CHAPTER 27

Seaweed Lipids as Nutraceuticals

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Contents

Abstract

Seaweeds are known as low-energy food. Despite low lipid content, ω-3 and ω-6 polyunsaturated fatty acids (PUFAs) introduce a significant part of seaweed lipids. PUFAs are the important components of all cell membranes and precursors of eicosanoids that are essential bioregulators of many cellular processes. PUFAs effectively reduce the risk of cardiovascular diseases, cancer, ostheoporosis, and diabetes. Because of the frequent usage of seaweeds in Asia and their increasing utilization as food also in other parts of the world, seaweeds could contribute to the improvement of a low level of ω-3 PUFAs, especially in the Western diet. The major

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commercial sources of ω-3 PUFAs are fish, but their wide usage as food additives is limited for the typical fishy smell, unpleasant taste, and oxidative nonstability. Nevertheless, growing requirements of healthy functional foods have led to produce PUFAs as nutraceuticals in controlled batch culture of marine microalgae, especially *Thraustochytrium* and *Schizochytrium* strains.

I. INTRODUCTION

Lipids belong to fundamental nutrients for human. Their components are fatty acids (FAs), which could be classified to saturated (SFAs—without double bonds), monosaturated (MUFAs—with one double bond), and polyunsaturated FAs (PUFAs—with two or up to six double bonds). Humans are able to synthesize both SFAs and MUFAs. Nevertheless, PUFAs with the first bond on the third or sixth carbon atom are essential because of the inability to be synthesized by the human body. Thus, they have to be obtained from the diet. Their main sources are chloroplasts of higher plants and fat of water organisms.

Nowadays, essential FAs (EFAs) are considered to be functional food and nutraceuticals with many health benefits including the potential of reducing the risk of cardiovascular diseases (CVD), cancer, ostheoporosis, and diabetes. CVD have been believed to be the main cause of death in most Western countries. Coronary heart disease (CHD) is closely connected with a progress of atherosclerosis evoked by interactions between plasma lipids, lipoproteins, monocytes, platelets, endothelium, and smooth muscle of arterial walls, which results in narrowing of coronary arteries. Thus, the composition of dietary lipids is an important factor of genesis of hearth diseases together with the quality and alluviation of arterial walls. This could lead to thrombosis and finally to coronary infarctions.

Dietary pattern has been modified throughout the human evolution. The origin composition of a hunter-gathered diet with a lower intake of total fats has been altered by a higher intake of total lipids with a high representation of saturated and trans-FAs, which are detrimental for health. Contemporary Western human diet is noted for a low content of ω-3 EFAs that results in an imbalance of ω-3 and ω-6 EFAs and in a progress of various pathophysiologies.

II. HEALTH IMPORTANCE OF PUFA

Dispensability of PUFAs for human has already been known for many decades. Oversized dietary intake of most SFAs and *trans*-FAs is harmful for health due to increasing the risk of CVD. The human body cannot synthesize PUFAs with the first double bond on the C3 and C6 from the

methyl-end. These FAs are EFAs, and their level in the human body depends on their intake from the diet. EFAs form two biologically important groups which are ω-3 and ω-6 EFAs according to the location of their first double bond from the methyl-end of FAs. They are also called long chains ω-3 and ω-6 PUFAs (LCPUFAs). However, some recent studies have concluded that humans of every age could transform α-linolenic acid (ALA, 18:3, ω-3) to docosahexaenoic acid (DHA, 22:6, ω-3) but only in the insufficient concentration (Brenna, 2002; Brenna *et al.*, 2009; Burdge and Calder, 2005; Burdge and Wootton, 2002).

In Fig. 27.1, there are shown the metabolic transformations of ω-3 and ω-6 PUFAs and their important derivates such as prostaglandins (PG), thromboxanes (TX), and leukotrienes (LK).

It is generally known that primary precursors of ω-3 and ω-6 EFAs are ALA and linoleic acid (LA), respectively. Both are formed by the gradual desaturation of oleic acid in the endoplasmic reticulum and chloroplasts of plantae. Because of the absence of Δ^{12} and Δ^{15} desaturases required for the synthesis of ALA from stearic acid (18:0), humans cannot synthesize ALA. It has to be obtained from the diet.

LCPUFAs are formed by series of reactions that are catalyzed by desaturases and elongases. Further, conversion of dietary ALA (18:3, ω-3) into EPA (20:5, ω-3) is limited because ALA and LA (18:2, ω-6) compete for common desaturation and elongation enzymes. The affinity of Δ^6 desaturase for ALA is greater than for LA (Burdge and Calder, 2005). It was proved that the relationship between ALA and LA is very important for the maintenance of their homeostasis. But the amount of ALA and LA in the diet is significant for ALA conversion to EPA (20:5, ω-3) and DHA and not for their ratio (Goyens *et al.*, 2006).

Polyunsaturated ω-3 LCPUFAs have significant roles in many biochemical pathways which result in different health promotion activities. Generally, LCPUFAs show cardioprotective effect that results from their considerable anthiatherogenic, antithrombotic, anti-inflammatory, antiarrhythmic, hypolipidemic effect and other health benefits, which are based on complex influence of concentrations of lipoproteins, fluidity of biological membranes, function of membraned enzymes and receptors, modulation of eicosanoids production, blood pressure regulation, and finally on the metabolism of minerals (Flachs *et al.*, 2005; Hu *et al.*, 2001; Kinsella *et al.*, 1990; Tvrzická *et al.*, 2009; Weiss *et al.*, 2005).

PUFAs of ω-3 series have many pleiothropic metabolic effects as ligands of peroxisome proliferator-activated receptors (PPAR-α). It is assumed that the activation of PPAR-α results in decrease of lipogenesis and secretion of a very low density lipoprotein (VLDL), further in growth of lipoprotein lipase activity and decrease of apolipoprotein C-III concentration, and on increased reverse transport of cholesterol (Corton and Anderson, 2000; Olivieri *et al.*, 2003; Tvrzická *et al.*, 2009).

