



# Astaxanthin and omega-3-rich oil from fermented *Acetes* (Cincalok) and its application as bioactive additive and sunscreen in lotion <sup>☆</sup>

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Received 21 October 2021 – Accepted 3 April 2022

**Abstract** – Shrimp species have been reported to contain astaxanthin, which has high antioxidant activities. They also contain omega-3 in the form of docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA), which can act as photoprotective agents that maintain healthy skin from reactive oxygen species (ROS) due to exposure to UV rays. In addition, fermentation has become an essential pre-treatment to extract the bioactive components in shrimp more easily. This study aims to extract oil from cincalok, a traditional Indonesian (especially in West Kalimantan) food made from *Acetes* shrimp fermented for 7–15 days. Cincalok oil was added to the lotion as a bioactive additive and sunscreen. Cincalok oil was extracted by the soxhletation method using n-hexane as solvent. The oil was then analyzed for its physicochemical properties, including density, viscosity, possible heavy metal contamination, and the profile of the fatty acids contained. The yield of cincalok oil extraction was  $1.09 \pm 0.05\%$ , with the highest fatty acid content of 21.70% palmitic acid, 10.99% DHA, and 10.33% EPA. Cincalok oil also contains astaxanthin of  $0.38 \pm 0.02$  mg/L oil. It has a viscosity of  $69.71 \pm 0.12$  cP with a density of  $0.93 \pm 0.03$  g/cm<sup>3</sup>. The analysis data of ICP-AES shows that there is no heavy metal contamination. The SPF value produced from cincalok oil lotion at 5 and 10% variations of cincalok oil was  $15.17 \pm 0.09$  and  $30.28 \pm 0.49$ , respectively. The SPF value of lotion with the addition of cincalok oil was much greater than that of the base lotion, which was  $2.16 \pm 0.12$ .

**Keywords:** *Acetes* / astaxanthin / fermentation / omega-3 / sunscreen

**Résumé** – Astaxanthine et huile riche en oméga-3 provenant d'*Acetes* fermentés (Cincalok) et son application comme additif bioactif et écran solaire dans une lotion. Les espèces de crevettes ont été signalées comme contenant de l'astaxanthine qui possède des activités antioxydantes élevées. Elles contiennent également des oméga-3 sous forme d'acide docosahexaénoïque (DHA) et d'acide eicosapentaénoïque (EPA), qui peuvent agir comme des agents photoprotecteurs contre les espèces réactives de l'oxygène (ROS) liées à l'exposition aux rayons UV. En outre, la fermentation est devenue un prétraitement essentiel pour extraire plus facilement les composants bioactifs des crevettes. Cette étude vise à extraire l'huile du cincalok, un aliment traditionnel indonésien (surtout dans le Kalimantan occidental, île de Bornéo) fabriqué à partir de crevettes *Acetes* fermentées pendant 7 à 15 jours. L'huile de cincalok a été ajoutée à la lotion en tant qu'additif bioactif et écran solaire. L'huile de Cincalok a été extraite par la méthode Soxhlet en utilisant le n-hexane comme solvant. L'huile a ensuite été analysée pour ses propriétés physico-chimiques, notamment sa densité, sa viscosité, son éventuelle contamination par des métaux lourds et le profil des acides gras qu'elle contient. Le rendement d'extraction de l'huile de cincalok était de  $1,09 \pm 0,05 \%$ .

<sup>☆</sup> Contribution to the Topical Issue "Lipids from aquatic environments / Lipides issus des milieux aquatiques".

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En termes d'acides gras, elle contient 21,70 % d'acide palmitique, 10,99 % de DHA et 10,33 % d'EPA. L'huile de Cincalok contient également de l'astaxanthine à raison de  $0,38 \pm 0,02$  mg/L d'huile. Sa viscosité est de  $69,71 \pm 0,12$  cP avec une densité de  $0,93 \pm 0,03$  g/cm<sup>3</sup>. Les données d'analyse de l'ICP-AES montrent qu'il n'y a pas de contamination par les métaux lourds. La valeur SPF produite à partir de la lotion à l'huile de cincalok à des taux d'incorporation de 5 et 10 % d'huile de cincalok était respectivement de  $15,17 \pm 0,09$  et  $30,28 \pm 0,49$ . La valeur SPF de la lotion avec l'ajout d'huile de cincalok était donc largement supérieure à celle de la lotion de base, qui était de  $2,16 \pm 0,12$ .

