Potential Production of Polyunsaturated Fatty Acids from Microalgae

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Abstract - **Currently, public awareness of healthcare importance increase. Polyunsaturated fatty acid is an essential nutrition for us, such arachidonic acid, docosahexaenoic acid and eicosapentaenoic acid. The need of Polyunsaturated fatty acid generally derived from fish oil, but fish oil has a high risk chemical contamination. Microalgae are single cell microorganism, one of Phaeodactylum tricornutum which have relatively high content of eicosapentaenoic acid (29,8%). Biotechnology market of Polyunsaturated fatty acid is very promising for both foods and feeds, because the availability of abundant raw materials and suitable to develop in the tropics. This literature review discusses about the content of Polyunsaturated fatty acid in microalgae, omega-3, omega-6, Polyunsaturated fatty acid production processes, and applications in public health.**

Keywords: **poly-unsaturated fatty acid, PUFA, microalgae, health.**

INTRODUCTION

Polyunsaturated fatty acid (PUFA) are an essential component which cannot be synthesized by human. Therefore, PUFA must be provided through the diet (Ward and Singh, 2005). Omega-3 (PUFA n-3) and omega-6 (PUFA n-6) are well known derivative products of PUFA.

PUFAs are found in animals, plants, fungi, and microalgae. Nowadays, food habits in society are characterized by a high consumption of fast food which contain low portion of PUFAs. Some authors have compared the composition of fast food with Japanese food (Rubio-Rodriguez et al., 2010; Kamei et al., 2002) and Mediterranean food (Rubio-Rodriguez et al., 2010; Thellen and Ohrogge, 2002) that mostly source in fish. These authors concluded that the n-6:n-3 ratio in blood, in people who consume Japanese and Mediterranean food, is close to 2:1; while people who consume fast food, it can reach up to 25:1, so far than desired. For these reason, nutritionists educate the need to consume fish and green vegetables to reduce the risk of heart diseases (Rubio-Rodriguez et al., 2010). In contrast, the availability of fish is highly dependent on season may hamper the continuity of food (Kalogeropoulos et al., 2010), thus encouraging the discovery of alternative sources of PUFA. Basically, fish do not produce PUFA, they will get it by eating microalgae. This realization has turned microalgae into one of the most important producers of PUFA (Agustini and Kabinawa, 2005; Bigogno et al., 2002; Guedes et al., 2011; Kalogeropoulos et al., 2010;Ward and Singh, 2005).

Exploration of microalgae is not used as food diversification effort only, but also intended to empower agricultural land which is not feasible. As a tropical country, Indonesia has suitable temperature and contains high level of salt so it is feasible for the growth of microalgae (Agustini and Kabinawa, 2005). This literature review discusses about the content of PUFA in microalgae, PUFA production processes, and applications in public health.

Polyunsaturated Fatty Acid (PUFA)

Microalgae are autotrophic organisms which grow rapidly through photosynthesis like land based plants. Their unicellular structure allows them to convert solar energy into chemical energy easily (Harun et al., 2010). Microalgae can grow to high densities population by using only light, carbon dioxide, water, and other inorganic compounds (John et al., 2011).

Microalgae have a composition of carbohydrate, protein, and fats are widely used to meet food needs and public's health. Nutritional supplement products from microalgae can be found in form of powders, tablets, capsules, and concentrates (Table 1). The average composition of proteins, fats, and carbohydrates in microalgae are 12-35%; 7,2-23%; 4,6-23% (%dry weight) (Anonymous, 2011). Different nutrition, environment, and growth phases can influence the nutritional composition in microalgae, including fatty acid composition (Mata et al., 2010; Anonymous, 2011).

Fatty acids are organic compounds formed by long chain hydrocarbon and carboxylic group (Rubio-Rodriguez et al., 2010). Based on their nature hydrocarbon chain, fatty acids can be classified into saturated and unsaturated. Polyunsaturated fatty Acid (PUFA) are a long chain of unsaturated fatty acid. PUFA composition in the Cod liver oil and some microalgae are shown in Table 2.

