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CHARACTERISTICS OF SARDINE FISH OIL EMULSION (*Sardinella* sp.) FLOUR BY-PRODUCT DURING STORAGE

Karakteristik Emulsi Minyak Ikan Sardin (*Sardinella* sp.) Selama Penyimpanan

Kristina Haryati

Fisheries Science Study Program, Cenderawasih University

Wolker Camp Street, Waena, Jayapura

*Corresponding Author: kristinaharyati40@gmail.com

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ABSTRACK

Sardine fish oil, which is mostly considered useless, is actually very beneficial because it contains various omega-3 fatty acids, especially EPA and DHA so that fish oil is made into an emulsion product. Emulsion is a two-phase system in which small droplets of one liquid are dispersed into another liquid. The purpose of this study was to determine the characteristics of the best emulsion formula for 30 days of storage. The methods in this study include the stage of determining the emulsion formula, and the emulsion testing stage. In making the emulsion formula, the types of emulsifiers used were Carboxymethyl Cellulose (CMC) and lecithin with a ratio of 1: 1; 1: 2; 3: 1; and 3: 2. The tests carried out included viscosity, pH, and percent stability. The results showed that the highest viscosity, pH, and percent stability tests during 30 days of storage were in formula 3: 1 and the lowest in formula 1: 2. During 30 days of storage, there was a decrease in the viscosity, pH, and emulsion stability values for all formulas. This finding shows that the best emulsion formula uses CMC and lecithin emulsifiers, namely 3:1. Thus, this study shows that sardine fish oil emulsion has the potential to be developed into an emulsion product using the existing formula.

Keywords: Carboxymethyl Cellulose (CMC), Emulsion, Fish Oil, Lecithin.

ABSTRAK

Minyak ikan sardine, yang sebagian besar dianggap tidak bermanfaat, sebenarnya sangat bermanfaat karena mengandung berbagai asam lemak omega-3, terutama EPA dan DHA sehingga minyak ikan dibuat menjadi produk emulsi. Emulsi adalah sistem dua fase di mana tetesan kecil dari salah satu cairan terdispersi ke dalam cairan lain. Tujuan dari penelitian ini yaitu mengetahui karakteristik formula emulsi terbaik selama 30 hari penyimpanan. Metode dalam penelitian ini meliputi tahap penentuan formula emulsi, dan tahap pengujian emulsi. Dalam pembuatan formula emulsi, jenis emulsifier yang digunakan yaitu Carboxymethyl Cellulose (CMC) dan lesitin dengan perbandingan 1:1; 1:2; 3:1; dan 3:2. Pengujian yang dilakukan meliputi viskositas, pH, dan persen stabilitas. Hasil menunjukkan bahwa pengujian viskositas, pH, dan persen stabilitas tertinggi selama penyimpanan 30 hari terdapat pada formula 3:1 dan terendah pada formula 1:2. Selama penyimpanan 30 hari, terjadi penurunan nilai viskositas, pH, dan stabilitas emulsi untuk semua formula. Temuan ini menunjukkan bahwa formula emulsi terbaik menggunakan emulsifier CMC dan lesitin yaitu 3:1. Dengan demikian, penelitian ini menunjukkan bahwa emulsi minyak ikan sardine memiliki potensi untuk dikembangkan menjadi produk emulsi menggunakan formula yang ada.

Kata Kunci: Carboxymethyl Cellulose (CMC), Emulsi, Lesitin, Minyak Ikan

INTRODUCTION

Fishery products are needed by the community to improve health because of their good nutritional content for the human body. One type of fishery product made to meet consumer desires is the processing of fish meal by-products into fish oil, which is expected to produce zero waste and reduce waste production in the sardine fish meal industry.

Sardine fish oil, which is largely considered to be of no benefit, is actually very beneficial because it contains various omega-3 fatty acids, especially EPA and DHA (Andhikawati *et al*., 2020; Dari *et al*., 2017; Manduapessy, 2017; Musbah *et al*., 2017; Pandiangan *et al*., 2023; Putri et al., 2022; Sarker, 2020). The omega-3 content in fish oil is very important for brain health and intelligence (Sarker, 2020), has mild and chronic cardiometabolic and pro-inflammatory effects (Gammone *et al*., 2019), and functions as an antioxidant and reduces triglyceride levels, which are the cause of free radicals (Fuentes et al., 2018).