**Mots clés** : Acétates / astaxanthine / fermentation / oméga-3 / écran solaire

## Highlights

- *Acetes* are a marginalized marine commodity.
- Oil extracted from their fermented products (cincalok) contains astaxanthin and omega-3, which are very beneficial for skin health.
- This oil can be used as a bioactive additive and sunscreen in lotion formulations.
- The addition of 10% cincalok oil in the lotion formulation resulted in SPF values of 30.28.

## 1 Introduction

Indonesia is a tropical country that gets sunshine all year round. Chronic exposure to ultraviolet (UV) rays from the sun can cause changes in the structure and composition of the skin and oxidative stress on the skin. UV is the most important modifiable risk factor for skin cancer and many others environmentally-influenced skin disorders (D'orazio *et al.*, 2013). Long-term exposure to ultraviolet light can cause premature skin aging (photoaging) (Huang and Chien, 2020; Geng *et al.*, 2021). Skincare is needed to overcome and prevent the side effects caused by UV rays. Therefore, additional skin protection is needed in skin-protective cosmetic preparations, namely sunscreen cosmetics in the form of lotion. Lotion should be formulated in such a way that it contains rich bioactive components and sunscreen (Korać and Khambholja, 2011).

As an archipelagic tropical country, Indonesia is also rich in marine natural resources. One of them is shrimp. Non-paneid shrimp species such as *Acetes* (*Acetes japonicus* or *Acetes sibogaesibogae*) are the shrimp group with the highest abundance on West Kalimantan, Indonesia. Despite its abundance, *Acetes* is not an export commodity, and the price is meager (Riswanto and Hediando, 2016), even though their nutritional content is relatively high. Balange *et al.* (2017) investigated that *Acetes* obtained from the Indian sea contain 9,12-octadecadienoic acid (17.08%), docosahexaenoic acid (DHA) (15.69%), and eicosapentaenoic acid (EPA) (13.45%) as the primary fatty acids. These compounds are well known for their health benefits, especially for treating several chronic diseases. Omega-3 fatty acids are polyunsaturated fatty acids that are good for maintaining skin moisture to prevent inflammation and dryness of the skin (Huang *et al.*, 2018).

Other active compounds contained in *Acetes* are chitosan, minerals, lipids, and carotenoids. The highest carotenoid in *Acetes* is astaxanthin (Fig. 1), reported as 14–40 µg/g dry weight depending on the species and size (Ung *et al.*, 2020; Lv *et al.*, 2021). Astaxanthin is one of the carotenoids that can

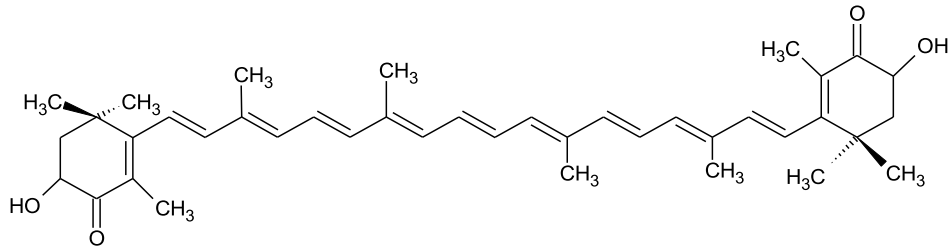
provide antioxidant effects to protect the body from free radical attacks (Ekpe *et al.*, 2018; Brotsudarmo *et al.*, 2020) and has a preventive effect on photoaging, inhibiting the formation of wrinkles, and increases skin elasticity (Komatsu *et al.*, 2017; Eren *et al.*, 2018). The use of active substances that are antioxidants can prevent various diseases caused by UV radiation, protecting against UV rays to act as a sunscreen (Ebrahimzadeh *et al.*, 2014).

*Acetes* are rarely consumed in fresh form but are first processed into dried shrimp, crackers, or paste (Balange *et al.*, 2017; Ung *et al.*, 2020). Another processed *Acetes* into a typical food of West Kalimantan is cincalok. Cincalok is made through the fermentation of *Acetes* by adding sugar and salt in a particular ratio and stored for 7–15 days (Khairina *et al.*, 2016). Fermentation has been a traditional and well-known preservation technology for a long time. Even though a simple technology produces it, fermented products are popular because they have a distinctive aroma and taste, high nutrients, and high functional properties (Sanlier *et al.*, 2017). Fermentation of *Acetes* is completed when the shrimp paste becomes bright pink and develops a strong, sour, and savory flavor. Low-temperature fermentation in the production process of cincalok is vital because it can separate omega-3 and astaxanthin without destroying them, thus facilitating the extraction process. Fermentation of cincalok is also reported to increase the astaxanthin content characterized by increasing pink intensity during incubation (Khairina *et al.*, 2016).

