with different superscripts are significantly different (p≤ 0.05)
^{ABC} Means ± SD in the same column with different superscripts are significantly different (p≤ 0.05)
 ns: not significant

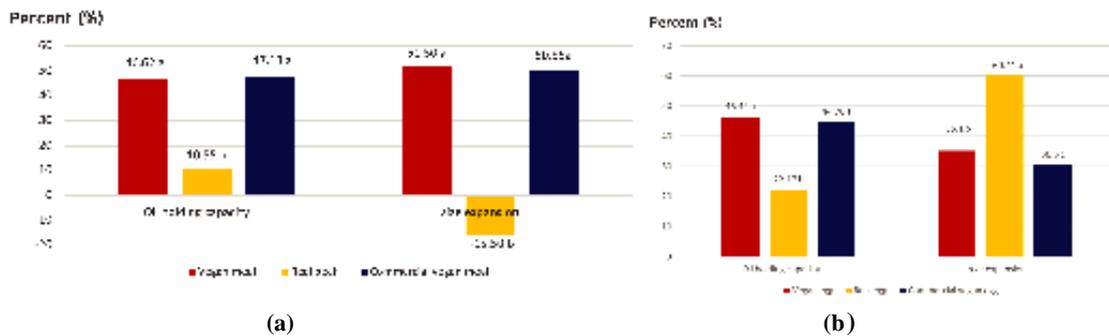


Figure 3 Oil holding capacity and size expansion of (a) vegan meat and (b) vegan egg compared with real beef, real egg, and commercial vegan products.

Table 3 Sensory evaluation of vegan meat and egg by vegan and general consumers

Attribute	Meat				Egg			
	Vegan consumers		General consumers		Vegan consumers		General consumers	
	Vegan meat	Commercial vegan meat	Vegan meat	Commercial vegan meat	Vegan egg	Commercial vegan egg	Vegan egg	Commercial vegan egg
Appearance	3.96±0.12 ^a	4.18±0.18 ^a	3.74±0.13 ^b	3.87±0.20 ^b	4.04±0.68 ^a	3.68±0.75 ^a	4.12±0.83 ^a	3.92±0.70 ^a
Color	4.11±0.13 ^a	4.16±0.11 ^a	3.84±0.19 ^b	3.85±0.23 ^b	4.12±0.60 ^a	3.60±0.71 ^b	3.96±0.89 ^{ab}	4.00±0.65 ^{ab}
Aroma	4.16±0.11 ^a	4.12±0.11 ^a	3.86±0.11 ^b	3.90±0.22 ^{ab}	3.80±0.87 ^{ab}	3.96±0.54 ^a	3.88±1.09 ^{ab}	3.40±1.00 ^b
Taste	4.08±0.10 ^a	4.03±0.18 ^a	3.82±0.44 ^b	3.96±0.25 ^{ab}	3.80±0.76 ^a	3.96±0.54 ^a	3.44±1.12 ^a	2.72±1.02 ^b
Texture	4.11±0.16 ^a	4.17±0.12 ^a	3.04±0.46 ^b	3.17±0.28 ^b	3.52±0.87 ^{ab}	3.92±0.57 ^a	3.72±1.10 ^{ab}	3.24±1.27 ^b
Overall liking	4.14±0.15 ^a	4.16±0.10 ^a	3.74±0.45 ^b	3.66±0.25 ^b	3.88±0.65 ^a	3.96±0.45 ^a	3.44±1.19 ^{ab}	3.04±1.06 ^b

^{abc} Means ± SD in the same row with different superscripts are significantly different (p≤ 0.05)

4.4 Acceptance test of the product

The sensory assessment was based on a target consumers of 200, categorized as 100 general consumers and 100 vegans, using a 5-point hedonic scale method on appearance, color, aroma, taste, texture, and overall liking. The consumer ages ranged from 18 - 65 years old.

It was found that vegan consumers accepted vegan meat and commercial vegan meat at the same level of 4 or “most-likeness” for all attributes. However, general consumers accepted the products in all attributes with a lower mean score of 3. The vegan group was familiar with vegan consumption, thus, accepting the texture of vegan meat.

