

## Mushroom keropok lekor, an innovative food product

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### Abstract

Keropok lekor is a popular Terengganu heritage traditional snack. Mushroom keropok lekor was developed as an alternative variation of nutritious products combination high in dietary fibre and protein. The aim of this study is to develop the mushroom keropok lekor from grey oyster mushroom (*Pleurotus sajor caju*) and investigated its physicochemical properties and shelf life. The boiled mushroom keropok lekor contained a good source of dietary fibre (3 g/100 g), protein (7.9 g/100 g), low in fat (0.5 g/100 g) and a cholesterol-free product. It contained 1.1% of polyunsaturated fatty acid (PUFA). The addition of mushrooms to fishery products improved physically the characteristic of minced fish products and enhance their nutritional value. The sensory evaluation of mushroom keropok lekor (T4) showed that the attribute of taste, texture, appearance-shape, colour, and overall acceptance is more than 4.0 with an interval scale (0-5). The shear force (N) for optimum formulation (T4) 3.75 newton was significantly different from the control sample 6.61 newton. The mushroom keropok lekor had a soft texture compared to conventional keropok lekor in the market. The shelf-life study showed that the total viable count was  $1.2 \times 10^2$  CFU/g, *Escherichia coli*, *Staphylococcus*, yeast, and mould was not detected after 24 months of frozen storage.

## 1. Introduction

Keropok lekor is found in Malaysia primarily in Terengganu and other state are Kelantan, Kedah, Pahang, and Johor. Keropok lekor is one of the popular traditional snacks on the East Coast of Peninsular Malaysia, for Malay people in Kelantan it is called *Keropok Batang*. It is traditionally made by mixing fresh meat from round scad 'selayang' (*Decapterus punctatus*) or fringescale sardinella 'tamban sisek' (*Chupea fimbriata*) with sago, tapioca flour and seasoning: salt, monosodium glutamate and sugar. Keropok lekor was formed into rolls of 6-12 inches (50-100 g) and cooked in boiling water (Che Rohani, 2013). The *keropok lekor* is consumed after being cooked either boiled, steamed, or fried. It is slightly greyish and had a fishy smell and prominent fish taste as it cools down after frying.

Recently, the innovation of keropok lekor was done by adding a variety of cheese and flavours, name as keropok lekor cheese, breaded keropok lekor cheese, and black paper keropok lekor. Keropok lekor cheese has been modified to form a ball and improved the taste and formulation for extending the shelf life.

In 2019, the landing of the capture of fish was 1,455,446 metric tonnes with a value of RM 11,336.32 million. This record had an increase in value by 0.21% compared to the previous year (1,452,862 metric tonnes) and a value of RM 11,312.83 million in 2018. (Department of Fisheries Malaysia, 2019). Agrofood Statistics 2019 reported that the export value for fish products was RM 18.3 million decreased by 31.7% compared to the export value in 2018 (RM 26.8 million) (Ministry of Agriculture and Food Industries, Malaysia, 2019). The imported fisheries product in 2019 increased by 16.4% (RM 82.8 million) compared to 2018, RM 69.2 million. This showed that the fisheries products for consumption in Malaysia had increased.

The quality of the products produced depends on the freshness of the fish, the type of fish used, the ratio of the main ingredients with flour (fish meat: flour) as well as the processing method. A good quality keropok lekor is determined by its taste and flavour, texture, and crispness. It comes in two main forms, lekor which is resembling like dough (sausage) and chewy (boiled or deep-fried). It is frequently served with dipping sauces. Boiled keropok lekor sometimes has some fishy smell,

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especially when they are no longer hot.

Consumers usually purchase keropok lekor from a local manufacturer, at restaurants, hawkers' stalls, and night markets among the local within their locality. Nowadays keropok lekor is widely known among Malaysians but has not been largely exposed to the tourists who visited Malaysia unless the tourists are introduced during their trip to Terengganu. Despite the popularity of keropok lekor in the country, this product is currently lacking in commercial value. Therefore, this product may need some form of innovation to attract foreign tourists as well as to sustain its market in the future (Omar *et al.*, 2011).