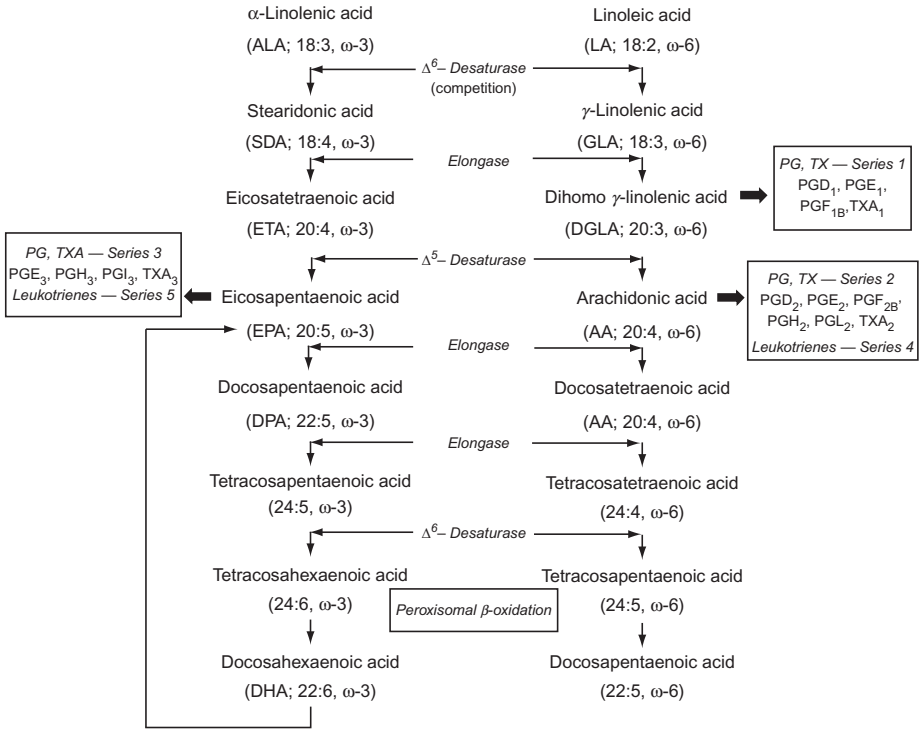

FIGURE 27.1 The metabolic transformation of ω-3 and ω-6 PUFAs and their derivates.

EPA and DHA are fundamental EPAs from ω-3 series of LCPUFAs. DHA is the main structural component of cell membranes, at high level in brain tissue and retina. DHA is formed from EPA by peroxisomal β-oxidation (Burdge and Calder, 2005). EPA and DPA (22:5, ω-3) can also be synthesized from DHA via β-oxidation in peroxisomes by catalytic activity of probably Δ-4 enoyl CoA reductase and Δ-2 enoyl CoA isomerase (Grønn *et al.*, 1991).

The principal LCPUFA of ω-6 series is arachidonic acid (AA; 20:4) acting as a precursor for eicosanoids synthesized from LA. LCPUFAs of ω-6 series have been considered as activators of PPAR-γ. Their metabolic effects include increased synthesis of cholesterol, increased activity of LDL receptors, increased activity of cholesterol 7 α-hydroxylase (Cyp 7A1), and decreased conversion of VLDL to LDL. As ligands of PPAR-γ, ω-6 PUFAs may improve insulin sensitivity, change fat distribution, and affect adipocyte differentiation (Chiang *et al.*, 2001; Corton and Anderson, 2000).

Biological activities of individual EFAs might be derived from the course of their interactions. Their major derivates are eicosanoids, signaling molecules having important functions in many regulation systems and performing as messengers in the central nervous systems (Hertting and Seregi, 1989; Leslie and Watkins, 1985).

Eicosanoids are divided into four following classes: PG, TX, prostacyclins, and LK. Further within each class, there are two or three series of eicosanoids. Eicosanoids derived from ω-3 and ω-6 FAs have antagonistic effects. Their amount depends on the composition of dietary FAs influenced by the competition with AA and EPA FAs as substrates for cyclooxygenases and 5-lipoxygenases (Kinsella *et al.*, 1990; Simopoulos, 2002a,b).

PG are oxygenated, unsaturated cyclic FAs responsible for the processes of many hormone-like actions. Arachidonic acid ω-6 PUFA is converted to an unstable intermediate hydroxyl-endoperoxide prostaglandin H_2 which is subsequently converted to PGE_2 by the enzymatic activity of cyclooxygenase-2 (COX-2). PGE_2 as proinflammatory eicosanoids have been related to carcinogenesis of breast and prostate, as well as cancer initiation (Kobayashi *et al.*, 2006; Terry *et al.*, 2003). EPA and DHA from marine oils inhibit COX-2 and suppress the production of PGE_2. It has been proved that EPA and DHA also inhibit lipoxygenases contributing to synthesis of hydroxyeicosatetraenoic acids and LK. 12-Hydroxyeicosatetraenoic acid has been connected with the suppression of apoptosis, stimulation of angiogenesis, and further with stimulation of tumor cell adhesion (Rose, 1996).

A. Significance of PUFAs in human diet

Lipids represent one of the main sources of energy for human metabolic processes. Lipid consumption in most Western countries is relatively high with the contribution of approximately 40% of total calories (Narayan

et al., 2006), despite the nutritious recommendation that 25% of energy should be covered by lipids (Sugano and Hirahara, 2000). Qualities of lipids are derived from their FAs composition which is various according to their sources. In general, vegetable oils from terrestrial plants are composed from SFAs and unsaturated FAs (UNFAs) with the chains formed by 16- and 18-carbon molecules, whereas the representation of individual FAs depends on plant species. Nevertheless, oils origined from marine organisms consist typically of UNFAs with the abundant amount of EPA and DHA, especially (Hu *et al.*, 2001).

The absolute amount of lipids in the diet is not the main promoter of CVD. The important factor is relative concentration and distribution of dietary FAs with proved effects on lowering a risk of CVD (Cordain *et al.*, 2002). Relative concentrations and distribution of dietary EFAs are different among various nationalities because of diverse dietary patterns. In Fig. 27.2, there are demonstrated trends of the total male and female mortality (CHD, CVD) in different countries in comparison with the distinct dietary intake of LCPUFAs. There is an evident dependency of the highest mortality in the countries with the lowest intakes of dietary *n*-3 LCPUFAs. The graphs have been constructed from data of several studies (Astorg *et al.*, 2004; Hibbeln *et al.*, 2006; Kris-Etherton *et al.*, 2000; Meyer *et al.*, 2003; Miyake *et al.*, 2010).