Another study reported that lactic acid bacteria (LAB) in the fermentation process proved to be effective in using a biological extraction approach for astaxanthin and omega-3. Cincalok contains LAB, which produces proteases that play a role in hydrolyzing shrimp shell proteins and releasing free astaxanthin from protein-chitin complexes, making it easier to extract. Lactate fermentation also dissolves vesicles from the shrimp exoskeleton, consisting of calcium salts and large amounts of astaxanthin (Prameela *et al.*, 2017; El-Bialy and El-Khalek, 2020). According to the nonpolar nature of astaxanthin and omega-3, they are present in the lipid fraction of organisms. Therefore, this study aimed to extract the lipid fraction from cincalok and characterize its physicochemical properties. The sun protection factor (SPF) values of the lotions formulated with cincalok oil were also determined.

## 2 Materials and method

*Acetes* shrimp obtained from Mempawah Waters, West-Kalimantan, Indonesia. Sugar and salt were bought from the market. Base lotion without sunscreen prepared by Dasilia *et al.* (2021). Astaxanthin ((3S,3'S)-3,3'-dihydroxy-β,



**Fig. 1.** Chemical structure of astaxanthin (3,3'-dihydroxy- $\beta,\beta$ -carotene-4,4'-dione).

$\beta$ -carotene-4,4'-dione, 3,3'-dihydroxy- $\beta,\beta$ -carotene-4,4'-dione, *trans*-Astaxanthin) analytical grade with purity > 98% was purchased from Sigma Aldrich (SML0982 SIGMA). Standard was stored in the dark at 4 °C until use. Butylated hydroxytoluene (BHT) antioxidant and solvent such as acetone, hexane, methanol, and H<sub>2</sub>O uHPLC grade were also purchased from Sigma Aldrich.

## 2.1 Preparation of Cincalok

*Acetes* were cleaned with seawater and separated from small fish contaminants and sand. The clean *Acetes* were added with sugar and salt with the mass ratio of shrimp:sugar:salt was 10:1:1 w/w, then was put in a tightly closed plastic container. The fermentation process was carried out for 9 days. Cincalok produced was filtered, and the residue was dried at 50 °C for 3 h using vacuum drying. Dry cincalok was ground using a screw press and ready for further treatment.

## 2.2 Extraction of oil from fermented Acetes

A 150 g of dried cincalok was placed onto a thimble, and the thimble was put into the soxhlet extractor. N-hexane solvent was poured into a two-neck-round bottom flask joined with the extractor and flask along with the condenser on the top to avoid any solvent losses. The whole assembly was then placed on the temperature controller heater to provide the required temperature. The temperature was measured by a thermometer that was inserted in one of the necks of the round bottom flask. After a specific interval, the experiment was stopped, and the trapped oil in the solvent was separated. The mixture of solvent and oil was separated using a distillation at a temperature of 65 °C continued by drying with N<sub>2</sub>. The resulting oil, in the following text, is called cincalok oil.

The oil obtained after solvent separation was weighed, and the oil yield was calculated using equation (1):

$$\text{Oil yield (wt \%)} = \frac{\text{Mass of extracted oil}}{\text{Mass of dried cincalok}} \times 100 \% \quad (1)$$

Testing physical properties of the oil include measurement of oil density and viscosity. The viscosity and density values were determined at 25 °C using an Ostwald viscometer and a pycnometer, respectively. Carotenoid astaxanthin content in oil was determined using UV-Visible Spectrophotometer and ultra-High Performance Liquid Chromatography (uHPLC).

Possible heavy metal contamination was determined using Inductively Coupled Plasma-Atomic Emission Spectrometry (ICP-AES) following the AOAC (2016) method in SUCOFINDO Laboratory, Pontianak, West-Kalimantan, Indonesia. Fatty acids in cincalok oil were analysed using Gas Chromatography-Mass Spectrometry (GC-MS) in Integrated Research and Testing Laboratory, Gadjah Mada University, Yogyakarta, Indonesia.

## 2.3 Determination of total carotenoid content

The total carotenoid concentration (presented as astaxanthin) in cincalok oil was measured spectrophotometrically using modified form the procedure by [Goula \*et al.\* \(2017\)](#) and [Corbu \*et al.\* \(2019\)](#). For the first experimental step, astaxanthin analytical standards were dissolved in acetone to prepare standard solutions at 0.5–7.0 mg/L concentrations. The spectrophotometric analyses were carried out using Spectrophotometer, Shimadzu, UV-1800 series, measuring the absorbance in 300–700 nm. Calculation of the total carotenoid concentration was done according to the standard curve of astaxanthin. Reading of astaxanthin concentrations was carried out at the maximum absorbance of standard solutions. Analyses of the samples were done on cincalok oil in triplicate. Cincalok oil analyzed did not saponified or neutralized before.