Traditionally, keropok lekor is cooked by boiling it in water. Problems such as bacterial activities resulting a slime on the surface of keropok lekor for about half day and mould can grow the next day. Boiled keropok lekor has a short shelf-life of fewer than 24 hrs at room temperature (Che Rohani and Mat Arup, 1999). Che Rohani and Mat Arup (1995) showed that the total viable count of bacteria in keropok lekor increased from less than  $1 \times 10^2$  CFU/g to  $1.5 \times 10^8$  CFU/g after 2 days at room temperature. Signs of spoilage for this product include sliminess and the formation of spots on the surface, which are resulted from bacterial growth.

Ready-to-eat keropok lekor and dipping sauce collected from 8 premises in Terengganu contained aerobic plate counts in the 1.8 to 5.5  $\log_{10}$  CFU/g and <1.0 to 5.1  $\log_{10}$  CFU/g range, respectively. Total coliforms were detected as unsatisfactory levels ( $> 1.7 \log_{10}$  CFU/g) in three keropok lekor samples. *Escherichia coli* was found in 10.29% of the samples and all of them were non-diarrheagenic serotypes (Wan-Hamat *et al.*, 2019). The microbiological quality of thirty ready-to-eat (RTE) keropok lekor was evaluated in comparison to microbiological guidelines for ready-to-eat foods. Results revealed that four samples (13.33%) contained Enterobacteriaceae counts that exceeded the recommended allowable counts of 4.0  $\log_{10}$  CFU/g. An unsatisfactory level of coliforms ( $> 1.7 \log_{10}$  CFU/g) was also observed in ten of the samples, two of which contained *E. coli* ( $2.1 \pm 0.17$  and  $3.7 \pm 0.02 \log_{10}$  CFU/g), suggesting poor hygiene and sanitation practice (Wan-Hamat *et al.*, 2020).

Mushrooms have been used in folk medicine and one of the sources of protein diets for thousands of years. The medicinal effects of mushrooms include antioxidant, antiviral, antibacterial, antifungal, detoxification, immunomodulatory, antitumor, radical scavengers, anti-inflammatory, anti-hypercholesterolemic, hepatoprotective, and antidiabetic (Wasser, 2001). In Malaysia, oyster mushroom has been shown to have high

-quality proteins, vitamins and medicinal values and are widely cultivated. It has been identified as one of the Beginning Projects (EPP) under the National Key Economic Area (NKEA) which involved massive mushroom cultivation in Malaysia. Mushroom production is estimated to increase by 16% per annum from 15,000 tonnes in 2010 to 67,000 tonnes in 2020 (Mohd Irwani *et al.*, 2013).

This mushroom is mostly popular in countries such as India, China, and Japan and is reported to be able to reduce the cholesterol level in the blood (Schneider *et al.*, 2011) and prevent hyperglycemia, insulin resistance, and inflammation in adipose tissue (Kanagasabapathy *et al.*, 2012). At the present, the use of mushrooms as the main ingredient of product food is limited and mostly sold as fresh products. *Pleurotus* mushroom is rich in fibre yet low in calories and fat. The dietary fibres in the mushroom consist of chitin, hemicelluloses, mannans, and  $\beta$ -glucans. Beta-glucans are polysaccharides with glucose residue linked by beta-glycosidic bonds. The fermentability of  $\beta$ -glucans and their ability to form highly viscous solutions in the human gut may constitute the basis of their anti-obesity benefits (Khoury *et al.*, 2012).

Even though the mushroom industry is expanding, it is now experiencing several problems. Fresh mushrooms are characterized by a short shelf life linked to postharvest changes. Mushroom production also faces the problem of seasonal glut, especially during the rainy season. Food processing could prolong the shelf life of perishable vegetables such as mushrooms. Therefore, efforts should be taken to develop the mushroom industry through research and development not only on the cultivation aspect but also the development of products from local mushrooms.

