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Improving the quality of Lemuru (*Sardinella lemuru*) oil using magnesol XL filter aid

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Abstract: This study was aimed at improving the quality of fish oil. A synthetic filter aid (Magnesol XL) was used at various concentration (1, 3 and 5%) and time levels (5, 10, 15 and 20 minutes) to adsorb the polar compound products of the oil. Some physical and chemical properties (viscosity, colour, density, acid value, peroxide value and free fatty acid) of the treated oil were determined. Results indicate that Magnesol XL at 1 and 3% levels significantly reduced the acid value, peroxide value and free fatty acid contents of the treated oil. Treatment of the fish oil with Magnesol XL at 1 and 3% levels was also better than treatment with 5% Magnesol XL on improving the fish oil quality. The fatty acid profile for Σ n3 at untreated and treatment adsorbent showed significant at 0.05 level but not significant at Magnesol XL adsorbent concentration 1-5%.

Keywords: lemuru (*Sardinella lemuru*) oil, Magnesol XL, polar compound and fatty acid

Introduction

Fish oil has been considered as an available source of long chain polyunsaturated n-3 fatty acids, especially Eicosapentaenoic acid (EPA) and Docosahexaenoic acid (DHA). These fatty acids are recognized to play an essential role in human health and nutrition (Newton, 1996). Although research on long chain n-3 polyunsaturated fatty acids continues to focus on a myriad of related conditions and diseases (Haumann, 1997), increasing research has also been conducted on extraction, concentration and stability of the long chain n-3 polyunsaturated fatty acid- rich fish oil.

Fish oil can be produced by several methods, including physical fractionation (Hirata et al., 1993), low temperature solvent fractionation (Moffat et al., 1993) and supercritical fluid extraction (Dunford et al., 1997). The conventional fish oil-refining steps include degumming, neutralizing, bleaching, and deodorizing. Free fatty acids (FFA) are precipitated as soaps and removed during the neutralization process (Young, 1978). Bleaching clays adsorb pigments from the oil, and oxidized components can be removed by deodorization. Although adsorbents are used to remove pigments during oil refining, they potentially adsorb FFA (Toro and Rocha, 1994).

Unrefined fish oil contains non-Triglycerides (TG), such as FFA and oxidized components that reduce the oil quality. The removal of impurities and non-TG components from crude fish oil is necessary to produce refined oil with a desirable and acceptable shelf life. The longer these components remain in the oil, the greater their negative effects on the quality of the refined oil. Farag et al. (2008) determined the minerals in Magnesol XL to be Si (34.75%), Mg (27.33%), Ca (4.36%), Na (1.92%) and Fe (1.09%) for improving the quality of fried soybean, sunflower, palm and cottonseed oils.

Therefore, the main objective of this study was to evaluate the feasibility of using magnesol XL as adsorbent to remove polar compounds and colour from fish oil to improve the quality of fish oil. The effects of the adsorbent on viscosity, colour, density, acid value, peroxide value, free fatty acid and profile fatty acid of the fish oil were also determined.

Material and Methods

Source of fish oil

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Lemuru (*Sardinella lemuru*) oil was a by-product from fish meal processing, and was obtained from a fish meal plant in East Java, Indonesia.

Filter aid

The filter aid used is Magnesol XL, a hydrous, white, amorphous and odourless powder that was obtained from Magnesol Product Division, Reagent Chemical and Research, Inc., Houston, Texas, 77 016, USA.

Adsorbent treatment

The lemuru oil (alkali refined) was centrifuged at 5000 rpm for 30 minutes, previously determined as the best treatment for removing soap stock and polar compounds (Suseno et al. 2009). Then it was mixed with 1, 3 and 5% (w/w) of Magnesol XL respectively for 5, 10, 15 and 20 minute at 25 Co. The oil was then separated from the adsorbent by centrifugation at 10,000 rpm for 30 minute. The physicochemical properties of the fish oil samples were analyzed before and after treatment with Magnesol XL.