As a comparative study, chromatographic analysis of astaxanthin was performed using a C3000-DIONEX uHPLC coupled with a diode array detector (DAD). Astaxanthin was detected in 433 nm and separated in uHPLC using a Sunfire column (Agilent, 50 mm length, internal diameter 2.1 mm, 1.8  $\mu$ m particle size, part number 9579). The mobile phase consisted of acetone (A), deionized water (B), and methanol (C) at a flow rate of 0.6 mL/min. A 20  $\mu$ L sample was injected into the column, and the temperature was maintained at 40 °C. All peaks were scanned at 274–660 nm and identified by comparison with the retention times of the standard and followed by quantification based on the standard curve. Astaxanthin analytical standards were dissolved in methanol and acetone 1:1 v/v with 0.1% BHT as an antioxidant. The samples were filtered using a 0.45  $\mu$ m membrane before injection.

## 2.4 Lotion formulation with cincalok oil and determination of their SPF value

The lotion formulation was made in 2 variations of the composition of cincalok oil, namely 5 and 10% by mass of 10 g of base lotion, which was stirred until homogeneous. Each

**Table 1.** Normalization function used in SPF calculation.

Wavelength (nm)	EE X I (normalization)
290	0.0150
295	0.0817
300	0.2874
305	0.3278
310	0.1864
315	0.0837
320	0.0180

formulation was made triplicate. The determination of the SPF value of lotion preparations was carried out by modifying the method of [Mbang et al. \(2014\)](#). Each lotion was weighed as much as 0.5 g and dissolved with ethyl acetate solvent using a 5 mL volumetric flask (done in triple). The solution was vortexed for 2 minutes and sonicated for 10 minutes. The solution was measured at a wavelength of 290–320 nm using Spectrophotometer, Shimadzu, UV-1800 series, and the SPF value was measured using the Mansur equation, which can be seen in [equation \(2\)](#) ([Tab. 1](#)):

$$SPF = CFx \sum_{290}^{320} EE(\lambda)xI(\lambda)x Abs(\lambda) \quad (2)$$

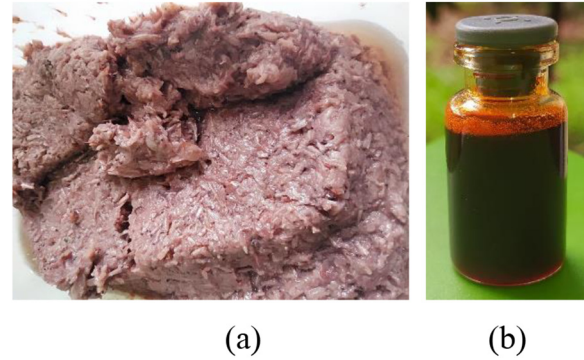
### 3 Results and discussion

#### 3.1 Physical characteristic of cinalok oil

Cinalok from *Acetes* fermentation for nine days in this study can be seen in [Figure 2a](#). This fermentation process aims to increase the intracellular productivity of shrimp ([Chen et al., 2017](#)). Before extraction, the drying stage of cinalok was carried out to minimize the water content because non-polar solvents were used in oil extraction. The water contained in the sample can hinder the diffusion of bioactive compounds out of the sample during extraction ([Silva et al., 2018](#)). Cinalok dried results yield 38.99% from 838.7 g of wet weight.

The treatment using a screw press in the next stage aims to crush shrimp shell and increase the surface area of the sample so that there is more possibility of sample-solvent contact. The screw press method has not been able to extract cinalok oil optimally because the oil content is very small. Therefore, the extraction of cinalok oil was carried out using the soxhletation method. The oil yield obtained was  $1.09 \pm 0.05\%$  of the dry sample weight of 150 g. The shrimp oil yield generally ranges from 1 to 8%, depending on the type of shrimp.

Cinalok oil has a viscosity of  $69.71 \pm 0.12$  cP with a density of  $0.93 \pm 0.03$  g/cm<sup>3</sup>. The viscosity value of cinalok oil is not much different from the viscosity of fish oil, which also contains omega-3. The viscosity of sardine oil obtained from the fishmeal was reported as 51.7 cP at 25 °C ([Suseno et al., 2015](#)) with a density of 0.92 g/cm<sup>3</sup> ([Mata et al., 2014](#)). [Ahmed et al. \(2017\)](#) have reported that the skins, scale, and bones oil of big eye tuna's viscosities range from 48–54 cP at 25 °C. The oil's viscosity can be affected by impurities in the oil, the oil's density, melting point, degree of unsaturation, and temperature ([Suseno et al., 2015](#); [Ahmed et al., 2017](#)).