Based on its benefits, fish oil is made into an emulsion product. Emulsion is a two-phase system in which small droplets of one liquid are dispersed into another liquid. Musbah *et al*. (2017) stated that emulsion technology is one method to suppress oxidative damage to oil. In the process of making emulsions, surfactants or emulsifiers are added so that the oil can be integrated with water. Various types of emulsifiers, both chemical and natural, are usually used to build and stabilize emulsions. The addition of surfactants to the emulsion can increase the kinetic stability of the emulsion by creating a protective layer between the droplets and reducing the tension between water and oil. Based on the description above, the characteristics of sardine fish oil emulsions as a source of omega 3, such as viscosity, pH, and percent stability of the combination of CMC and lecithin emulsifiers, then the emulsion was stored for 30 days and testing was carried out every two days.

RESEARCH METHODS

Time and Location of Research

This study was conducted from February to April. The sardine or lemuru fish oil used came from fish oil produced by the flour industry in Bali. The purification of fish oil to testing was carried out in several laboratories of the Bogor Agricultural University such as the Fish Oil Laboratory of the Department of Aquatic Product Technology, the Biochemistry Laboratory of the Department of Aquatic Product Technology, and the Physical Chemistry Laboratory of the Department of Chemistry of the Bogor Agricultural University.

Tools and materials

The tools used in the study were centrifuge (PLC Series), digital scales (Quattro), Stirrer (Coming PC-420 D), Brookfield Viscometer, pH meter model 410 A, vortex, electric stove, water bath, and other glassware. The materials used in this study were crude fish oil, distilled water, Sodium Hydroxide (NaOH) (Merck), magnesol XL, Carboxymethyl Cellulose (CMC), lecithin, essence, benzoate.

Purification Stage

At this stage it is divided into 3, namely the degumming process which is done by pouring 100 mL of oil into a beaker glass then heating it to a temperature of 50°C. Add 2 mL of water to the hot oil while stirring for 10 minutes. Furthermore, the neutralization process is carried out on the oil by adding NaOH or caustic soda 20°Be while stirring for 20 minutes. The neutralized oil is centrifuged to separate the oil from other impurities including water. The last process is bleaching the centrifuged oil which is done by adding magnesol XL 5% of the oil weight. The oil is heated while stirring before adding magnesol. After magnesol binds the pigment and other unwanted components in the oil, centrifuge it. The result is fish oil that has undergone a series of purifications.

Formula Determination Stage

The purified fish oil is then used in determining the formula. In determining the formula, the types of emulsifiers used are CMC and lecithin, where the emulsifier ratio that is the focus of the study is the CMC and lecithin ratio of 1:1; 1:2; and 3:2 (w/w). The emulsifier ratio is grouped into the water phase and the oil phase, where the water phase is done by mixing CMC, distilled water, essence, and benzoate while homogenizing for 10 hours. While the oil phase is done by mixing 10% of the purified fish oil with lecithin while homogenizing for 1 hour. Furthermore, the oil and water phases are mixed, and the emulsion is stored at room temperature for 30 days.

Viscosity Testing Stage

The emulsion of the 4 formulas that have been made is then tested for viscosity every 2 days. The viscosity test of the sample is carried out by measuring a 60 mL emulsion sample then selecting the spindle for viscosity testing, turning on the viscometer and waiting for the rotor to read the number or viscosity value to stop. The value is expressed in cP (centiPoise) units.

pH Testing Stage

In addition to viscosity testing, pH testing was also carried out on emulsions from 4 formulas every 2 days. Sample pH testing was carried out by calibrating the tool first before the tool was used. After calibration, the tool can be used by dipping the electrode in the emulsion sample, leaving it for a few minutes until the number or value read is stable.

Stability Percentage Testing Phase

The percentage of stability is calculated if emulsion separation occurs using the formula:

$$\%\, stability = \frac{A - B}{B}\, x\ 100\%$$

Information: A = total volume height of emulsion (cm)
B = high volume creaming

Data analysis

The data were analyzed using SPSS with a Completely Randomized Design (CRD) with 2 replications. If significantly different, then continued with Duncan's Advanced Test.

RESULTS

Table 1 shows that the highest viscosity value during 30 days of storage was in formula 3:1 at 1300.67 cP; followed by formula 3:2 at 892.98 cP; formula 1:1 at 84.98; and the lowest was in formula 1:2 at 43.71 cP. The viscosity values shown in Table 1 generally show that during 30 days of storage, there was a decrease in the emulsion viscosity value for all formulas.