For vegan eggs, it was found that the vegan group rated the preference for vegan eggs and commercial vegan eggs of all attributes in the level of “most-likeness” but preferred the color of the omelet from the vegan eggs more than the color of the commercial vegan eggs. For general consumers, it was found that they preferred the taste of the vegan eggs more than the commercial vegan eggs, rated for appearance, color, aroma, and texture at the level of “most-likeness,” while no comment on the aroma, taste, texture and overall preferences of the commercial vegan egg. However, it can be inferred that the consumers in the vegan group and the general group accepted the vegan egg as not inferior to the commercial vegan egg. The feature that needed to be improved was the texture.

4.5 Shelf-life study of the products

The shelf-life assessment of vegan meat was studied in three different packagings: transparent plastic bag, laminate bag, and aluminum foil bag, and different packings under normal pressure and vacuum. The storage temperature was

in a freezer with a clear glass door at -18°C for 18 months for the total microorganisms test and rancidity analysis.

To assess the shelf life of vegan egg powder, which had a longer shelf life than vegan meat, the short-established ASLT method was used to predict the shelf life of various storage products. The storage conditions at 45°C and 55°C were selected as the accelerated temperature conditions and then used to test the product quality. To determine the product storage time, the vegan egg powder was packed in a zip-locked transparent plastic bag under normal pressure and was stored at accelerating temperatures of 45 °C and 55 °C for 12 months. The samples were taken every 3 months until 12 months by total microorganism test and rancidity analysis. The shelf-life assessment of the vegan meat was done in the same procedure.

Summary of microbial monitoring, the total microbial count of vegan meat and vegan egg was found less than the maximum permissible threshold. No increase in the total microorganisms was observed over their shelf life (18 months for vegan meat and 12 months for vegan egg). Besides, it was found that, when increasing the storage period for a longer time, the total number of microorganisms tended to be constant.

The findings of 2-thiobarbituric acid (TBA) content as an index of rancidity in the vegan meat showed no change over 18 months of the shelf life, no difference in all three packaging types, and no difference in packing form. It could be said that any form of packaging can be chosen without interrupting the costs and without the need for vacuum packing, as shown in Figure 4.

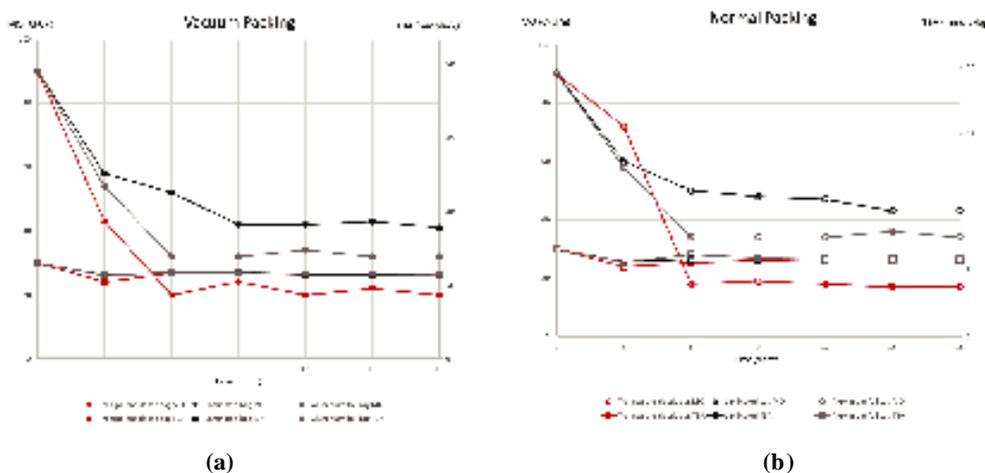


Figure 4 Total microorganism count and TBA value of vegan meat in three different packaging types by different packing; (a) vacuum and (b) normal packing storages at -18°C for 18 months.