Physico-chemical properties of the oils

Colour

A Lovibond tintometer was used to measure the colour of the oil samples under investigation. The yellow glass filter was fixed at 35 and the intensity of the red glass colour was measured (Nielsen, 1998).

Viscosity

The relative flow time was measured as an indication of oil viscosity using an Ostwald viscometer (Joslyn, 1950).

Density

The lemuru oil was measured using pyknometer or specific gravity bottle. A flask with a close-fitting ground glass stopper with a fine hole through it, so that a given volume can be accurately obtained. This enables the density of a fluid to be measured accurately (Nielsen, 1998).

Chemical properties

Acid value

A known weight of oil sample (~2 g) was dissolved in neutralized alcohol (50 mL) and titrated with KOH (0.1 mol L-1) (AOAC, 2000).

Peroxide value

A known weight of oil sample (~2 g) was dissolved in 30 mL chloroform: acetic acid (3:2, v/v) then 1 mL freshly prepared saturated KOH solution was added and the mixture vortexed for exactly 1 min. Distilled water (30 mL) and stock solution (0.5 mL, 1%) were added and the liberated iodine was titrated with sodium thiosulfate (0.1 mol L-1) (AOAC, 2000).

Free fatty acid

The percentage of FFA in each sample was determined by the titration method as described in AOCS method Ca 5a-40 (AOCS, 1998). Briefly, oil was weighed into a flask followed by neutralized 95% ethyl ethanol and a phenolphthalein indicator. The mixture was then titrated against sodium hydroxide solution (0.1 M) until a permanent pink colour persisted for at least 30 s. Weight percentage of FFA was calculated on either an oleic, palmitic, or lauric acid basis, depending on the type of oil being analyzed. Each sample was titrated in triplicate.

FAME analysis

Methyl esters were prepared by transmethylation using 2M KOH in methanol and n-heptane according to the method described by Ichihara et al. (1996) with minor modification. Lemuru oil (0.1 g) was dissolved in 2 ml hexane. After the addition of 4 ml of 2 M methanolic KOH, the tube was vortexed for 2 min at room temperature, and then centrifuged at 4000 rpm for 10 min. The hexane layer was then taken for analysis by gas chromatography (GC)The fatty acid composition was analysed by GC Shimadzu 2010 with autosampler (Shimadzu, Japan) equipped with a flame ionization detector and a fused silica capillary SGE column (30 m x 0.32 mm, ID x 0.25 lm, BP20 0.25 UM, USA). The oven temperature was 140oC, held 5 min, raised to 200oC at a rate of 4oC/min and to 220oC at a rate of 1oC/min, while the injector and the detector temperatures were set at 220oC and 280oC, respectively. The sample size was 2 µl and the carrier gas was controlled at 16 psi. The split used was 1:100. Fatty acids were identified by comparing the retention times of FAME with a standard 37 component FAME mixture (Supelco). Two replicate GC analyses were performed and the results were expressed in GC area % as a mean value and ± standard deviation.

Evaluation of the efficiency of adsorption process

The passive and depth filtration and also their combinations were evaluated for their performance in improving oil quality. This was determined by calculating percentage improvement of the treated oil over the untreated oil as Percentage Improvement (% PI)

$$\% \text{ PI} = \frac{(\text{Value of untreated oil} - \text{Value of treated oil})}{(\text{Value of untreated oil})} \times 100\%$$

The percentage improvement by the adsorbents

for both passive and depth filtration was calculated and reported as average percentage improvement (API).

Statistical analysis

The experiment was run in triplicate with a 2x3 factorial in a completely randomized design (CRD). The data were subjected to analysis of variance (ANOVA). The differences among the treatments were determined using Duncan's multiple range test (DMRT) (Steel and Torrie, 1980).