Typical Western diet with oversized intake of ω-6 PUFAs (LA-rich oils from vegetable sources) leads to overproduction of proinflammatory ω-6 PG and cytokines, which could be suppressed by higher intake of ω-3 PUFAs from fish oils. Simopoulos (2002a,b) reported that high intake of ALA (about 15 g/day) would suppress human protein interleukin (IL-1) and tumor necrosis factor.

Many studies have been conducted on marine fish oil consumption and relation to risk of breast or prostate cancer. The inhibition of eicosanoids production from ω-6 PUFAs by higher consumption of fish oil with high levels of ω-3 PUFAs, which is a common feature of lowering a cancer risk, was reported (Bagga *et al.*, 1997; Terry *et al.*, 2004).

Eicosanoids derived from AA are biologically active in small quantities. Their large amounts lead to the formation of thrombi and atheromas, and to the development of allergic and inflammatory disorders (Simopoulos, 2002a,b). In general, ω-6 PUFAs have been associated with the enhancement of the promotional phase of mammary carcinogenesis (Rose, 1997).

However, contradictory results of studies on the effect of ω-3 and ω-6 PUFAs have been reported. It has been shown that AA also inhibits the growth of A549 human lung adenocarcinoma cells, even though DHA has been more effective than AA (Trombetta *et al.*, 2007).

Differences between *cis*- and *trans*-configuration of PUFAs and the implication of their dietary intake on the human health have also been

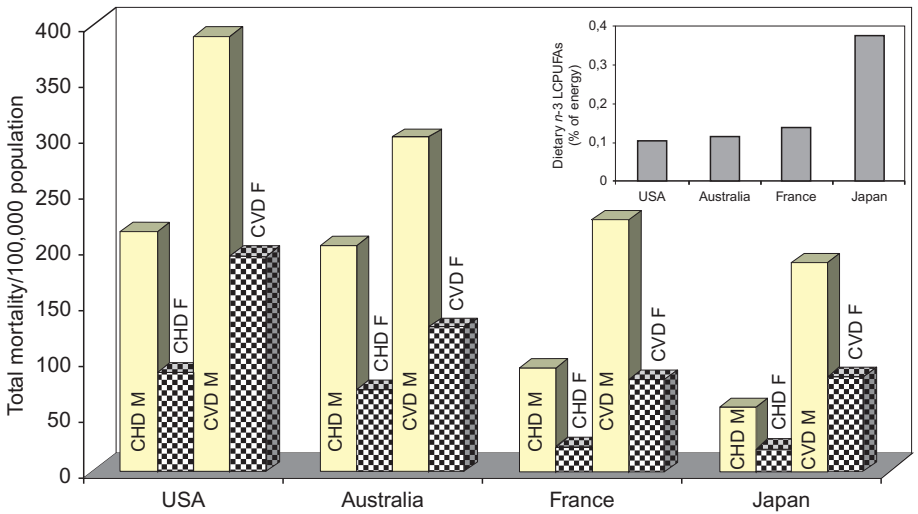

FIGURE 27.2 Trends of total man and female mortality (CHD, CVD) in some countries with dietary intake of LCPUFAs.

reported. Trans isomers of monounsaturated octadecenoic acid (C18:1) were found as the most common *trans*-FAs in the diet of many European countries (Hulshof *et al.*, 1999).

Recommended intakes of ω-3 LCPUFAs were often discussed in the scientific quarters and varied in different countries because of dissimilar dietary intake of ω-3 LCPUFAs. Approximate estimation of the consumption of ω-3 LCPUFAs is 0.1–0.5 g/day in Europe, 0.1–0.2 g/day in the United States, while in Japan, it is higher up to 2 g/day (Gómez Candela *et al.*, 2011) due to higher consumption of fish.

B. The ω-6/ω-3 ratio as health-promoting factor

The significance of the ω-6/ω-3 ratio has also been discussed many times in research papers within the context of evolutionary aspects of the human diet. The origin ratio of ω-6/ω-3 was 1. Nowadays, the new lifestyle with the alteration of dietary pattern has caused the change of dietary intake of lipids, especially the distribution of ω-3 and ω-6 PUFAs in Western countries. At the beginning of the twentieth century, the consumption of vegetable oils and fats has risen. These oils and fats are responsible for an excessive dietary level of ω-6 PUFAs and a lowering concentration of ω-3 PUFAs in the Western diet (Cordain *et al.*, 2005). Further contributor of altered composition of received FAs is the oversized consumption of margarine and shortening produced from refined vegetable oils by hydrogenation process resulting in the production of *trans*-isomers of FAs (Hu *et al.*, 2001). The modification of present dietary pattern has led to higher intake of ω-6 PUFAs and that has caused an increase of the ω-6/ω-3 ratio up to 20–30:1 (Gómez Candela *et al.*, 2011). The relationship between low ω-6/ω-3 ratio and rare occurrence of CHD in Inuits has already been described (Bjerregaard *et al.*, 2003). The significance of the balance of the ω-6/ω-3 ratio is based on the fact that mammalian cells cannot convert ω-6 to ω-3 FAs due to the absence of the converting enzyme, omega-3 desaturase (Simopoulos, 2006). However, the significance of this ratio has been challenged on behalf of separate recommendations for ALA, marine ω-3 PUFAs, and LA (de Deckere *et al.*, 1998).

III. LIPIDS AND PUFAS IN SEAWEED

Primary product of ω-3 FAs in trophic chain for fish is a product of marine microorganisms and algae. Fish and fish oil are considered as the main source of PUFAs, especially DHA (C22:6, ω-3) and AA (C20:4, ω-6). However, fish do not synthesize these EFAs, and their high level in fish oil results from the diet composed of marine zooplankton fed on phytoplankton (Yap and Chen, 2001; Yongmanitchai and Ward, 1993).