If the traditional food is well known, the products may have demand from a foreign country and can be exported to foreign countries. By using modern technologies to replace traditional techniques, manufacturers could produce a more hygienic way of processing and preserving food (De Roest and Menghi, 2000). Thus, there is an urgent need to upgrade the processing parameter of traditional foods like keropok lekor to be ready-to-cook product, more hygienic and be one of the menus for the fast-food category. In Europe, the registration of traditional foods possibly will motivate their small-scale production throughout the region and expand their export potential to other countries (Tregear *et al.*, 2007). Moreover, according to Trichopoulou *et al.* (2007), the mixture of 'healthy and palatable' is very attractive to the food industry and traditional foods could potentially be mass-produced and exported.

## 2. Materials and methods

### 2.1 Materials

New product innovation by using mushrooms in the ingredients of traditional keropok lekor was used to compare with the traditional keropok lekor in the markets. The formulation of mushroom keropok lekor was designed using a simple lattice design, there are four treatments of mushroom keropok lekor added with a variation percentage of mushroom (15-45%) and one control sample (without mushroom) (Table 1). The formulation of mushroom keropok lekor had been registered as a trade secret with intellectual property Malaysia (MYIPO), number: MDI/SI/SI01/PA/073/5/58. The mushroom keropok lekor processing principle is a frozen food made from minced fish, the main ingredients are mixed with mushrooms from the type of grey oyster mushroom (*Pleurotus sajor caju*) and other materials such as sago flour, salts are blended during the mixing process then roll to formed into sausage shape 6 inches with 50 g each and cooked in boiling water. Frozen foods are defined as foods that have been reduced to at least -18°C at the centre of the food. Frozen products do not require the addition of preservatives because the growth of microorganisms is retarded when the food is kept frozen at temperatures below -18°C.

### 2.2 Determination of fatty acid profile and polyunsaturated fatty acid.

The sample of oil 100 g was first hydrolyzed with methanolic Potassium hydroxide (KOH). Hydrochloric acid (HCL) was added to release the fatty acids, which were then extracted with heptane. The fatty acids were then injected into the GC with a bicyanopropyl polysiloxane capillary column (30 m × 0.25 μm × 0.20 μm). The chromatographic condition for the determination of fatty acids was set with the column temperature at 180°C, injector temperature at 230°C and flame ionization detector temperature at 250°C, IUPAC 7<sup>th</sup> edn II.D.19/II.D.25 (Dieffenbacher and Pocklington, 1991).

Table 1. Treatment of mushroom fish sausage

Ingredients	Control	T1 (keropok lekor with mushroom) (%)	T2 (keropok lekor with mushroom) (%)	T3 (keropok lekor with mushroom) (%)	T4(keropok lekor with mushroom) (%)
Selayang meat	ND	ND	ND	ND	ND
Sagu flour	ND	ND	ND	ND	ND
Salt	ND	ND	ND	ND	ND
Water with ice	ND	ND	ND	ND	ND
Ten ingredients registered as trade secret	ND	ND	ND	ND	ND
Total	100	100	100	100	100
Mushroom	0	ND	ND	ND	ND

ND: Non-disclosure, the formulation had been registered as trade secret at Intellectual Property Malaysia MYIPO with number: MDI/SI/SI01/PA/073/5/58.

### 2.3 Microbiological analysis

Total viable count (TVC) was determined according to McLandsborough (2005). Approximately 25 g of sample was aseptically weighed and transferred to a sterile stomacher bag, 225 mL of 0.1% sterile peptone water (Merck, Germany) was added to make a 10<sup>1</sup> dilution and homogenized for 30 s at 230 rpm using a paddle blender (Seward Stomacher model 400, England). Homogenized samples were then subjected to determine the total viable count. Serial dilutions were made, and 1 mL of each appropriate dilution was poured plated using plate count agar (PCA) and sterile petri plate, allowing the agar to solidify. All the plates were inverted and incubated at 35±2°C for 48 hrs. Plates showing 25–250 colonies were counted.