Results and Discussion

Peroxide value

The result of this study showed that 1% magnesol could remove FFA. Figure 1 shows that the peroxide value (PV) of untreated fish oils was 5.5 mg/Kg. All treatments could decrease PV value except at various 3% for 20 minute and 5% level. Treatment with magnesol XL at 1 and 3 % level significantly reduced the PV value, but the treatment at 5% level significantly increased the PV value. This may be due to the capabability of the Magnesol XL at 5% level to adsorb all polar compounds and antioxidants in the fish oil, leading to higher oxidative activities, hence higher PV values compared to the oil treated with the 1 and 3% Magnesol XL

According to Dimic et al. (1994) the bleaching process was considered particularly undesirable, as large amounts of bleaching earth promote a loss of tocopherols, further affecting the oil stability. Then, List et al. (1972) also reported that sunflower oils treated in the laboratory with a high concentration of bleaching earth (6%) had poorer flavour and oxidative stability than the same oil bleached with a lower concentration of earth (2%). Apparently bleaching removes, destroys or inactivates some unknown minor constituent essential for optimum keeping qualities. Alpaslan et al. (2001), the losses in total tocopherol content were approximately 3.5 and 7.9% during bleaching stages of the physical and chemical refining, respectively. The average losses of total tocopherol content during the chemical and physical refining processes were found to be 30.2% and 35.5%, respectively (Tasan and Demirci, 2005).

The adsorbent treatment with a combination of conditions of 1% concentration for 10 minutes, and 3 % Magnesol concentration for 5 minutes reduced PV by 60% and 59.5% respectively, giving the treated fish oil much lower peroxide values. Adsorptive bleaching is often used commercially to improve fish oils by removing coloured matter and natural pigments. In addition to the removal of coloured compounds, adsorptive bleaching improves the brightness of oils by removing the suspended mucilaginous and colloid-like matter (Gauglitz and Gruger, 1965)

Acid value

Figure 2 shows that the initial acid value (AV) of fish oil was 1.8 The treatment with Magnesol XL at 1, 3 and 5% levels reduced the Acid value. Treatment with 1% Magnesol XL resulted in a reduction of the acid value by 46%. The use of 3% Magnesol XL resulted in no further reduction of the acid values. In contrast, treatment at 5% level could have resulted in the adsorbsion of polar compounds as well as other minor compounds (tocopherol and pigment) that would promote the oxidation process and caused the increased AV in lemuru oil observed

Free fatty acid

The adsorbent Magnesol XL at 1% level reduced FFA by 68% (Figure 3). It was significant ($p<0.05$) with all treatments. Filtration with filter aid was recommended as it not only removes particles but also adsorbs soluble deterioration products, including FFA, polar materials and colour compounds. Generally, oils prepared at higher temperatures had higher FFA and there are two sources of free fatty acids: (1) the reaction of oil with water in the foods and (2) the secondary oxidation substances. This result indicated that the hydrolysis of ester bonds of triglyceride occurred less at lower temperatures (Nawar, 1996). The percentage improvement in the quality of oil by adsorbents was calculated and reported as average percentage improvement (API) for removal of FFA (Figure 3). Among these, treatment conditions B (1% Magnesol, 5 min) and C (1% Magnesol, 10 min) gave the best improvement of 71.2% for removal FFA. This was followed by treatment conditions E (68.6%), F (65.3), G (62.7%), L (61.0), D (61.0%), H (58.5%), K (57.6), J (54.2) and I (43.2), respectively. In general, 1 and 3 % Magnesol XL treatment exhibited higher Percentage Improvement than treatment at 5%.

Density and Viscosity

Table 1 shows that Magnesol XL treatment with variation in concentration levels of up to 5% did not significantly affect the density of fish oil. This is because the magnesol XL only adsorb the polar compounds but does not contribute to any weight or volume changes after its removal from the oil sample. The treatment viscosity values of fish oil were gradually and significantly increased toward the end of the treatment.