A. The variability of seaweed lipid composition

Lipid content and FA composition of seaweed are very changeable in dependence on different environmental conditions. It was reported that different levels of light and salinity could have positive or negative effect on the content and distribution of FAs across all groups of seaweeds (Floreto and Teshima, 1998). The lipid metabolism in algae is also influenced by different season period. In brown seaweed *Costaria costata*, the higher content of total lipids was established in May, whereas the abundant amount of storage lipids (triacylglycerols—TAGs) was observed in July; ω-3 PUFAs were prevailing in April, but the level of ω-6 PUFAs was similar in spring and summer (Gerasimenko *et al.*, 2010). According to Kim *et al.* (1996), seasonable changes in the connection with lipids and FAs were observed also in brown seaweed *Fucus serratus*. While the highest contents of total lipids and storage lipids (TAG) were in summer, the lowest contents of both total lipids and TAG were established in spring. However, Honya *et al.* (1994) concluded that in another brown seaweed *Laminaria japonica* was the lowest content of total lipids and SFAs in midsummer, whereas the highest amount of ω-6 PUFAs was established during summer and ω-3 PUFAs content culminated during the cold months. These variabilities of lipid composition could be connected with the level of nitrogen in the seawater which is also a factor affecting the lipid contents in seaweeds. Nitrogen deficiency has been reported to cause the poor algal biomass production and lower lipid content. Nitrogen supplementation has led to increased amount of lipids in red seaweed *Palmaria palmata* (Mishra *et al.*, 1993).

Further variation of lipid metabolism has been observed as the dependence on the environmental pollution by heavy metals (Cu, Cd, and Pb), herbicides, and also in the consequence of manganese deficiency in a cultivation medium for photosynthetic algae (Constantopoulos, 1970).

Finally, differences of lipid composition, in general, between wild and cultured strains of various plants and seaweeds were reported (Saito *et al.*, 2010; Simopoulos, 2002a,b, 2004). In contrast to that, Mishra *et al.* (1993) observed no difference in the lipid contents of wild and cultured strains of *P. palmata*, but wild strains of *P. palmata* contained lower amount of nonpolar lipid fractions.

B. The lipid composition of seaweeds

Seaweeds are known as low-energy food. Lipid content in commonly used seaweeds does not obviously exceed 5% of dry matter. Nevertheless, the main part of lipids is formed by wide range of FAs. In Tables 27.1 and 27.2, lipid contents and FA profiles of some commercially used seaweeds from several studies are shown (Dawczynski *et al.*, 2007; Kamlangdee and

TABLE 27.1 Content of some FAs (% of total FAMEs) and total lipids (TL; % of dry matter) in some brown seaweed

FAs	*Laminaria* sp.[a]	*Laminaria japonica*[b]	*Laminaria japonica*[c]	*Undaria pinnatifida*[a]	*Undaria pinnatifida*[b]	*Undaria pinnatifida*[d]	*Hizikia fusiformis*[a]	*Hizikia fusiformis*[c]
C14:0	2.88	5.30	6.84	2.25	4.40	3.17	0.76	7.19
C16:0	36.00	12.30	35.40	13.50	26.80	16.51	26.80	41.40
C18:0	1.49	1.00	6.15	0.86	2.90	0.69	0.30	2.60
$C16:1_{\omega7}$	1.71	3.90	0.68	0.44	0.10	3.70	0.15	6.58
$C16:3_{\omega4}$						2.31		
$C18:1_{\omega9}$		8.40			17.90	6.79		
∑C18:1	12.80		31.80					16.10
$C18:2_{\omega6}$ (LA)	5.48	8.40	8.30	7.41	6.20	6.23	3.56	5.96
$C18:3_{\omega3}$ (ALA)	0.76	6.10		11.20	5.80	11.97		
$C18:3_{\omega6}$ (GLA)	1.60	4.20		1.71	1.20			
∑C18:3			2.37					7.40
$C18:4_{\omega3}$	1.24	13.90		25.80	8.70	22.60		
$C20:1_{\omega9}$	1.55		0.53				4.09	
$C20:2_{\omega6}$							0.97	0.44
$C20:3_{\omega6}$ (DGLA)	0.01			0.57			3.21	
$C20:4_{\omega6}$ (AA)	12.40	14.00	6.65	13.30	12.70	15.87	5.30	10.40
$C20:5_{\omega3}$ (EPA)	16.20	14.00		13.20	7.50	9.43	42.40	
$C22:6_{\omega3}$ (DHA)								
ω6/ω3	1.3:1	0.81:1		0.5:1	0.94:1	0.49:1	0.3:1	
TL	1.00	0.58	1.83	4.50	0.38	1.05	1.40	1.17

[a] Dawczynski *et al.* (2007).
[b] Khotimchenko (1998).
[c] Mišurcová, Ambrožová, and Samek (unpublished datas).
[d] Sánches-Machado *et al.* (2004).

TABLE 27.2 Content of FAs (% of total FAMEs) and total lipids (TL; % of dry matter) in red and green seaweed and *Schizochytrium* sp.