Omega-3 fatty acids (PUFA n-3) are unsaturated fatty acids and contained in food as α-linolenic acid (ALA, C18:3, n-3) (Chew et al., 2008; Kalogeropoulos et al., 2010). ALA is the shortest chain of n-3 and mainly found in vegetables oil and nuts (Chew et al., 2008; Kalogeropoulos et al., 2010). Eicosapentaenoic acid (EPA, C20:5, n-3) and docosahexaenoic acid (DHA, C22:6, n-3) are derivatives product from n-3 found in fish and many other microorganisms, such as microalgae and bacteria (Guedes et al., 2011; Kalogeropoulos et al., 2010; Ward and Singh, 2005). ALA can be converted into EPA and DHA in the body, but the conversion is very limited and inefficient (Kalogeropoulos et al., 2010), therefore -3 must be provided in the form of dietary supplement.

TABLE 1. COMMERCIAL FOOD PRODUCTS FROM MICROALGAE (GOUVEIA, L., ET AL., 2008)

Microalgae	Producer	Product	Production (ton/year)
Spirulina	Hainan Simai Pharmacy Co. (China)	Powders, tablets, dan extract	3000
	Earthrise Nutritionals (USA)	Tablets, powders, drinks	
		Extract	
	Cynotech Corp. (USA)	Tablets, chip, liquor, liquid	
	Myanmar Spirullina Factory (Myanmar)	extract	
Chlorella	Taiwan Chorella Manufacturinh Co. (Taiwan)	Tablets, powders, nectar, mie	2000
	Klotze (Jerman)	Powders	
Dunaliella salina	Cognis Nutrition and health (Australia)	Powders of β-caroten	1200
Aphanizoenon flos-aquae	Blue Green Foods (USA)	Capsules, crystal	500
	Vision (USA)	Powders, capsules	

TABLE 2. PUFA COMPOSITION IN THE COD LIVER OIL AND SOME MICROALGAE (MEDINA, A.R., ET AL., 1997)

Organism	PUFA (% w of total fatty acid)				
	LA	AA	ALA	EPA	DHA
Isochrysis galbana	0,9	0,7	1,2	22,6	8,4
Phaeodactylum tricornutum	2,2	3,4	0,6	29,8	0,8
Porphyridium cruentum	6,2	23	1	23,9	0,2
Cod liver oil	1,5	2,7	0,8	12,5	9,9

EPA and DHA have positive effects on health. Recently, EPA proven to prevent coronary heart disease, and lowers blood cholesterol (Medina et al., 1999). DHA also has an important role in the development of central nervous system of infants (Medina et al., 1999). EPA is widely found in Porphyridium cruentum, and Monodus subterraneus (Ward and Singh, 2005), while the best source of DHA derived from microalgae with Schyzochytrium genus (Ward and Singh, 2005). Currently, some government agencies and nutritional organizations recommended daily intake level of DHA and EPA ranging from 0,2-0,3 g/day for the general population (Kalogeropoulos et al., 2010 ; Ruxton et al, 2004) up to 1,0-4,0 g/day for patient with coronary heart disease (Kalogeropoulos et al., 2010). Chemical structure of ALA, EPA, and DHA are shown in Figure 1.

Omega-6 fatty acid (PUFA n-6) are polyunsaturated fatty acid contained in food as linoleic acid (LA, C18:2, n-6) (Kalogeropoulos et al., 2010; McCusker et al, 2010). LA is the shortest chain from n-6 and arachidonic acid (AA, C20:4, n-6) is one of derivatives product from n-6.

AA is almost excluded from the water alga and most marine species. The only microalgae which produce AA in significant quantitative is the unicellular rhodophyte: P. cruentum (Bigogno et al., 2002). Other Rhodophytes such as Gracilaria sp has a composition of AA up to 60% of total fatty acids, but the dry weight content doesn't exceed 0,2% (Bigogno et al., 2002). AA is a biogenetic precursor of the biologically active prostaglandin and leucotrienes with important role in circulatory and central nervous systems (Medina dkk., 1999). AA is necessary for visual acuity, and it is valuable as an ingredient in formulations of artificial baby food (Medina dkk., 1999). Chemical structure of AA is shown in Figure 2.