Colour

The colour characteristics of *lemuru* oil at different

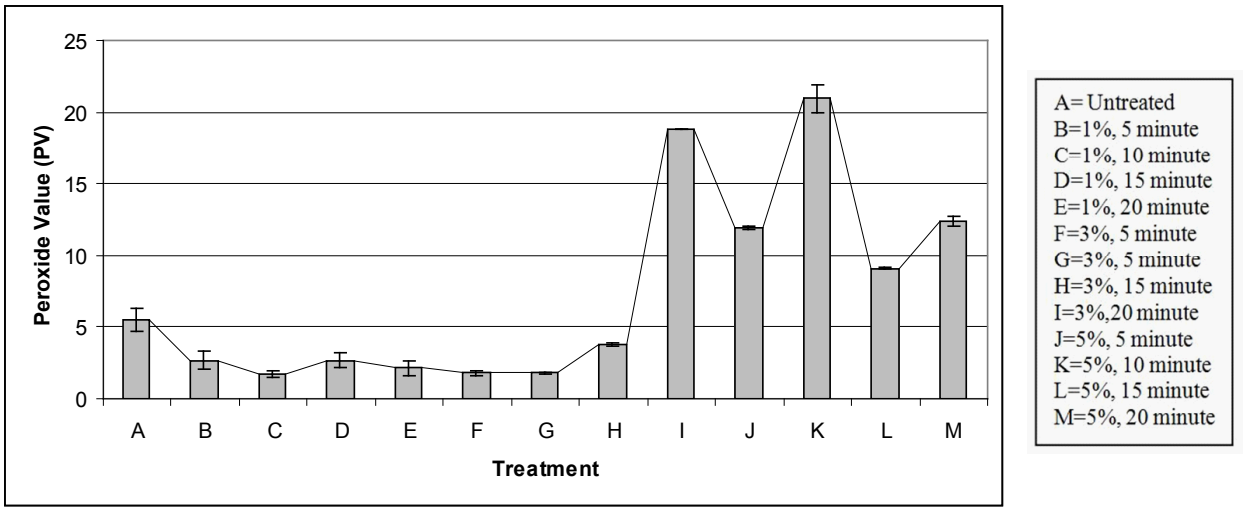

Figure 1. PV values of Lemuru oil with various levels of Magnesol XL treatment

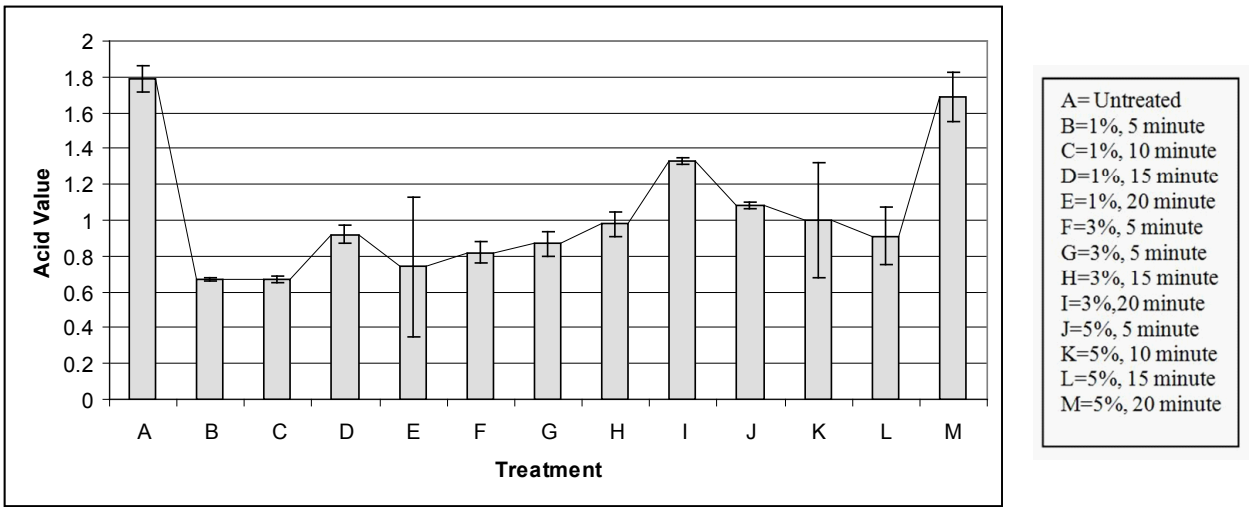

Figure 2. Acid Values of Lemuru oil with various levels of Magnesol XL treatment