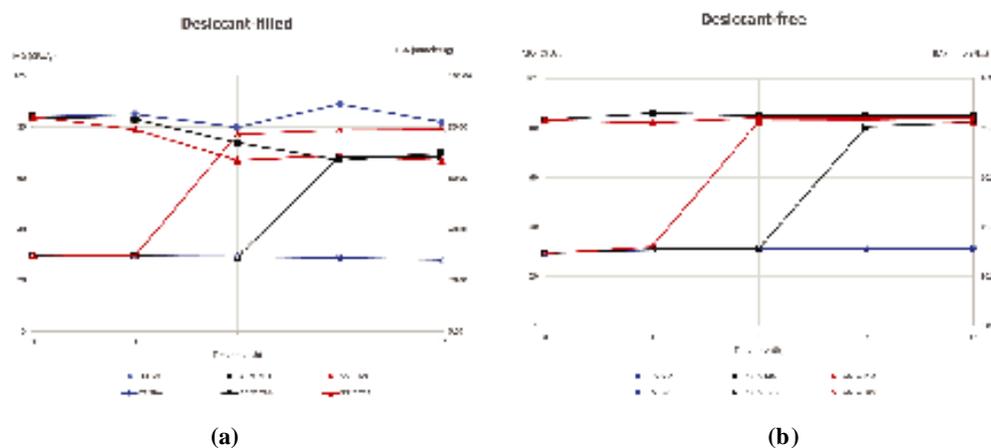


Figure 5 Total microorganism counts and TBA values of vegan eggs at three different temperatures by different packing: (a) desiccant-filled and (b) desiccant-free storages for 12 months.

In Figure 5, the shelf-life assessment of the vegan eggs showed that the total microbial count was less than the maximum permissible threshold. There was no increase in all microorganisms over the 12 months and no difference in all three temperatures. Although no microbial change was observed over the 12 months of storage, changes in TBA were observed in accelerated conditions at 45 °C at month 9 and 55 °C at month 6. The TBA value above 60 mmole/kg is the level at which the consumers could feel the rancid smell of the product and no longer accept it. Rancidity is caused by the reaction between oxygen and polyunsaturated fatty acid, resulting in many off-odor substances (Ottaway, 1993). This reaction was also associated with a reduction in the amount of vitamin B1, the main vitamin found in rice and used as the principal raw material. The results also indicated that the vegan eggs could be packed in zip-lock bags without the need for desiccant.

According to the shelf-life study of vegan egg powder by the ASLT method using a Q10 factor of 1.5, the Q1 value can be calculated to predict a shelf life at other temperatures. The results showed that the shelf-life of the vegan egg powder could last from 1.3- 5 years (496-1674 days), depending on the storage temperature. The product can be kept longer at lower storage temperatures. The results of this study enable a comprehensive logistics plan for the benefit of the business.

Our innovative allergen-free vegan meat and egg products were purposely designed to be

consumer-friendly and to combine plant-based food with a functional food. Besides, they were not contained allergenic ingredients such as gluten or soy protein like the commercial products. The commercial allergen-free vegan egg offers 2 optional flavoring agents to the consumer; flavoring agent formula 1 which composes rice protein isolate powder mixed with yellow color from pumpkin powder and flavoring agent formula 2 which composes rice protein isolate mixed with red color from dragon fruit. The allergen-free vegan meat costs 400 Baht per kilogram, which is cheaper than the commercial vegan meat brands imported from overseas as detailed in Table 4. The allergen-free vegan meat is sold in form of minced beef and burger packed in a non-vacuum seal plastic bag and is kept frozen (Figure 6a). The new products are introduced to the target consumers through social media platforms, promotion booths, and events at a lower price compared with similar domestic or imported products found in the market and delivered using the distribution channels in the form of consignment and online. The package of the vegan eggs is designed eco-friendly by packing bags of powder (24g/bag) in 6-egg cartons (Figure 6b). The cost of the allergen-free vegan egg is 81 Baht per pack, which is cheaper than the commercial vegan egg brands that are all imported from overseas. It will be introduced to the customer through social media or by presenting at events and will be sold in both convenience stores and online stores.