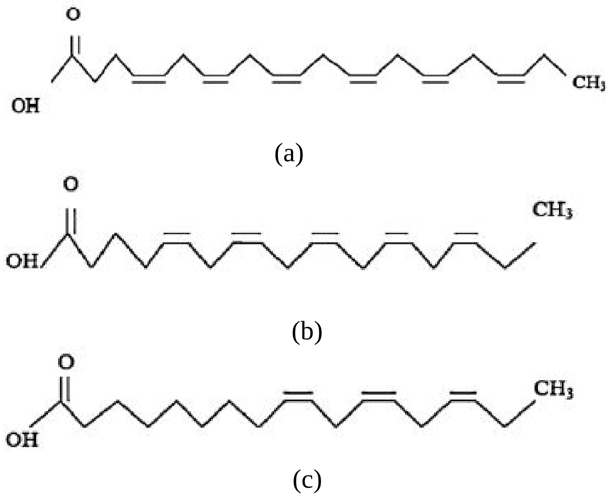


Figure 1. (a) DHA, (b) EPA, (c) ALA (McManus, A., et al., 2011)

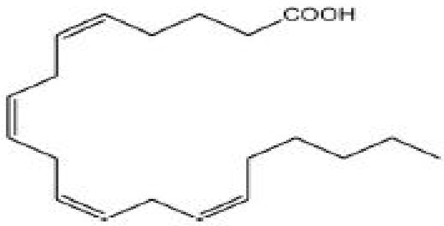


Figure 2. AA (Kiefer, J., et al., 2010)

Production of PUFA

Microalgae production consists of five important stages as shown in Figure 3.

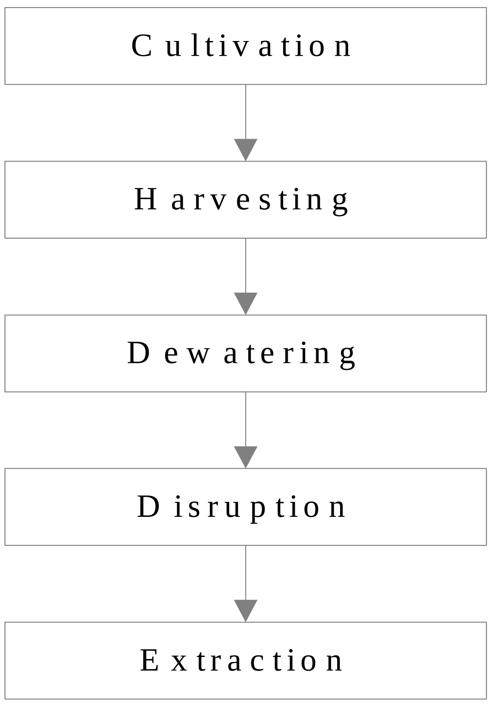
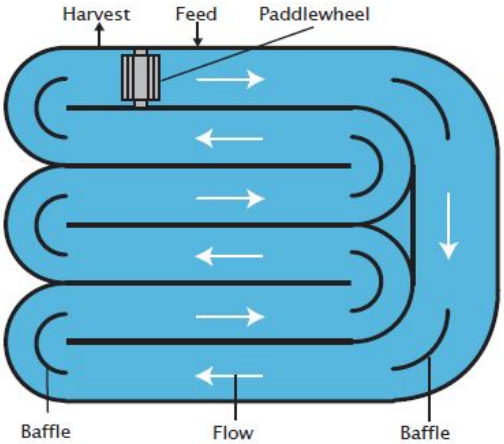


Figure 3. Microalgae production processes

Cultivation condition of microalgae. Most microalgae use light and carbon dioxide (CO2) as an energy and carbon sources (photoautotrophic organism) (Chisty et al, 2007; Wen et al, 2009). Temperature must remain generally 15-30˚C for optimum growth. Growth medium must provide inorganic elements that function in cell formation, such as nitrogen, phosphor, and iron (Agustini and Kabinawa, 2005; Chisti, 2007).

Cultivation technique of microalgae. Nowadays, there are two most commonly used techniques to cultivate microalgae. These are open race way ponds and photobioreactors (Harun et al., 2010). Open ponds are the oldest and simplest systems for cultivation (Chisti, 2007). Enclosed photobioreactors have been used to overcome the contamination and evaporation problems encountered in open ponds (Wen et al, 2009). These systems are made of transparent materials and commonly placed outdoors for illumination by natural lights (Harun et al., 2010). Photobioreactors have a large surface area to volume ratio (Chisti, 2007; Wen et al, 2009). Microalgae productivity of photobioreactors can reach 13 times more than open ponds (Chisti, 2007; Wen et al, 2009). Figure 4 shows the open raceway ponds and photobioreactors systems.



(a)

(b)

Figure 4. (a) open raceway pond and (b) photobioreactor (Wen, Z., at al., 2009).