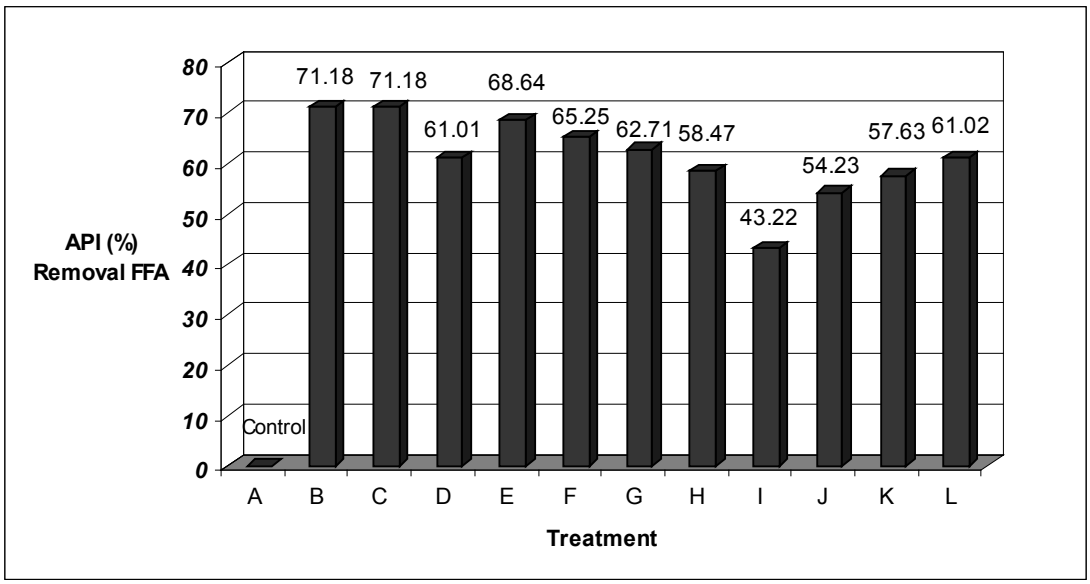

Figure 3. The percentage removal of FFA using adsorbent with various levels of Magnesol XL treatment

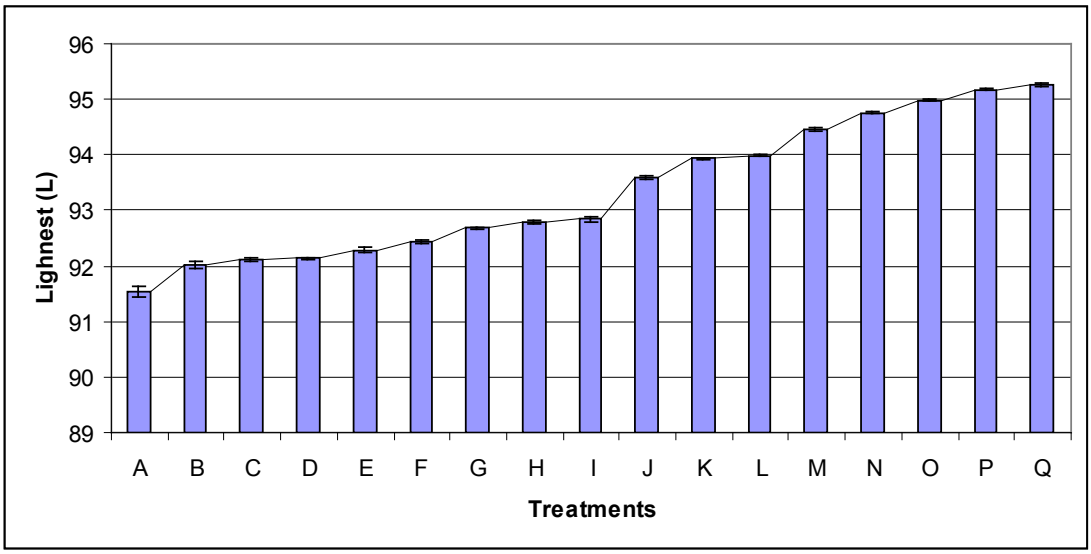

Figure 4. The lightness values (colour) of Lemuru oil with various levels of Magnesol XL treatment