	Red seaweed						Green seaweed		Brown microalga
FAs	*Palmaria* sp.[a]	*Palmaria palmata*[b]	*Porphyra* sp.[a]	*Porphyra* sp.[c]	*Porphyra* sp.[b]	*Laurencia papillosa*[d]	*Ulva lactuca*[d]	*Ulva tubulosa*[d]	*Schyzochytrium* sp.[e]
C14:0	13.76	9.67	0.53	2.68	2.93	4.87	2.74	1.13	
C16:0	45.44	54.10	63.19	30.80	47.60	37.80	43.00	49.20	19.7–29.4
C18:0	1.28	8.28	1.23	0.66	2.91	1.86	3.27	4.22	0.7–1.3
$C16:1_{\omega7}$	5.26	2.66	6.22	2.24	2.77	2.25	5.77	2.05	
$C16:3_{\omega4}$	1.20		1.56						
$C18:1_{\omega9}$	3.13		6.70			2.14	17.80	18.60	
$\sum$C18:1		4.98		7.16	13.30				
$C18:2_{\omega6}$ (LA)	0.69	1.47	1.17	3.86	3.13	2.72	9.44	10.50	
$C18:3_{\omega3}$ (ALA)			0.23	5.66			2.25	1.02	0.1–0.4
$C18:3_{\omega6}$ (GLA)				0.31		0.31	3.22	2.24	
$\sum$C18:3		0.20			1.20				
$C18:4_{\omega3}$	0.74		0.24	3.37					
$C20:1_{\omega9}$		0.45	4.70	1.42	2.25				
$C20:3_{\omega6}$ (DGLA)									
$C20:4_{\omega6}$ (AA)	1.45		6.80	8.00		21.40	2.50	1.83	0.3–0.9
$C20:5_{\omega3}$ (EPA)	24.05		6.03	20.90		22.80	0.87	2.37	0.5–1.1
$C22:6_{\omega3}$ (DHA)									30.3–36.1
ω6/ω3	0.13		1.21	0.6:1		1.07	3.03	1.83	
TL	1.80	0.64	1.03	2.80	0.93	1.73	1.27	2.13	

[a] Sánches-Machado *et al.* (2004).
[b] Mišurcová, Ambrožová, and Samek (unpublished datas).
[c] Dawczynski *et al.* (2007).
[d] Kumari *et al.* (2010).
[e] Kamlangdee and Fan (2003).

Fan, 2003; Khotimchenko, 1998; Kumari *et al.*, 2010; Mišurcová, Ambrožová, and Samek, unpublished datas; Sánches-Machado *et al.*, 2004).

From these tables could be concluded that lipid content and FAs-composition are very changeable between different groups of seaweeds, and even within the same species. The content of total lipids ranged from 0.4% to 4.5% of dry seaweed matter. It is evident that palmitic acid was the most abundant SFA in all genera seaweeds.

Red seaweeds predominantly contained polyunsaturated 20C-PUFAs—eicosapentaenoic acid (EPA; ω-3, C 20:5) and AA (ω-6, C 20:4). Further, PUFAs with 18 and 20 carbon molecules were the most abundant in selected brown seaweeds. Finally, 18C-PUFAs were predominantly determined in the chosen green seaweeds. For comparison, FAs-composition of brown microalga *Schizochytrium* sp. as the significant source of DHA is also introduced in Table 27.2.

C. Distribution of PUFAs in seaweed lipids

Various FA compositions have been established in different groups of seaweed lipids. In algal lipids, there are presented two classes of polar and nonpolar lipids. TAGs have a storage function and form the main part of seaweed lipids. However, the group of polar lipids (phospholipids, glycolipids, and sulpholipids) is the main structural part of all cell membranes, which have the crucial function for the living processes (Gerasimenko *et al.*, 2010; Mishra *et al.*, 1993). Generally, acylglycerides were found better for human utilization like esters due to their easy incorporation into the plasma (Linko and Hayakawa, 1996).

As far as PUFAs distribution in seaweed lipids is concerned, it has been reported that majority of PUFAs has been distributed in TAGs (Gerasimenko *et al.*, 2010; Mishra *et al.*, 1993; Saito *et al.*, 2010). According to the same pattern as FA composition of total seaweed lipids, the abundant amount of SFAs palmitic acid was obviously observed in all parts of seaweed lipids. However, in red seaweed *P. palmata*, the highest amount of palmitic acid was in polar lipids, while in the TAGs, there was the lowest concentration of it. The abundant PUFA in all lipid parts was EPA; nevertheless, it was in the highest concentration in the TAGs (Mishra *et al.*, 1993).

IV. THE LIPID COMPOSITION OF MARINE MICROALGAE

Some marine microalgae from the kingdom Chromista and Protozoa were observed as a rich natural source of PUFAs. Thraustochytriacea is the significant family of Chromista in which have been included generas *Schizochytrium* and *Thraustochytrium* (Yokoyama and Honda, 2007) with

the ability to form a high level of DHA, particularly (Lewis *et al.*, 1999). It was published that Δ^4 desaturase in marine heterokont brown algae *Thraustochytrium* sp. is responsible for the direct conversion of DPA (22:5, *n*-3) to DHA (22:6, *n*-3) in contrast to fish and mammals (Qiu *et al.*, 2001). It was also proved that Δ^4 desaturase gene (Fad4) from *Thraustochytrium* sp. could be transfected into human lymphocytes. This fact is significant for its utilization for perspective increase of human DHA and also for the treatment of patients with the Zellweger syndrome, which causes metabolic defect in DHA synthesis (Martinez *et al.*, 2010). It was observed that *Thraustochytrium* sp. could accumulate more than 50% of its lipids in the form of DHA (Ward and Singh, 2005) and *Schizochytrium* sp. 35.6% of total lipids in the form of DHA (Yaguchi *et al.*, 1997). *Crypthecodinium cohnii*, red marine microalga from the kingdom Protozoa, was identified also as a good natural source of DHA that produces no other PUFAs. DHA forms 99.2% of total lipids in this alga (Mendes *et al.*, 2007, 2009).

Different distribution of FAs in various parts of microalgal lipids as well as in seaweed lipids was established. According to Fan *et al.* (2007), marine microalga of strain *Schizochytrium mangrovei* contained 68% of total lipids of dry cell weight and 93% of FAs were distributed in TAGs. Interestingly, a higher amount of PUFAs was determined in polar lipids, fundamental components of cell membranes, with the high degree of unsaturation of the fatty acyl groups responsible for their normal functions. Lipids of *S. mangrovei* were primarily composed of palmitic FA and DHA, 50.3% and 29.7% of total lipids, respectively. With respect to DHA distribution, it was the major PUFA in all lipid classes. The highest amount of DHA was primarily contained in TAGs, which reached 93.6% of the total DHA.

Thraustochydrids strains could be used for the commercial production of DHA for an infant formula (Ward and Singh, 2005). Currently, the production of DHA by marine microalgae is the subject of intensive research because of the fact that microalgae oil has the advantage of presenting neither an unpleasant odor nor a high amount of cholesterol and contains squalene and phytosterols, which have additional benefits to human health (Rubio-Rodríguez *et al.*, 2010).