For samples analyzed using Compact Dry 'Nissui' and 3M<sup>TM</sup> Petrifilm method, 25 g of sample was aseptically weighed and transferred to a sterile stomacher bag, 225 mL of 0.1% sterile peptone water (Merck, Germany) was added to make a 10<sup>-1</sup> dilution and homogenized for 30 s at 230 rpm using paddle blender (Seward Stomacher model 400, England). Homogenized samples were then subjected to determine the total viable count and other pathogens. Approximately 1 mL of the specimen was pipetted and perpendicular on the middle of the dry sheet of the Compact Dry plate or 3M<sup>TM</sup> Petrifilm. For *E. coli* and *Staphylococcus aureus* (meat, poultry, and seafood AOAC 998.08), the 3M<sup>TM</sup> Petrifilm for *E. coli*/Coliform Count and *S. aureus* plate was incubated for 24±2 hrs at 35±1°C, the blue colonies with gas were count as *E. coli* and red or blue colonies with gas was count as coliform. The 3M Petrifilm Staph Express disk should be used whenever colonies other than red-violet are present on the plate, for example (black or blue-green colonies) as they may obscure *S. aureus* (black colonies may or may not be *S. aureus*, blue-green colonies are not *S. aureus*). *Staphylococcus aureus* colonies may vary in size, count all red-violet colonies regardless of size. Compact dry for yeast and

mould were incubated for 3-7 days at 20-25°C, on day 3 of incubation, blue, white or cream with clear boundaries was observed for yeast and on day 7 the cottony colonies with a characteristic colour (large colonies) was observed for mould.

The microbiological limit was based on the guideline for ready-to-eat or ready-to-cook shrimp at the point of sale (Table 2) and the recommendation of microbiology limit for seafood products (Table 3). The total viable count was limited to  $<10^5$  for seafood products and  $<20$  CFU/g for *staphylococcus aureus* was under the satisfactory category at the point of sale.

#### 2.4 Texture analysis

Shear force determination was carried out at  $25\pm 2^\circ\text{C}$ . The maximum force required to cut the sample (Newton) was recorded. The sheer force of samples 25 g was measured using a texture analyser (EZ test shimadzu) using a fish ball probe. The operating parameters used are crosshead speed of the machine 50 mL/min, load stress 20 Newton. The measurement was replicated ten times for each sample with two replicates.

#### 2.5 Chemical analysis

Approximately 100 g of the sample was homogenized before the proximate analysis. Moisture content was determined by drying a known amount of

sample in an air oven at  $100\pm 2^\circ\text{C}$  for 5-12 hrs to constant weight AOAC 950.46 (AOAC, 2005). Protein was determined by the Kjeldahl Method AOAC 928.8 (AOAC, 2005). The crude fat content of the sample was extracted using Solvent Extraction Method AOAC 991.6 (AOAC, 2005). Ash content was determined by heating at  $550^\circ\text{C}$  for 4-5 hrs using a muffle furnace AOAC 920.153 (AOAC, 2005). All analysis was done in two replicates. Total dietary fibre AOAC 2009.01 (AOAC, 2005). All analyses were done in triplicate.

#### 2.6 Rancidity (peroxide value)

The most common chemical method of measuring the oxidative deterioration of oils. Although hydroperoxides decompose to a mixture of volatile and non-volatile products and they also react further to endoperoxides and other products, the peroxide value measurement is a useful method of monitoring the oxidative deterioration of oils. The sample 10 g was weighed into a clean dry boiling tube and 1 g of powdered potassium iodide, and 20 mL of solvent mixture (2 volumes of glacial acetic acid plus 1 volume of chloroform) were added. The tube was placed in boiling water and the sample was boiled within 30 seconds. Pour the contents quickly into a flask containing 20 mL of potassium iodide solution (5%), wash out the tube twice with 25 mL of water and titrated with 0.002 M sodium thiosulphate solution using starch

Table 2. Guideline of microbiological quality ready-to-eat or to be cooked shrimp for chilled and frozen storage at the point of sale