Table 1. Density and viscosity values of Lemuru oil with various levels of Magnesol XL treatment

Treatment Code	Magnesol XL level	Time	Density	Viscosity
A	0	0	22.75 ± 0.01[a]	51.73 ± 0.15 [d]
B	1	5	22.91 ± 0.01[a]	44.23 ± 0 .11 [a]
C	1	10	22.83 ± 0.01[a]	64.13 ± 0.78 [l]
D	1	15	22.75 ± 0.00[a]	54.63 ± 0.06 [e]
E	1	20	22.73 ± 0.01[a]	59.50 ± 0.10 [f]
F	3	5	22.76 ± 0.00[a]	63.46 ± 0.15 [h]
G	3	10	22.73 ± 0.01[a]	60.20 ± 0.30 [g]
H	3	15	22.75 ± 0.00[a]	51.13 ± 0.23 [c]
I	3	20	22.71 ± 0.01[a]	66.63 ± 0.23 [j]
J	5	5	22.72 ± 0.00[a]	45.73 ± 0.06 [b]
K	5	10	22.74 ± 0.01[a]	70.60 ± 0 .10 [l]
L	5	15	22.69 ± 0.00[a]	69.17 ± 0.11[k]
M	5	20	22.73 ± 0.00[a]	82.30 ± 0.30[m]

Values given represent means of triplicate measurements.
Values in each row followed by the same letter are not significantly different at P = 0.05.

A= Untreated, B=1%, 5 minute, C=1%, 10 minute, D=1%, 15 minute, E=1%, 20 minute
F=3%, 5 minute, G=3%, 5 minute, H=3%, 15 minute, I=3%,20 minute, J=5%, 5 minute
K=5%, 10 minute, L=5%, 15 minute, M=5%, 20 minute

Table 2. The colour of Lemuru oil with various levels of Magnesol XL treatment

Concentration	Time (Minute)	a*	b*	C*	h	(ΔE)
Untreated		-1.23±0.04^{n}	48.99±0.04^{r}	49±0.05^{q}	91.44±0.04^{a}	0.18±0.07^{d}
0.5	5	-1.45±0.04^{m}	47.72±0.03^{q}	47.69±0.08^{p}	91.74±0.04^{b}	0.1±0.04^{a,b,c}
0.5	10	1.49±0.03^{l,m}	47.53±0.04^{p}	47.55±0.03^{o}	91.79±0.03^{b,c}	0.05±0.03^{a,b}
0.5	15	-1.5±0.03^{l}	47.48±0.02^{o}	47.5±0.01^{o}	91.81±0.03^{c}	0.04±0.01^{a,b}
0.5	20	-1.57±0.02^{k}	47.04±0^{n}	47.07±0.01^{n}	91.91±0.03^{d}	0.05±0.03^{a,b}
1	5	-1.67±0.01^{j}	46.43±0.03^{m}	46.46±0.03^{m}	92.06±0.01^{e}	0.04±0.01^{a}
1	10	-1.81±0.02^{i}	45.36±0.02^{l}	45.39±0.02^{l}	92.28±0.02^{f}	0.13±0.02^{c,d}
1	15	-1.85±0.05^{h,i}	45.29±0.04^{k}	45.33±0.04^{k}	92.34±0.05^{f,g}	0.06±0.05^{a,b}
1	20	-1.88±0.02^{h}	44.79±0.03^{j}	44.83±0.03^{j}	92.39±0.03^{g}	0.08±0.04^{a,b,c}
3	5	-2.36±0.01^{g}	42.85±0.02^{i}	42.91±0.02^{i}	93.15±0.02^{h}	0.08±0.03^{a,b,c}
3	10	-2.6±0.01^{f}	41.25±0.04^{h}	41.33±0.04^{h}	93.61±0.02^{i}	0.05±0.03^{a,b}
3	15	-2.62±0.01^{f}	40.93±0.03^{g}	41.02±0.03^{g}	93.67±0.01^{i}	0.04±0.03^{a,b}
3	20	-2.74±0.02^{e}	39.09±0.02^{f}	39.19±0.02^{f}	94.01±0.04^{j}	0.06±0.03^{a,b}
5	5	-2.91±0.02^{d}	38.22±0.01^{e}	38.33±0.01^{e}	94.35±0.03^{k}	0.03±0.01^{a}
5	10	-3±0.02^{c}	37.57±0^{d}	37.69±0.01^{d}	94.57±0.03^{l}	0.04±0.01^{a,b}
5	15	-2.99±0.03^{c}	36.43±0.03^{c}	36.55±0.03^{c}	94.68±0.05^{m}	0.05±0.04^{a,b}
5	20	-3.15±0.02^{b}	35.97±0.01^{b}	36.11±0.01^{b}	95±0.03^{n}	0.03±0.01^{a}
Refined cod liver oil		-3.83±0.01^{a}	17.36±0.01^{a}	17.78±0.01^{a}	102.44±0.05^{o}	0.06±0^{a,b}