Tabel 1. Nilai viskositas formula emulsi selama penyimpanan 30 hari

Day	Nilai Viskositas			
	1:1	1:2	3:1	3:2
0	346.23 ± 14.31^{d}	122.69 ± 10.26^{e}	2991.66 ± 49.29^{h}	3245.33 ± 25.36^{h}
6	268.76 ± 84.05^{bcd}	110.11 ± 3.41^{cde}	2668.60 ± 376.46^{gh}	2420.55 ± 126.78^{ef}
12	191.60 ± 111.26^{abc}	100.85 ± 5.92^{bcde}	2035.55 ± 257.46^{def}	1541.24 ± 119.98^{cd}
18	131.28 ± 32.76^{ab}	93.38 ± 8.51^{bcde}	1669.72 ± 95.32^{abcd}	1032.19 ± 295.54^{ab}
24	94.84 ± 3.57^{a}	74.24 ± 1.06^{abcd}	1447.85 ± 12.55^{abc}	943.91 ± 61.73^{ab}
30	84.98 ± 0.05^{a}	43.71 ± 3.06^{ab}	1300.67 ± 0.64^{a}	892.98 ± 9.99^{a}

Note: Different superscript letters in the same column indicate significant differences ($p<0.05$). The experiment was repeated 2 times.

Table 2 shows that the highest pH value during 30 days of storage was in formula 3:1 at 4.71; followed by formula 3:2 at 4.44; formula 1:1 at 3.78; and the lowest was in formula 1:2 at 3.73. The pH values shown in Table 2 generally show that during 30 days of storage, there was a decrease in the pH value of the emulsion for all formulas, as was the case with viscosity.

Table 2. pH value of emulsion formula during 30 days of storage

Day	pH value			
	1:1	1:2	3:1	3:2
0	6.42 ± 0.38^{h}	6.20 ± 0.65^{k}	6.52 ± 0.41^{i}	6.36 ± 0.64^{h}
6	5.29 ± 0.16^{fg}	5.10 ± 0.04^{ij}	5.51 ± 0.01^{gh}	5.38 ± 0.04^{g}
12	4.87 ± 0.24^{def}	4.66 ± 0.04^{fgh}	5.16 ± 0.71^{def}	5.13 ± 0.11^{defg}
18	4.63 ± 0.26^{bcd}	4.46 ± 0.05^{defg}	5.03 ± 0.09^{abcde}	4.92 ± 0.11^{bcdefg}
24	4.26 ± 0.01^{b}	4.11 ± 0.13^{abcd}	4.85 ± 0.07^{abcd}	4.19 ± 0.58^{abc}
30	3.78 ± 0.01^{a}	3.73 ± 0.01^{a}	4.71 ± 0.02^{a}	4.44 ± 0.05^{abcdef}

Note: Different superscript letters in the same column indicate significant differences ($p<0.05$). The experiment was repeated 2 times.

Table 3 shows that the highest stability percentage value during 30 days of storage is in formula 3:1 at 96.30%; followed by formula 3:2 at 11.67%; formula 1:1 at 10.84%; and the lowest is in formula 1:2 at 9.31%. The stability percentage values shown in Table 3 show that there is a decrease in the emulsion stability percentage value for all formulas, as is the case with viscosity and pH.

Table 3. Percentage stability value of emulsion formula during 30 days of storage

Day	Emulsion stability % value (v/v)			
	1:1	1:2	3:1	3:2
0	100±0.00^{e}	100±0.00^{d}	100±0.00^{a}	100±0.00^{b}
6	44.59±17.44^{cd}	27.72±4.99^{ab}	100±0.00^{a}	100±0.00^{b}
12	20.59±1.75^{ab}	25.00±5.70^{ab}	100±0.00^{a}	72.69±21.21^{b}
18	18.34±2.36^{ab}	18.24±1.41^{ab}	100±0.00^{a}	29.11±10.52^{a}
24	13.34±4.72^{ab}	11.90±2.68^{a}	100±0.00^{a}	18.34±2.36^{a}
30	10.84±1.18^{a}	9.31±0.98^{a}	96.30±0.99^{a}	11.67±2.36^{a}

Note: Different superscript letters in the same column indicate significant differences ($p<0.05$). The experiment was repeated 2 times.