Criteria	Microbiology quality (CFU/g)		
	Satisfactory	Acceptable	Unsatisfactory
Total viable count	$<10^5$	$10^5 - <10^6$	$>10^6$
<i>Listeria</i> spp. (total)	$<20$	$20 - <10^3$	$\geq 10^3$
<i>Escherichia coli</i> (total)	$<20$	$20 - <100$	$\geq 10^2$
Faecal Coliform	$10^2$	$10^2 - 10^3$	$> 10^3$
<i>Staphylococcus aureus</i>	$<20$	$20 - <100$	$10^2 - <10^4$
Yeast	$<10^4$	$10^4 - 10^6$	$>10^6$

Source: Stannard (1997); Gilbert, Roberts and Bolton (2000)

Table 3. Recommended microbiological limits for seafood

Product	Test	n	c	Limit per gram	
				m	M
Prawns and shrimps (raw, frozen)	Aerobic plate count at $35^\circ\text{C}$ (/g)	5	2	$5\times 10^5$	$5\times 10^6$
	Coagulase producing <i>Staphylococcus</i> (/g)	5	2	102	103
	Faecal coliform (/g)	5	2	102	103
	Salmonella (/25 g)	5	0	0	0

n: Number of representative sample units.

c: Maximum number of acceptable sample units with bacterial counts in m and M.

m: Maximum recommended bacterial counts for good quality products.

M: Maximum recommended bacterial counts for marginally acceptable quality products.

Plate counts below "m" are considered good quality. Plate counts between "m" and "M" are considered marginally acceptable quality but can be accepted if the number of samples does not exceed "c." Plate counts at or above "M" is considered unacceptable quality.

Source: ICMSF (1986), Food Administration Manual (1995)

as an indicator (1%). A blank should be performed at the same time AOAC 965.33 (AOAC, 2005). Peroxide value is measured in milliequivalent of peroxide per 100 g sample.

$$\text{Peroxide value} = \frac{\text{Titration value} \times \text{Normality of acid used} \times 100}{\text{Weight of sample used}}$$

### 2.7 Sensory analysis of mushroom fish sausage

The sensory analysis of the products was evaluated by 20 trained panels with two replicates. The sensory characteristics of the processed mushroom fish sausage were assayed by Quantitative Descriptive Analysis (QDA) (Aminah, 2000) simple descriptive using five category scales (numeric 0 to 5) to know the intensity of every sensory attribute from weaker to stronger. A score above 4 overall will be considered as the margin for the selection of the highest score for all the attributes of the product. The sensory attributes covered under the taste panel are 1) appearance: colour, 2) taste 3) texture: tenderness 4) shape 5) overall acceptability.

The optimum formulation of mushroom fish sausage will be selected according to the acceptability in sensory evaluation, dietary fibre, and result from texture analysis. The optimum formulation will be compared with commercial keropok lekor. Product specification and processing parameters were determined. The optimum formulation was verified using verification analysis.

#### 2.7.1 Selection and training of sensory panels

The sensory evaluation training session followed the method of training panels from Watts *et al.* (1989), Aminah (2000) and Rehbein and Oehlschlager (2009). For the training of sensory panels, the questionnaire was distributed to find the panellists that keropok lekor and did not follow any diet. The panellists were selected among Food Technology officers and staff who were willing to do the sensory evaluation. The training of sensory panels begins by describing the procedures of the sensory evaluation and what is expected of the panellists. The nature and limits of the sense organs are described, such as the importance of breathing deeply and resting between samples during odour evaluation. In the first training session for the basic taste recognition test, the sensory panels were given a questionnaire with samples (salt, sugar, citric acid, and caffeine) solution to test the sensitivity of their taste bud (Table 4). The answer was given on the same day. If the panels failed the test, they could repeat it for a second time.

The second training session was for the QDA training session, 4 hrs of the training session was conducted two times on different days. In the training session the attributes were elaborated to the panels and the panels were asked to evaluate 4 samples for one

session and a total of eight samples of mushroom keropok lekor were given to the panels in the training session.