+a* values = redness , -a* values = greenness; +b* values = yellow and -b* values = blue, h =Hue angle and C*= chroma. (ΔE) = total colour difference values

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Values given represent means of triplicate measurements.

Values in each row followed by the same letter are not significantly different at P = 0.05

Table 3. Chemical characterization of lemuru oil before and after treatment at various levels of Magnesol XL

Fatty acid Composition (% methyl ester)	Untreated	Magnesol 1%	Magnesol 3%	Magnesol 5%
C 10:0	0.26	0.37	0.31	1.55
C 11:0	0.13	0.07	0.11	1.08
C 12:0	0.33	0.34	0.28	0.74
C 13:0	0.19	0.14	0.15	0.29
C 14:0	10.04	7.32	7.86	7.38
C 15:0	0.73	0.58	0.57	0.58
C 16:0	20.25	22.86	22.5	20.96
C 17:0	1.25	0.94	0.99	0.93
C 18:0	5.48	16.62	15.38	11.67
C 20:0	1.65	0.2	1.54	1.52
C 21:0	0.24	1.64	0.24	0.26
C 22:0	0.38	0.32	0.35	0.48
C 23:0	0.44	0.25	0.19	0.26
C 24:0	0.45	0.51	0.62	0.0
Σ SFA	**41.43**	**51.72**	**50.67**	**45.07**
C 14 :1	0.03	0.02	0.02	0.03
C 15 :1	0.11	0.11	0.09	0.14
C 16: 1	12.02	10.26	8.26	9.32
C 17: 1	0.78	0.49	0.67	0.69
C 18: 1 n9	2.13	2.61	2.4	2.27
Σ MUFA	**15.07**	**13.49**	**11.44**	**12.45**
C 18: 2 n6 cis	0.7	0.74	0.69	0.68
C 18: 3 n3	2.5	2.67	2.44	1.32
C 20: 2	1.89	1.64	1.65	1.74
C 20: 3 n3	15.36	14.13	14.3	10.23
C 20: 4 n6	0.32	0.62	0.56	0.6
C 20: 5 n3	7.31	2.43	3.73	3.95
C 22: 2	0.43	1.64	0.48	0.25
C 22: 6 n3	3.92	6.35	5.43	10.14
Σ PUFA	**32.43**	**30.22**	**29.28**	**28.91**
PUFA/SFA	**0.78**	**0.58**	**0.58**	**0.64**
Σ n6	**1.02**	**1.36**	**1.25**	**1.28**
Σ n3	**29.09**	**25.58**	**25.9**	**25.64**
n6/n3	**0.04**	**0.05**	**0.05**	**0.05**
unidentified	**11.07**	**4.57**	**8.61**	**13.57**