**Fig. 2.** Cinalok (a) and cinalok oil (b).**Table 2.** Analysis data of ICP-AES.

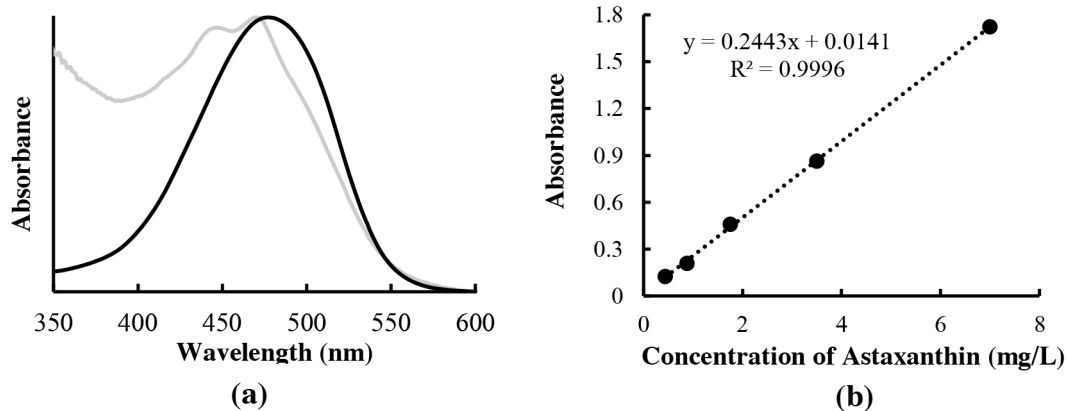
Characteristics	Result (ppm)
Lead as Pb	< 0.0010
Cadmium as Cd	0.0021
Zinc as Zn	0.0877
Iron as Fe	0.0408
Copper as Cu	< 0.0010
Arsenic as As	< 0.0010
Mercury as Hg	< 0.0002

#### 3.2 Metal content in cinalok oil

Based on ICP-AES analysis data ([Tab. 2](#)), there is no contamination of heavy metals in the oil except for cadmium (Cd) metal (0.0021 ppm). However, this value is far below the contamination threshold required for raw materials for pharmaceutical preparations, which is < 0.3 ppm. Zinc (Zn) and iron (Fe) are present in a concentration of 0.0877 and 0.0408 ppm, respectively. Zinc has been reported to have antioxidant properties and has been found to prevent UV-induced skin damage and reduce the incidence of malignancies ([Gupta et al., 2014](#)). In another study, Fe can play a crucial role in oxidative stress and photo-induced skin damage caused by reactive oxygen species (ROS) generated in the skin by UVA (320–400 nm). Iron is not actively excreted from the body; however, the skin is a critical organ in iron hemostasis as iron is lost through the skin by desquamation ([Wright et al., 2014](#)).

#### 3.3 Carotenoid content in cinalok oil

As well as other shrimp oil, cinalok oil contains the carotenoid compound astaxanthin. This is indicated by the deep red color of the cinalok oil (see [Fig. 2b](#)). In general, the highest carotenoid content is found in the cephalothorax (the united head and thorax) in shrimp ([Scurria et al., 2020](#)). Analysis using a UV-Vis spectrophotometer was carried out to determine the total carotenoid as astaxanthin content in cinalok oil. The spectrum of cinalok oil compared with standard astaxanthin in acetone solvent is presented in [Figure 3a](#).



**Fig. 3.** Astaxanthin standard (black) and cincalok oil (grey) spectrum in acetone (a) and calibration curve of astaxanthin standard at 477nm (b).

Based on Figure 3a, astaxanthin analytical standard and cincalok oil dissolved in acetone show the absorption spectrum in the range 350–600 nm. Conjugated double bonds cause the absorption spectra at these wavelengths in carotenoid structures that can absorb visible light (Rahmalia *et al.*, 2014; Macernis *et al.*, 2014). However, in the spectrum of cincalok oil, two absorption peaks were observed at 470 and 447 nm, whereas in the astaxanthin standard spectrum, there is only one peak at a wavelength of 477 nm in accordance with Rodriguez-Amaya (2002). The peaks difference between cincalok oil and astaxanthin standard is due to the presence of other pigments contained in cincalok oil. As reported by Gulzar *et al.* (2020), in addition to containing astaxanthin as the largest carotenoid component, shrimp also contain  $\beta$ -carotene, lutein, and zeaxanthin. The maximum wavelength for other carotenoid components in shrimp species such  $\beta$ -carotene, lutein, and zeaxanthin in acetone has been reported by some literature to be around 445–450 nm (Rodriguez-Amaya, 2002; Casella *et al.*, 2020). The presence of the other components also causes the maximum wavelength of cincalok oil spectra (470 nm) to shift slightly to hypochromic compared to the maximum wavelength of standard astaxanthin spectra (477 nm).