The panels gave their feedback on what they understood about every attribute in the sensory evaluation score sheet. After that, the sensory evaluation form was developed according to the attributes that the panels understood and suitable to evaluate the mushroom keropok lekor. The sensory analysis was done by twenty trained panels with the age from 25 to 45 years old, 15 panellists were married, and the rest were single.

Table 4. Basic taste for recognition test

Basic taste	Substance	Concentration
Sweet	Sucrose	1.0%
Salty	Sodium Chloride	0.2%
Sour	Citric acid	0.04%
Bitter	Caffeine	0.05%

Source: Watts *et al.* (1989)

### 3. Results and discussion

Mushroom keropok lekor was an innovative product from grey oyster mushroom (*Pleurotus sajor caju*) which is mixed with other ingredients. It gave a soft texture with a moisture content of 68.7% after 24 months at cold storage -20°C (Figure 1). This mushroom keropok lekor is a good quality product as the ingredients used can hold mushroom mixtures with other ingredients during frozen storage and after cooking. The shelf life of the product was up to 24 months, the total viable count was  $1.2 \times 10^2$  CFU/g, *E. coli*, *Staphylococcus*, yeast and mould were not detected at cold storage -20°C (Table 5). Chilled boiled fish sausage (keropok lekor) was introduced in 1995, and the shelf-life extended to seven days at 2°C or 30 days when the product containing 3% sodium lactate (vacuum-packed and pasteurized before storage (Che Rohani and Mat Arup, 1995). Later, frozen keropok lekor was introduced to local processors, the shelf life was extended to more than 10 months at -18°C (Che Rohani, 2001). Some of the entrepreneurs used the steam process to cook keropok lekor, but it took about 2 hrs, for boiled keropok lekor it took about 45-60 minutes. These methods involved high energy costs for about 65%

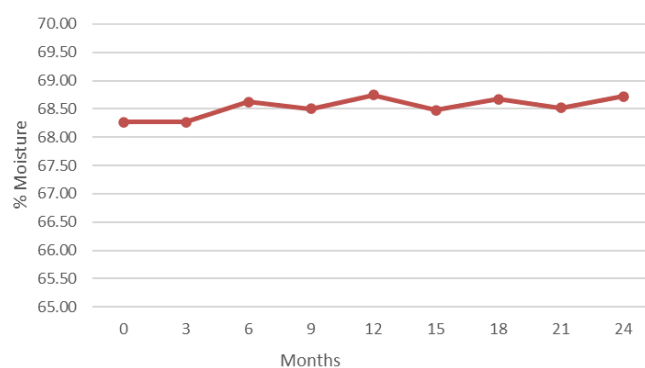


Figure 1. Moisture of mushroom keropok lekor for 24 months

Table 5. Microbiological analysis for 24 months

Months	Total Plate Count (CFU/g)	Yeast and Mould (CFU/g)	<i>Staphylococcus aureus</i> (CFU/g)	Coliform ( <i>Escherichia coli</i> )* (CFU/g)
0	< 1.0×10	ND	ND	ND
3	< 1.0×10	ND	ND	ND
6	< 1.0×10	ND	ND	ND
9	< 1.0×10	ND	ND	ND
12	< 1.0×10	ND	ND	ND
15	< 25×10 (2.0 ×10)	ND	ND	ND
18	< 25×10 (2.0×10)	ND	ND	ND
21	< 25×10 (4.0×10)	ND	ND	ND
24	< 25×10 (1.2×10 <sup>2</sup> )	ND	ND	ND

ND, Not detected in 25 g sample.

of the total production cost (Yu and Low, 1992).