Dewatering of microalgae. Flocculation, centrifugation, and filtration are commonly techniques for dewatering microalgae (Agustini and Kabinawa, 2005; Borowitzka, 1999; Harun et al., 2010). Flocculation can be used as initial step that will enhance the ease of next processing. Microalgae have a negative charge, thus they need cationic chemical flocculants for coagulating biomass, such as $Al_2(SO_4)_3$, $FeCl_3$, dan $Fe_2(SO_4)_3$ (Borowitzka, 1999; Harun et al., 2010). Filtration has proved to be the most competitive technique (Harun et al., 2010). The types of filtration that can be used are dead end filtration, microfiltration, ultrafiltration, pressure filtration, and tangential flow filtration (Harun et al., 2010).

Disruption is the separation process of lipids and other non lipids chain (Medina, A.R., et al, 1997). This process is necessary before extraction stage. Disruption agents must not degrade the lipids (Medina, A.R., et al, 1997). There are five methods of disruption that can be used, (i) autoclave (125˚C, 1,5 MPa), (ii) bead-beater (bead diameter 0,1 mm, speed 2800 rpm), (iii) microwave (100˚C, 2450 MHz), (iv) sonication using sonicator (resonance 10 KHz for 5 min), and (v) osmotic shock processes (Lee, J.Y. et al, 2010).

Extraction of microalgae can be classified into two methods, (1) mechanical methods (Expression/Expeller press and Ultrasonic-assisted extraction) and chemical methods (Hexane Solvent Method, Soxhlet extraction, and Supercritical fluid Extraction) (Mata, et al, 2010; Anonymous, 2011). Each of these methods has drawbacks: (a) the mechanical methods requires drying process which is energy intensive, (b) the use of chemical solvent should be considered to the level of safety and health, (c) supercritical extraction requires a high pressure equipment that is expensive and energy intensive (Anonymous, 2011).

Supercritical Fluid Extraction (SFE)

Figure 5 shows pressure-temperature phase diagram for carbon dioxide. In this diagram, the vapor-liquid equilibrium curve starts at the triple point (TP), where both of three phases, solid, liquid, and vapor are in equilibrium. This curves ends at the critical point (CP), where the meniscus separating vapor and liquid phases disappears and only a single phases formed.

Carbon dioxide (CO_2) is in the supercritical state if its temperature and pressure higher than its critical temperature (304,1K) and pressure (7,38MPa). In its critical region, CO_2 is sensitive with very small changes in temperature and pressure (Mendes et al, 2003). Supercritical CO_2 has been the most used supercritical fluid, because non-flammable, non-toxic, inexpensive, and relatively inert (Mendes et al, 2003). On the other hand, the adding of entrainer, a substance which has a volatility intermediate between the compound to be extracted and the supercritical fluid, can increase the solvent power of CO_2 (Mendes et al, 2003). Due to its non polar behavior, CO_2 cannot be used to extract the polar compound. However, the addition of polar entrainer such as water, ethanol, and methanol, thus polar compound extraction can be done (Mendes et al, 2003).

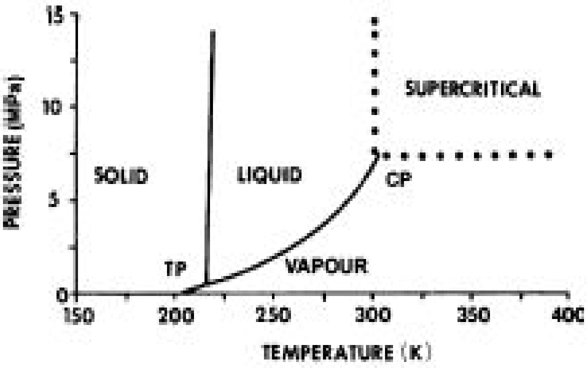


Figure 5. Pressure – temperature phase diagram for carbon dioxide (CO_2) (Mendes, R.L., et al., 2003).

CONCLUSIONS

PUFA are polyunsaturated fatty acid that has positive effects on health such as lowering cholesterol level and the risk of heart disease. Due to their continuity of their raw materials, easily developed in the tropics, and not susceptible to chemical contamination, microalgae are an alternative source of PUFA which is potential to be developed in Indonesia. PUFA extraction from microalgae can be done by several methods, mechanical and chemical methods. The potency of microalgae as feedstock PUFA is very large, it is necessary to study deeper about the extraction and purification of PUFA which is more economic with good quality and quantity.

ACKNOWLEDGEMENTS

The authors would like to thank Dr. Hadiyanto, MSc and Dessy Ariyanti, ST, MT for their important contributions to this paper.

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