For quantification analysis based on spectrophotometry, the maximum wavelength absorbance is usually used as the working wavelength. This helps achieve higher sensitivity and higher accuracy by reaching the maximum absorbance ratio between the target and interference materials. Hence, in this work, calculating the total carotenoid concentration in cincalok oil was carried out at wavelengths of 477 nm. The calibration curve of standard astaxanthin at a wavelength of 477 nm is presented in Figure 3b. The linear regression value ( $R = 0.9996$ ) as can be seen in Figure 3b show that a linear relationship exists between absorbance and astaxanthin concentration according to Lambert–Beer's Law.

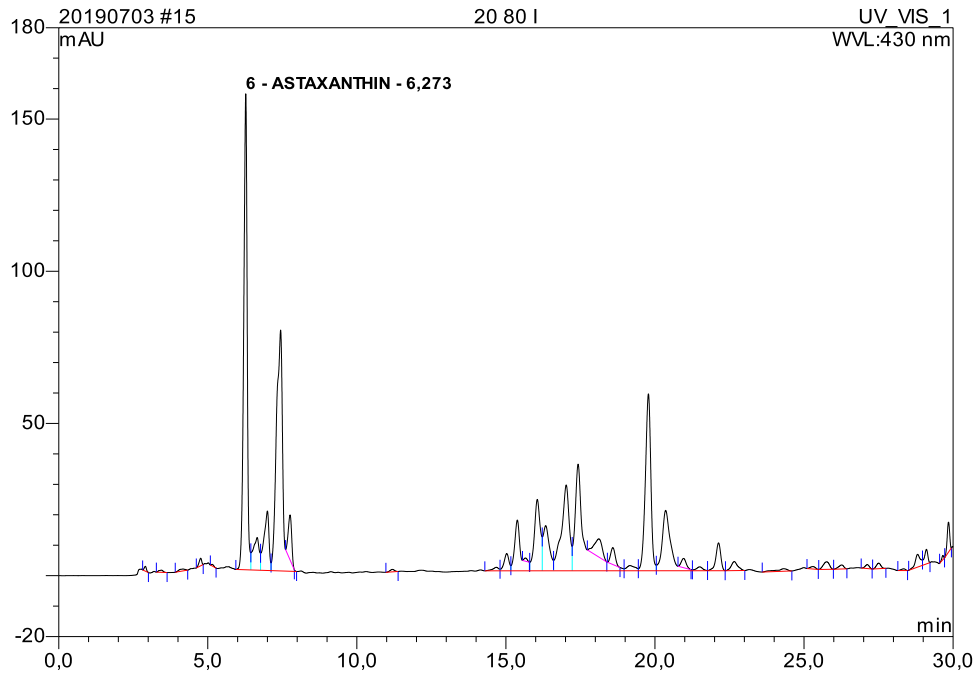
Following the linear equation  $y = 0.2443 \times \pm 0.0141$ , that the total concentration of carotenoids in cincalok oil is  $0.556 \pm 0.033$  mg/L or  $0.598 \pm 0.031$  mg/g of oil. Total astaxanthin acquired by the uHPLC-DAD analysis was only  $0.375 \pm 0.008$  mg/L, but it is generally accepted that some carotenoids with their isomers present in low concentration might not be detected as peaks and are not include in the uHPLC method. In contrast, in the spectrophotometric analysis, the absorbance

is highly increased by other than carotenoids compounds dissolved in lipids also active in the carotenoid spectral range. In addition, the standard astaxanthin used in this study was *trans*-astaxanthin from *Blakesleatrispora*. In contrast, in shrimp oil, astaxanthin is also present in other isomers and possibly still in the form of esters so that it is detected at different peaks from the standard. Shrimp oil contains appreciable content of astaxanthin at 0.1 to 1.5 mg/mL depending on processing methods. Other carotenoids such as zeaxanthin, lutein, and  $\beta$ -carotene are present as minor carotenoids (Carvalho and Caramujo, 2017). Chromatograms of cincalok oil compared to standard astaxanthin are presented in Figure 4, where the retention time of standard *trans*-astaxanthin is 6.273 minutes.