The optimum formulation mushroom keropok lekor (T4) contains a good source of dietary fibre (3 g/100 g),  $\beta$ -glucan 0.2 g/100 g, protein (7.9 g/100 g) and fat (0.5 g/100 g) (Table 6). The product (100 g) contains 1.10 g polyunsaturated fatty acid (0.5 g omega 3 and 0.59 g omega 6), 0.3 g DHA and 0.08 g EPA. According to the study by Kanagasabapathy *et al.* (2013),  $\beta$ -glucan is an agent of anti-cholesterol, this  $\beta$ -glucan prevents obesity and oxidative stress. Katakura *et al.* (2012) wrote that polyunsaturated fatty acids (PUFAs) can induce neurogenesis and recovery from brain diseases. Polyunsaturated fatty acids (PUFAs) are critical for the developing brain and are classified into omega-3 PUFAs, such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), and omega-6 PUFAs, such as arachidonic acid (AA). Omega-3 (n-3) fatty acid docosahexaenoic acid (DHA) benefits in reducing risk for cardiovascular disease, its role in the resolution of inflammation, its importance in cognitive function in infants and inhibiting the progression of neurodegenerative diseases in the elderly (Whelan *et al.*, 2009).

Dietary fibre is a group of compounds containing carbohydrate polymers and non-carbohydrate components. Dietary fibre has gained considerable attention in recent years due to its potential role in

improving human health. For instance, it has been shown to prevent heart disease, obesity, and cancers (Elluech *et al.*, 2011; Huang *et al.*, 2013). Dietary fibre from mushrooms could enhance the texture and at the same time increased the value of traditional snacks considerably, both in the physical and nutritional of products.

James *et al.* (2009), reported that dietary fibre significantly lowers the risk of developing coronary heart disease, stroke, hypertension, diabetes, obesity, and certain gastrointestinal diseases. Several large-scale, randomized clinical trials have shown that dietary intake of omega-3 PUFAs improves the prognosis of patients with symptomatic heart failure or recent myocardial infarction. Therefore, dietary consumption of omega-3 PUFA is recommended in international guidelines for the general population to prevent the occurrence of cardiovascular diseases (CVDs) (Endo and Arita, 2015).

The peroxide value of mushroom keropok lekor with optimum formulation was 0.31 meq/kg (Table 7), and the maximum limit of peroxide value is 30 meq/kg for fish products (Pearson, 1962). This new innovative technology could make a variety of fish products which rich in nutritional value. Jamilah (1983) wrote that steaming of keropok lekor presently did not prove to be feasible. However, it is suggested that steaming can be adopted in the precooking process due to the overall

Table 6. Nutritional facts of mushroom fish sausage

Treatment	Energy (kcal)	Protein (%)	Fat (%)	Carbohydrate (%)	Dietary fibre (%)	Moisture (%)	Ash (%)	Peroxide Value (meq)
Control	135	10.0±0.2	0.1±0.03	23.5±0.2	0.1±0.03	64.8±0.2	1.6±0.03	0.35±0.03
T1	132	8.3±0.2	0.3±0.03	23.9±0.2	0.5±0.03	66.1±0.2	1.4±0.03	0.33±0.03
T2	133	7.9±0.3	0.2±0.02	24.9±0.2	1.1±0.03	65.2±0.3	1.8±0.03	0.34±0.03
T3	107	7.3±0.2	0.4±0.03	18.6±0.2	1.4±0.03	72.2±0.2	1.5±0.03	0.33±0.03
T4	113	7.9±0.2	0.5±0.03	19.3±0.2	3.3±0.03	70.6±0.2	1.7±0.03	0.31±0.03

Control: Keropok lekor without mushroom, T1-T4: Mushroom keropok lekor

lower microbial count of steamed samples ( $1 \times 10^2$  less than the boiled). Fried keropok lekor also tends to become stale and rancid if the manufacturer used recycled oil or used the same oil for a bulk of keropok lekor on the same day of manufacturing.