DISCUSSION

Emulsion stability is influenced by mechanical factors, temperature, formation process, changes in pH value and viscosity (Sayuti *et al*., 2016). The oil phase in water is oil that is distributed in the form of droplets throughout the outer part of the water.

Emulsion Viscosity.

Viscosity testing is carried out to determine the thickness of a preparation or mixture. The emulsion formed will become thicker as the viscosity level increases. The characteristic of a liquid known as viscosity is closely related to flow resistance; the level of liquid viscosity is related to the flow resistance (Hizmadin *et al*., 2024). The level of viscosity of an emulsion produced is also influenced by the amount and type of emulsifier and the size of the emulsifier particles (Usman *et al*., 2016). Changes in the thickness of the emulsion arise due to differences in the concentration of the emulsifier used.

Kombinasi The combination of emulsifier types used in this study were CMC and lecithin. CMC is a derivative form of natural polymers that is most often used in many fields, including the food, pharmaceutical, soap, fabric, and cosmetic sectors (Yuniarti *et al*., 2022). In contrast, the dual polarity of lecithin allows it to function as an emulsifier. The bipolar group of lecithin consists of a polar (hydrophilic) phosphate ester and a 2-chain fatty acid chain that is non-polar (hydrophobic), which allows it to interact well with the oil and water phases (Hamad *et al*., 2016).

In this study, CMC and lecithin were used because they have thick and stable properties so that the resulting emulsion can last for 30 days of storage. The combination of the use of CMC and lecithin emulsifiers produces oil dispersed in water so that the surface tension of the emulsion decreases.

During the storage period, the overall viscosity of the fish oil emulsion decreased. Research by Fransisca et al. (2023) found that the presence of short hydrocarbon chains of lecithin makes it more difficult to dissolve the dispersed phase in the dispersing phase, resulting in a decrease in the viscosity of the emulsion. Followed by a decrease in viscosity, the emulsion preparation becomes less stable. This is due to the fact that the preparation is increasingly dilute, which means that the dispersed phase, or globules, will move more easily through the dispersing medium. As a result, the possibility of collisions between globules increases and globules tend to associate to form larger particles.

Emulsion pH

The pH test shows the extent of acidity and alkalinity of a solution. pH stability is one of the most important factors in determining emulsion stability. Lipid oxidation can occur and will affect pH, where the pH value of the emulsion will decrease and have an impact on decreasing emulsion stability (Husni *et al*., 2019). The decrease in pH value in this study is thought to be due to the addition of benzoic acid as a preservative. The results showed that the pH for all formulas containing CMC and lecithin decreased over 30 days.

Menurut According to research by Lim *et al*. (2015), a decrease in pH value causes emulsion instability for 24 hours. The addition of CMC type emulsifier causes an increase in pH due to the presence of weak acid and strong base salt content so that it has basic properties (Hizmadin et al., 2024). Furthermore, according to Fitriyaningtyas & Widyaningsih (2015), the pH of an emulsion can increase if the pH of the emulsifier used is neutral.

Emulsion Stability Percentage

Emulsion stability is a condition in which the molecular size of the dispersing phase and the dispersed phase have the same configuration. The resistance of an emulsion to prevent separation or breakdown of the emulsion is called emulsion stability (Hizmadin et al., 2024). The creaming index is used to analyze emulsion stability. As stated by Husni et al. (2019) when oil globules combine and form large globules, this indicates that the emulsion is unstable. One factor that causes the failure of oil globule coating is the use of emulsifiers and the addition of inappropriate emulsifier concentrations. Another factor that is often an obstacle in making emulsion preparations is that the resulting emulsion is not thick. The globules cannot stay in position, which causes the separation to be greater. By using a 3% CMC emulsifier, the fish oil emulsion is more stable for longer than with a 1% CMC concentration. This shows that increasing the concentration of CMC causes the emulsion to become thick because water is bound and particles are mobilized, which has an impact on the stability of the emulsion. In addition, the use of lecithin reduces the surface tension of oil and water, which makes the emulsion more stable during storage (Fitriyaningtyas & Widyaningsih, 2015).

CONCLUSION

During 30 days of storage, the formula of each emulsion decreased, but the best emulsion formula from the combination of CMC emulsifier and lecithin was a ratio of 3: 1. This can be seen from the high viscosity, pH, and emulsion stability values during 30 days of storage.

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