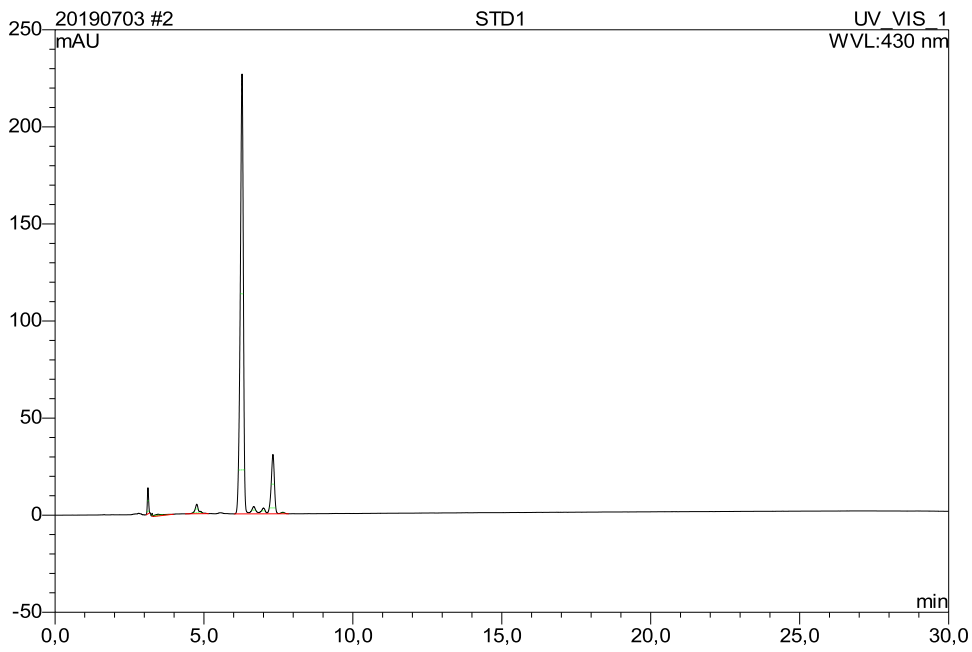
### 3.4 Fatty acid profile of cincalok oil

Cincalok oil was characterized using GC-MS instrumentation to determine the profiles of fatty acids contained in cincalok oil. The results of the GC-MS analysis are presented in Table 3. Based on the table, there are 19 types of fatty acids in cincalok oil extracted with five main components: palmitic acid, DHA, EPA, stearic acid, and phthalic acid with a percentage of 21.70 each; 10.99; 10.13; 9.93; and 9.06%. This results in a good agreement with Balange *et al.* (2017), who reported that dried Acetes contain EPA (13.11%) and DHA (8.19%). The slight difference in the DHA and EPA values of this study with the results reported by Balange *et al.* (2017) is due to differences in the origin and maybe the size of shrimp. From these results, it can be concluded that cincalok oil can be a good source of health-beneficial omega-3 fatty acids, *i.e.*, DHA and EPA. EPA and DHA play an essential role for skin health in maintaining skin moisture in lotion preparations. Palmitic acid has been reported to affect an antioxidant that functions as a catcher of the harmful effects of free radicals that cause skin damage, such as the appearance of wrinkles, scales, dryness, and cracks (Mieremet *et al.*, 2019).

The principal fatty acids in the marine shrimps, *Penaeus brasiliensis*, *Penaeus schimitti*, and *Xiphopenaeus kroyeri* were reported as palmitic acid, EPA, DHA, 13-octadecenoic acid, stearic acid, palmitoleic acid, and 5,8,11,14-eicosatetraenoic acid. The primary fatty acids found in the freshwater



(a)



(b)

Fig. 4. Chromatograms of cincalok oil (a) and standard astaxanthin (b).