Table 7. Texture analysis of mushroom fish sausage

Treatment	Newton
Control	6.61±0.71 <sup>a</sup>
T1	6.41±1.30 <sup>a</sup>
T2	5.96±0.37 <sup>b</sup>
T3	4.57±0.19 <sup>c</sup>
T4	3.75±0.48 <sup>d</sup>

Values are presented as mean±SD. Values with different superscripts within the same column are significantly different ( $p > 0.05$ ). Control: Keropok lekor without mushroom, T1-T4: Mushroom keropok lekor

Quality evaluation of mushroom keropok lekor was analyzed using sensory evaluation for 20 trained panellists by quantitative descriptive analysis (QDA) tests. The results of the optimum formulation for mushroom keropok lekor (T4) showed that in terms of the attribute (taste, texture, appearance-shape, colour and overall acceptance) the score is more than 4.0 with an interval scale (0-5) (Table 7).

While the result for texture analysis of mushroom keropok lekor using texture analysis (EZ test shimadzu) showed that, the texture of optimum formulation (T4) was 3.75 Newton (Table 8). The texture of mushroom keropok lekor was significantly different from control sample 6.61 Newton. The mushroom keropok lekor had a soft texture, and this product is one of the innovative products that differentiates from traditional keropok lekor.

Omar *et al.* (2011), reported that from 212 respondents that participated in his study, 70% of the respondents chose to eat keropok lekor because of the taste. Half of the respondents choose black pepper as an innovative supplementary flavour of keropok lekor. Therefore, a new value-added recipe that gives flavour for keropok lekor needs to be studied. These new frozen mushroom keropok lekor make a variety of fish products.

Table 8. Sensory evaluation of mushroom fish sausage

Treatment	Overall acceptability	Colour	Texture	Taste	Shape
Control	3.69±0.71 <sup>a</sup>	3.87±0.70 <sup>a</sup>	3.56±0.80 <sup>a</sup>	3.74±0.83 <sup>a</sup>	3.67±0.68 <sup>a</sup>
T1	3.93±0.61 <sup>a</sup>	4.02±0.63 <sup>a</sup>	3.94±0.69 <sup>a</sup>	4.02±0.64 <sup>a</sup>	3.91±0.63 <sup>a</sup>
T2	3.79±0.60 <sup>a</sup>	3.89±0.63 <sup>a</sup>	3.84±0.65 <sup>a</sup>	3.88±0.68 <sup>a</sup>	3.95±0.67 <sup>a</sup>
T3	3.99±0.53 <sup>a</sup>	3.99±0.58 <sup>a</sup>	3.90±0.56 <sup>a</sup>	4.10±0.53 <sup>a</sup>	3.92±0.57 <sup>a</sup>
T4	4.11±0.58 <sup>a</sup>	4.15±0.50 <sup>a</sup>	4.03±0.73 <sup>a</sup>	4.07±0.69 <sup>a</sup>	4.08±0.53 <sup>a</sup>

Values are presented as mean±SD. Values with different superscripts within the same column are significantly different ( $p > 0.05$ ). T1-T4: Mushroom keropok lekor.

#### 4. Conclusion

The mushroom fish sausage was the innovative product of grey oyster mushrooms (*Pleurotus sajor caju*) that are mixed with fish meat and other ingredients. It was an alternative healthier fish product in the market. The addition of mushrooms to fishery products not only improve the physically characteristic of minced fish products but also improve functional foods. Mushroom keropok lekor, contained a good source of dietary fibre (3 g/100 g), protein (7.9 g/100 g), low in fat (0.5 g/100 g) and a cholesterol-free product. In hundred grams of mushroom keropok lekor, it contained 1.10 g polyunsaturated fatty acid (PUFA). Promoting the health benefits of mushroom keropok lekor to the consumer, will contribute to the awareness of functional food products from fish in the market.

For the texture of mushroom keropok lekor, the shear force for optimum formulation (T4) 3.75 newton was significantly different from the control sample 6.61 newton. The mushroom keropok lekor had a soft texture with a moisture content of 68.7% after 24 months at cold storage  $-20^{\circ}\text{C}$  and this product is one of the innovative products that differentiate it from traditional keropok lekor. The shelf life of the product was up to 24 months at  $-20^{\circ}\text{C}$ , the total viable count was  $1.2 \times 10^2$  CFU/g, and *E. coli*, *Staphylococcus*, yeast, and mould were not detected.

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