V. CONCLUSION

Nowadays, process of finding new sources of ω-3 PUFAs has been continuing. Their major commercial sources are fish and fish fat. EPA and DHA produced from fish oil are used as nutraceuticals and functional ingredients in industrial foods. However, their wide usage as food additives is limited for the typical fishy smell, disagreeable taste, and

finally for the oxidative nonstability. Further, chemical forms of PUFAs are important for their better utilization by human.

Nevertheless, the growing requirements of healthy functional foods and escalation of the environment pollution on the worldwide basis have led to produce PUFAs in controlled batch culture of marine microalgae, especially of *Thraustochytrium* and *Schizochytrium* strains. Besides marine microalgae, seaweeds seem to be an interesting natural source of ω-3 PUFAs, thanks to their better utilization.

Seaweed lipids are presented in a very small amount that does not exceed 5% of dry seaweed matter. Despite this low lipid content in seaweeds, ω-3 and ω-6 PUFAs represent the significant part of seaweed lipids. LCPUFAs of ω-3 and ω-6 series are precursors of eicosanoids which are important bioregulators of many cellular processes. Lipids composition of seaweed is very changeable due to the adaptation mechanism improving their tolerance to the environmental conditions.

In spite of health benefits of ω-3 PUFAs for human, the unfavorable effects within the context of their easy auto-oxidation should not be ignored. This fact means the incorporation of expensive steps during purifying process in the industrial production of PUFAs.

Seaweed could be potentially used in the production of low fat foods due to their high level of important PUFAs. Functional food products enriched with ω-3 LCPUFAs are widely spread nowadays. The utilization of marine microalgae to produce high value lipids is suggested as the alternative sources of PUFAs. The utilization of algal enzymes in order to produce EPA and DHA from modified crops seems to be also the perspective interest.

REFERENCES

Astorg, P., Arnault, N., Czernichow, S., Noisette, N., Galan, P., and Hercberg, S. (2004). Dietary intakes and food sources of n-6 and n-3 PUFA in french adult men and women. *Lipids* **39**, 527–535.

Bagga, D., Capone, S., Wang, H. J., Heber, D., Lill, M., Chap, L., and Glaspy, J. A. (1997). Dietary modulation of omega-3/omega-6 polyunsaturated fatty acid ratios in patients with breast cancer. *J. Natl. Cancer Inst.* **89**, 1123–1131.

Bjerregaard, P., Young, T. K., and Hegele, R. A. (2003). Low incidence of cardiovascular disease among the Inuit—What is the evidence? *Atherosclerosis* **166**, 351–357.

Brenna, J. T. (2002). Efficiency of conversion of α-linolenic acid to long chain n-3 fatty acids in man. *Curr. Opin. Clin. Nutr. Metab. Care* **5**, 127–132.

Brenna, J. T., Salem, N., Jr., Sinclair, A. J., and Cunnane, S. C. (2009). α-Linolenic acid supplementation and conversion to n-3 long-chain polyunsaturated fatty acids in humans. *Prostaglandins Leukot. Essent. Fatty Acids* **80**, 85–91.

Burdge, G. C. and Calder, P. C. (2005). Conversion of α-linolenic acid to longer-chain polyunsaturated fatty acids in human adults. *Reprod. Nutr. Dev.* **45**, 581–597.

Burdge, G. C. and Wootton, S. A. (2002). Conversion of eicoapentaenoic, docosapentaenoic and docosahexaenoic acids in young women. *Br. J. Nutr.* **88**, 411–420.

Chiang, J. Y. L., Kimmel, R., and Stroup, D. (2001). Regulation of cholesterol 7 α-hydroxylase gene (CYP7A1) transcription by the liver orphan receptor (LXRα). *Gene* **262**, 257–265.

Constantopoulos, G. (1970). Lipid metabolism of manganese-deficient algae. *Plant Physiol.* **45**, 76–80.

Cordain, L., Eaton, S. B., Brand Miller, J., Mann, N., and Hill, K. (2002). The paradoxical nature of hunter-gatherer diets: Meat-based, yet non-atherogenic. *Eur. J. Clin. Nutr.* **56**, 542–552.

Cordain, L., Eaton, S. B., Sebastian, A., Mann, N., Lindeberg, S., Watkins, B. A., O´Keefe, J. H., and Brand-Miller, J. (2005). Origins and evolution of the Western diet: Health implications for the 21st century. *Am. J. Clin. Nutr.* **81**, 341–354.

Corton, J. C. H. and Anderson, S. P. (2000). Central role of peroxisome proliferator-activated receptors in the actions of peroxisome proliferators. *Annu. Rev. Pharmacol. Toxicol.* **40**, 491–518.

Dawczynski, C. H., Schubert, R., and Jahreis, G. (2007). Amino acids, fatty acids, and dietary fibre in edible seaweed products. *Food Chem.* **103**, 891–899.

de Deckere, E. A. M., Korver, O., Verschuren, P. M., and Katan, M. B. (1998). Health aspects of fish and n-3 polyunsaturated fatty acids from plant and marine origin. *Eur. J. Clin. Nutr.* **52**, 749–753.

Fan, K. W., Jiang, Y., Faan, Y. W., and Chen, F. (2007). Lipid characterization of mangrove thraustochytrid—*Schizochytrium mangrovei*. *J. Agric. Food Chem.* **55**, 2906–2910.

Flachs, P., Horakova, O., Brauner, P., Rossmeisl, M., Pecina, P., Franssen-van Hal, N., Ruzickova, J., Sponarova, J., Drahota, Z., Vlcek, C., Keijer, J., Houstek, J., *et al*. (2005). Polyunsaturated fatty acids of marine origin upregulate mitochondrial biogenesis and induce β-oxidation in white fat. *Diabetologia* **48**, 2365–2375.

Floreto, E. A. T. and Teshima, S. (1998). The fatty acid composition of seaweeds exposed to different levels of light intensity and salinity. *Bot. Mar.* **41**, 467–484.