**Table 3.** Fatty acids profile of cinalok oil.

Fatty acids	Formula	% fatty acids in the oil
Dodecanoic acid (lauric acid)	C <sub>13</sub> H <sub>26</sub> O <sub>2</sub>	0.55
Tetradecanoic acid (myristic acid)	C <sub>15</sub> H <sub>30</sub> O <sub>2</sub>	4.01
Pentadecanoic acid	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	0.91
9-hexadecenoic acid (palmitoleic acid)	C <sub>17</sub> H <sub>32</sub> O <sub>2</sub>	8.10
Hexadecanoic acid (palmitic acid)	C <sub>17</sub> H <sub>34</sub> O <sub>2</sub>	21.70
Heptadecanoic acid (margaric acid)	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	2.09
Stearidonic acid	C <sub>19</sub> H <sub>30</sub> O <sub>2</sub>	0.60
9,12-octadecadienoic acid (linoleic acid)	C <sub>19</sub> H <sub>34</sub> O <sub>2</sub>	1.37
13-octadecanoic acid	C <sub>19</sub> H <sub>34</sub> O <sub>2</sub>	8.28
Octadecanoic acid (stearic acid)	C <sub>19</sub> H <sub>38</sub> O <sub>2</sub>	9.93
Nonadecanoic acid	C <sub>20</sub> H <sub>40</sub> O <sub>2</sub>	0.67
5,8,11,14-eicosatetraenoic	C <sub>21</sub> H <sub>34</sub> O <sub>2</sub>	3.90
5,8,11,14,17-eicosapentaenoic acid (EPA)	C <sub>21</sub> H <sub>32</sub> O <sub>2</sub>	10.13
Eicosanoic acid	C <sub>21</sub> H <sub>42</sub> O <sub>2</sub>	2.00
4,7,10,13,16,19-docosahexaenoic acid (DHA)	C <sub>23</sub> H <sub>34</sub> O <sub>2</sub>	10.99
Docosanoic acid	C <sub>22</sub> H <sub>32</sub> O <sub>2</sub>	1.54
Phthalic acid	C <sub>24</sub> H <sub>38</sub> O <sub>4</sub>	9.06
Cholesterol	C <sub>27</sub> H <sub>46</sub> O	3.40
Tetracosanoic acid	C <sub>25</sub> H <sub>50</sub> O <sub>2</sub>	0.74

prawn *Macrobrachium rosenbergii* were palmitic acid, EPA, 13-octadecenoic acid, stearic acid, DHA, linoleic acid, and margaric acid (Bragagnolo and Rodriguez-Amaya, 2001). The fatty acid profile of cinalok oil indicated the presence of most of the fatty acids in Crustacea reported earlier.

### 3.5 Sun protector factor values of lotion formulated with cinalok oil

Because of the nutritional content in cinalok oil, it has the potential as a nutritional additive and sunscreen in lotion. The lotion is one of the emollients (softener) cosmetic preparations. The lotion consists of an oil phase and a water phase stabilized by an emulsifier and contains one or more active ingredients. In this study, cinalok oil was used as the active ingredient of the base lotion. The activity test of cinalok oil in the lotion was focused on determining the SPF value. The measurement of the SPF value was carried out using a UV-Vis Spectrophotometer to know how much the protection value of a lotion preparation was in protecting the skin from UV exposure. The SPF value of the lotion added with 5 and 10% cinalok oil was  $15.17 \pm 0.092$  and  $30.28 \pm 0.489$ , respectively. This SPF value is much higher than the SPF value of the base lotion ( $2.16 \pm 0.116$ ).

The higher the concentration of cinalok oil applied to the lotion, the greater the concentration of astaxanthin content, so the SPF value of the lotion is also higher. In addition, it may also be caused by the content of Zn and Fe, which in some studies have also been reported to be active as sun protectors. The lotion added with cinalok oil also did not change the homogeneity and pH. The pH range is at 7–8 and stable for 1 month of analysis time.

## 4 Conclusions

Cinalok is a new source that can be used as a nutritional enhancer and sunscreen in cosmetic lotions. The content of palmitic acid (21.70%), omega-3 especially DHA (10.99%) and EPA (10.33%), and astaxanthin ( $0.375 \pm 0.008$  mg/L) are the main components of cinalok oil. These components are very beneficial for skin health. There is no heavy metal contamination in the oil. The SPF value produced from cinalok oil lotion at 5 and 10% variations of cinalok oil was  $15.17 \pm 0.092$  and  $30.28 \pm 0.489$ , respectively. The SPF value of lotion with the addition of cinalok oil was much greater than that of the base lotion, which was  $2.16 \pm 0.116$ . For further studies, it is also necessary to consider extracting cinalok oil using solvents other than n-hexane, such as non-toxic eutectic solvents or vegetable oils.

*Acknowledgment.* This study received financial support from Tanjungpura University through Innovation Research 2020–2022. We thank SUCOFINDO Laboratory, Pontianak, West-Kalimantan and Integrated Research and Testing Laboratory, Gadjah Mada University, Yogyakarta for assistance with the analysis by ICP-AES and GC-MS.

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**Cite this article as:** Rahmalia W, Dasilia C, Usman T, Prayitno D I, Nurbaeti S N. 2022. Astaxanthine et huile riche en oméga-3 provenant d'*Acetes* fermentés (Cincalok) et son application comme additif bioactif et écran solaire dans une lotion. *OCL* 29: 18.