Table 4 Comparison of allergen-free vegan meat and vegan egg product with commercial products sold in Thailand

¹ Product Brands	Product Appearance	² Storage	Compared Items									
			content/serve							Allergen-Free	GMO-Free	Price
			Source of protein	Wt./Vol.	Protein (g)	Fat (g)	Fiber (g)	Kcal	Sodium (mg)			
THB/Kg												
MEAT												
Beyond Meat (USA)	Mince Burger	FZ	Pea protein, Rice protein	100g	17.7	15.9	1.8	232	309	√	√	1,430
Impossible Meat (USA)	Mince	FZ	Soy protein, Potato protein	100g	16.8	12.4	2.7	212	327	X	X	874
Omni Meat (HK)	Mince	FZ	Soy,Pea & Rice protein, Shitake	100g	12.5	0.8	4.6	69	340	X	√	400
More Meat (TH)	Mince Burger	FZ	Soy protein	100g	16.0	5.0	4.0	130	180	X	X	445
Let's Plant Meat (TH)	Mince	FZ	Soy protein	100g	13.3	8.8	4.0	221	301	X	X	664
Mud Jai Meat (TH)	Burger	FZ	Soy, Mushroom (splitgill)	100g	13.3	8.8	2.7	220	300	X	X	1,225
Meat Avatar (TH)	Mince	FZ	Soy protein, Wheat protein	100g	17.0	1.7	5.8	113	40	X	X	537
Meat Zero (TH)	Mince Burger	FZ	Soy protein	100g	11.0	7.0	5.0	140	210	X	√	340
³ PEN MEAT (TH)	Mince Burger	FZ	Rice protein	100g	17.0	1.0	6.0	161	5	√	√	400
THB/g												
EGG												
HOBOTAMA (JP)	Liquid, Yellow	FZ	Soy	100g	2.3	13.6		178	0.8	X	X	2.10
Just Egg (USA)	Liquid, Yellow	RF	Mung bean protein	44ml	5.0	5.0	0	70	170	√	√	0.70
Simply Eggless (USA)	Liquid, Yellow	RF	Lupin	67ml	3.0	4.0	1.0	50	20	X	√	0.44
Follow Your Heart (USA)	Powder, Yellow	TA	Soy	10g	3.0	1.0	4.0	35	150	X	X	4.09
Bob's Red Mill (USA)	Powder, Cream	TA	Potato	10g	0	0	1.0	30	320	√	√	2.89
	Powder, Dark yellow	TA	Soy, Algae	28g	20	2.0	0	110	290	X	X	6.85
ORGAN (AUS)	Powder, Dark yellow	TA	Chickpea, vegetable protein	50g	7.5	2.0	8.0	175	1010	X	√	0.74
⁴ PEN EGG (TH)	Powder, Cream	TA	Rice protein	24g	7.7	0.5	2.5	75	60	√	√	0.32

¹Product Brands = Brand name (Manufacturing Country)

²Storage condition ; FZ=Frozen , RF= Refrigerate , TA= Ambient Temperature

³PEN MEAT and ⁴PEN EGG are vegan meat and vegan egg from our research.

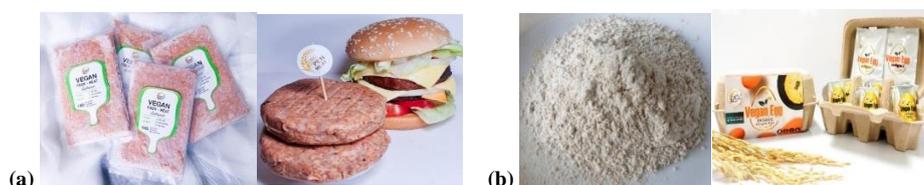


Figure 6 Allergen-free vegan meat and egg from our research.

(a) Allergen-free vegan meat; minced and burger and (b) allergen-free vegan egg powder and packaging.

5. Conclusion

The vegan meat and egg products obtained from this research are allergen-free, plant-based, and beneficial as nutrient-rich food. These novel products of vegan meat and egg are developed mainly from rice-based raw materials delivering low fat and high fiber with fewer food additives than the commercial products. The nutritional values of the products are equivalent to international commercial products. In terms of energy conservation and environmental friendliness, employing by-products from rice processing to make vegan meat and vegan eggs will help businesses save energy and money and will also alleviate the problem of storing space for rice processing by-products or wastes. The manufacturing of the allergen-free vegan meat and eggs requires only a few pieces of equipment, resulting in a reduction in electric energy and production costs. Finally, the results obtained from this research should be of benefit in increasing the value of rice processing by-products and can continue as a commercial for the community. The innovative products from this research have already been scaled up to a commercial scale in Thailand. The production process increases the value-added to the Thai rice and related Thai rice products, sustaining the Thai businesses. They have the potential to scale up as a global health innovation and bring a reputation to Thailand.

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