Gerasimenko, N. I., Busarova, N. G., and Moiseenko, O. P. (2010). Seasonal changes in the content of lipids, fatty acids, and pigments in brown alga *Costaria costata*. *Russ. J. Plant Physiol.* **57**, 205–211.

Gómez Candela, C., Bermejo López, L. M., and Loria Kohen, V. (2011). Importance of a balanced omega 6/omega 3 ratio for the maintenance of health. Nutritional recommendations. *Nutr. Hosp.* **26**, 323–329.

Goyens, P. L. L., Spilker, M. E., Zock, P. L., Katan, M. B., and Mensink, R. P. (2006). Conversion of α-linolenic acid in humans is influenced by the absolute amounts of α-linolenic acid and linoleic acid in the diet and not by their ratio. *Am. J. Clin. Nutr.* **84**, 44–53.

Grønn, M., Christensen, E., Hagve, T. A., and Christophersen, B. O. (1991). Peroxisomal retroconversion of docosahexaenoic acid (22:6(n-3)) to eicosapentaenoic acid (20:5(n-3)) studied in isolated rat liver cells. *Biochim. Biophys. Acta* **1081**, 85–91.

Hertting, G. and Seregi, A. (1989). Formation and function of eicosanoids in the central nervous system. *Ann. N. Y. Acad. Sci.* **559**, 84–89.

Hibbeln, J. R., Nieminen, L. R. G., Blasbalg, T. L., Riggs, J. A., and Lands, W. E. M. (2006). Healthy intakes of n-3 and n-6 fatty acis: Estimations considering worldwide diversity. *Am. J. Clin. Nutr.* **83**, 1483–1493.

Honya, M., Kinoshita, T., Ishikawa, M., Mori, H., and Nisizawa, K. (1994). Seasonal variation in the lipid content of cultured *Laminaria japonica*: Fatty acids, sterols, β-carotene and tocopherol. *J. Appl. Phycol.* **6**, 25–29.

Hu, F. B., Manson, J. A. E., and Willett, W. C. (2001). Types of dietary fat and risk of coronary heart disease: A critical review. *J. Am. Coll. Nutr.* **20**, 5–19.

Hulshof, K. F. A. M., van Erp-Baart, M. A., Anttolainen, M., Becker, W., Church, S. M., Couet, C., Hermann-Kunz, E., Kesteloot, H., Leth, T., Martins, I., Moreiras, O., Moschandreas, J., *et al*. (1999). Intake of fatty acids in Western Europe with emphasis on trans fatty acids: The TRANSFAIR study. *Eur. J. Clin. Nutr.* **53**, 143–157.

Kamlangdee, N. and Fan, K. W. (2003). Polyunsaturated fatty acids production by *Schizochytrium* sp. isolated from mangrove. *Songklanakarin J. Sci. Technol.* **25**, 643–650.

Khotimchenko, S. V. (1998). Fatty acids of brown algae from the Russian Far East. *Phytochemistry* **49**, 2363–2369.

Kim, M. K., Dubacq, J. P., Thomas, J. C., and Giraud, G. (1996). Seasonal variations of triacylglycerols and fatty acids in *Fucus serratus*. *Phytochemistry* **43**, 49–55.

Kinsella, J. E., Lokesh, B., and Stone, R. A. (1990). Dietary n-3 polyunsaturated fatty acids and amelioration of cardiovascular disease: Possible mechanisms. *Am. J. Clin. Nutr.* **52**, 1–28.

Kobayashi, N., Barnard, R. J., Henning, S. M., Elashoff, D., Reddy, S. T., Cohen, P., Leung, P., Hong-Gonzales, J., Freedland, S. J., Said, J., Gui, D., Seeram, N. P., *et al*. (2006). Effect of altering dietary ω-6/ω-3 fatty acid ratios on prostate cancer membrane composition, cyclooxygenase-2, and prostaglandin E_2. *Clin. Cancer Res.* **12**, 4662–4670.

Kris-Etherton, P. M., Taylor, D. S., Yu-Poth, S., Huth, P., Moriarty, K., Fishell, V., Hargrove, R. L., Zhao, G., and Etherton, T. D. (2000). Polyunsaturated fatty acids in the food chain in the United States. *Am. J. Clin. Nutr.* **71**, 179–188.

Kumari, P., Kumar, M., Gupta, V., Reddy, C. R. K., and Jha, B. (2010). Tropical marine macroalgae as potential sources of nutritionally important PUFAs. *Food Chem.* **120**, 749–757.

Leslie, J. B. and Watkins, W. D. (1985). Eicosanoids in the central nervous system. *J. Neurosurg.* **63**, 659–668.

Lewis, T. E., Nichols, P. D., and McMeekin, T. A. (1999). The biotechnological potencial of traustochytrids. *Mar. Biotechnol.* **1**, 580–587.

Linko, Y. Y. and Hayakawa, K. (1996). Docosahexaenoic acid: A valuable nutraceutical? *Trends Food Sci. Technol.* **7**, 59–63.

Martinez, M., Ichaso, N., Setien, F., Durany, N., Qiu, X., and Rosler, W. (2010). The Δ4-desaturation pathway for DHA biosynthesis is operative in the human species: Differences between normal controls and children with Zellweger syndrome. *Lipids Health Dis.* **9**, 1–10.

Mendes, A., da Silva, T. L., and Reis, A. (2007). DHA concentration and purification from the marine heterotrophic microalga *Crypthecodinium cohnii* CCMP 316 by winterization and urea complexation. *Food Technol. Biotechnol.* **45**, 37–44.

Mendes, A., Reis, A., Vasconcelos, R., Guerra, P., and da Silva, T. L. (2009). *Crypthecodinium cohnii* with emphasis on DHA production: A review. *J. Appl. Phycol.* **21**, 199–214.

Meyer, B. J., Mann, N. J., Lewis, J. L., Milligan, G. C., Sinclair, A. J., and Howe, P. R. C. (2003). Dietary intakes and food sources of omega-6 and omega-3 polyunsaturated fatty acids. *Lipids* **38**, 391–398.

Mishra, V. K., Temelli, F., Ooraikul, B., Shacklock, P. F., and Craigie, J. S. (1993). Lipids of the Red Alga, *Palmaria palmata*. *Bot. Mar.* **36**, 169–174.