processing steps compared with those of the refined cod liver oil are presented in Table 2. The refined cod liver oil and bleached *lemuru* oil were lighter (higher L*) than untreated oil. Bleaching increased the colour lightness of *lemuru* oil. All treatments had a negative a* value, indicating a slight greenish colour, and a positive b* (yellowish) value. The lowest b* value was observed for the bleached lemuru oil. The total colour difference (ΔE) values for all treatments were greater than 1.0. Hue angle values of all treatments were higher than 90°. Oils with a hue angle value between 90 and 180° were more greenish-yellow in colour. Commercially refined cod liver oil had the highest hue angle (102.44°). Its colour was observed to be a very light greenish-blue.

Profile of fatty acid from lemuru oil

Lemuru fish oil with untreated, 1%, 3% and 5% magnesol XL had high amounts of palmatic acid (C 16: 0), stearic acid (C18 : 0), palmitoleic acid (C 16: 1) and eicosatrienoic acid C 20: 3 n3 (>10%). Among the saturated fatty acids palmitic acid (C16:0) was a dominant fatty acid. Palmitoleic acid (C16:1) and eicosatrienoic acid (C 20: 3 n3) was a major monounsaturated and polyunsaturated fatty acid, respectively. Table 4 showed that fat content of a range of untreated, magnesol 1%, magnesol 3% and magnesol 5% lemuru fish oil consist of Σ n3 and Σ n6 were 29.09, 25.58, 25.9, 25.64, 1.02, 1.36, 1.25, and 1.28 respectively. The saturated fatty acid (SFA) composition is significant at the 0.05 level for all treatments, also mono unsaturated fatty acid (MUFA). The omega 3 is significant at the 0.05 level for untreated with all treatments but not significant at 1 to 5 magnesol XL concentrations. The result of treatment show that Σ n3 of treatment (1%, 3% and 5% magnesol) decreased be compared with untreated. According to Taylor and Gallavan (1988) treated frying oil with a mixture of gel-derived alumina or magnesium silicate and noted a significant reduction in the amount of fatty acids, aldehydes, ketones and odour- and colour-forming compounds. Lin et al. (1999) used combinations of four commonly used filter aids to recover the used frying oils: Britesorb, Hubersorb, Frypowder and Magnesol reduced the free fatty acids by 82.6–87.6% and the absorbance at 460nm by 5.6–8.6%. The fatty acid profile generally exhibits a dominance of the two classes, SFAs and PUFAs (Table 3). The ratio of PUFA/SFA on untreated, 1%magnesol, 3% magnesol and 5% magnesol were 0.78, 0.58, 0.58 and 0.64 respectively. The proportions of PUFAs-n3 and PUFAs-n6 on untreated, 1% magnesol, 3% magnesol, 5% magnesol were 0.04, 0.05, 0.05 and 0.05, respectively. The UK Department of Health recommends an ideal ratio of n6/n3 of 4.0 at maximum (HMSO, 1994). Values higher than the maximum value are harmful to health and may promote cardiovascular diseases (Moreira et al., 2001).

Conclusion

Overall, the data shown that the magnesol XL under study possess useful scavenging properties, i.e. they remove the oxidation product from fish oil and regenerated oil may improve the quality fish oil in the food industry. Generally the present work provides information on improving the quality of fish oil. The data indicate that treatment with 1% and 3% levels had nearly the same effect significant and both levels were better than treatment with the 5% level on improving the fish oil quality. The data of chemical analysis indicate that 1 and 3% level decrease PV, AV and FFA values. But, the physical analysis for density resulted no differences at all treatments. While for viscosity increased at 1, 3 and 5% level for 5, 10, 15 and 20 minute. Then, L (lightness) value was increased at all levels and time. Significant ($p<0.5$) for Σ n3 on untreated and treatment was not significant for each concentration adsorbent.

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