Miyake, Y., Sasaki, S., Tanaka, K., Fukushima, W., Kiyohara, Ch., Tsuboi, Y., Yamada, T., Oeda, T., Miki, T., Kawamura, N., Sakae, N., Fukuyama, H., *et al*. (2010). Dietary fat intake and risk of Parkinson´s disease: A case-control study in Japan. *J. Neurol. Sci.* **288**, 117–122.

Narayan, B., Miyashita, K., and Hosakawa, M. (2006). Physiological effects of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA)—A review. *Food Rev. Int.* **22**, 291–307.

Olivieri, O., Bassi, A., Stranieri, Ch., Trabetti, E., Martinelli, N., Pizzolo, F., Girelli, D., Friso, S., Pignatti, P. F., and Corrocher, R. (2003). Apolipoprotein C-III, metabolic syndrome, and risk of coronary artery disease. *J. Lipid Res.* **44**, 2374–2381.

Qiu, X., Hong, H., and MacKenzie, S. L. (2001). Identification of a Δ4 fatty acid desaturase from *Thraustochytrium* sp. involved in the biosynthesis of docosahexanoic acid by heterologous expression in *Saccharomyces cerevisiae* and *Brasica juncea*. *J. Biol. Chem.* **34**, 31561–31566.

Rose, D. P. (1996). The mechanistic rationale in support of dietary cancer prevention. *Prev. Med.* **25**, 34–37.

Rose, D. P. (1997). Dietary fatty acids and cancer. *Am. J. Clin. Nutr.* **66**, 998S–1003S.
Rubio-Rodríguez, N., Beltrán, S., Jaime, I., de Diego, S. M., Sanz, M. T., and Carballido, J. R. (2010). Production of omega-3 polyunsaturated fatty acid concentrates: A review. *Innovat. Food Sci. Emerg. Technol.* **11**, 1–12.
Saito, H., Xue, Ch., Yamashiro, R., Moromizato, S., and Itabashi, Y. (2010). High polyunsaturated fatty acid levels in two subtropical macroalgae, *Cladosiphon okamuras* and *Caulerpa lentillifera*. *J. Phycol.* **46**, 665–673.
Sánches-Machado, D. I., López-Cervantes, J., López-Hernández, J., and Paseiro-Losada, P. (2004). Fatty acids, total lipid, protein and ash contents of processed edible seaweeds. *Food Chem.* **85**, 439–444.
Simopoulos, A. P. (2002a). Omega-3 fatty acids in inflammation and autoimmune diseases. *J. Am. Coll. Nutr.* **21**, 495–505.
Simopoulos, A. P. (2002b). Omega-3 fatty acids in wild plants, nuts and seeds. *Asia Pac. J. Clin. Nutr.* **11**, S163–S173.
Simopoulos, A. P. (2004). Omega-3 fatty acids and antioxidants in edible wild plants. *Biol. Res.* **37**, 263–277.
Simopoulos, A. P. (2006). Evolutionary aspects of diet, the omega-6/omega-3 ratio and genetic variation: Nutritional implications for chronic diseases. *Biomed. Pharmacother.* **60**, 502–507.
Sugano, M. and Hirahara, F. (2000). Polyunsaturated fatty acids in the food chain in Japan. *Am. J. Clin. Nutr.* **71**, 189S–196S.
Terry, P. D., Rohan, T. E., and Wolk, A. (2003). Intakes of fish and marine fatty acids and the risks of cancers of the breast and prostate and of other hormone-related cancers: A review of the epidemiologic evidence. *Am. J. Nutr.* **77**, 532–544.
Terry, P. D., Terry, J. B., and Rohan, T. E. (2004). Long-chain (n-3) fatty acid intake and risk of cancers of the breast and the prostate: Recent epidemiological studies, biological mechanisms, and directions for future research. *J. Nutr.* **134**, 3412S–3420S.
Trombetta, A., Maggiora, M., Martinasso, G., Cotogni, P., Canuto, R. A., and Muzio, G. (2007). Arachidonic and docosahexaenoic acids reduce the growth of A549 human lung-tumor cells increasing lipid peroxidation and PPARs. *Chem. Biol. Interact.* **165**, 239–250.
Tvrzická, E., Staňková, B., Vecka, M., and Žák, A. (2009). Fatty acids 1. Occurrence and biological significance (In Czech). *Čas. Lék Čes.* **148**, 16–24.
Ward, O. P. and Singh, A. (2005). Omega-3/6 fatty acids: Alternative sources of production. *Process Biochem.* **40**, 3627–3652.
Weiss, L. A., Barrett-Connor, E., and von Mühlen, D. (2005). Ratio of n-6 to n-3 fatty acids and bone mineral density in older adults: The Rancho Bernardo Study. *Am. J. Clin. Nutr.* **81**, 934–938.
Yaguchi, T., Tanaka, S., Yokochi, T., Nakahara, T., and Higashihara, T. (1997). Production of high yields of docosahexaenoic acid by *Schizochytrium* sp. strain SR21. *J. Am. Oil Chem. Soc.* **74**, 1431–1434.
Yap, C. Y. and Chen, F. (2001). Polyunsaturated fatty acids: Biological significance, biosynthesis, and production by microalgae-like organisms. *In* ''Algae and Their Biotechnological Potential'', (F. Chen and Y. Jiang, Eds), 1-4020-0162-2, pp. 1–32. Kluver Academic Publishers, Dordrecht.
Yokoyama, R. and Honda, D. (2007). Taxonomic rearrangement of the genus *Schizochytrium* sensu lato based on morphology, chemotaxonomic characteristics, and 18S rRNA gene phylogeny (Thraustochytriaceae, Labyrinthulomycetes): Emendation for *Schizochytrium* and erection of *Aurantiochytrium* and *Oblongichytrium* gen. nov. *Mycoscience* **48**, 199–211.
Yongmanitchai, W. and Ward, O. P. (1993). Positional distribution of fatty acids, and molecular species of polar lipids, in the diatom *Phaeodactylum tricornutum*. *J. Gen. Microbiol.* **139**, 465–472.