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BIOAVAILABILITY OF OMEGA-3 PUFA FROM FOODS ENRICHED WITH FISH OIL – A MINI REVIEW

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This study presents a review of the research on the influence of omega-3 polyunsaturated fatty acids consumed in the form of foods enriched with fish oil on health. The study indicates that the bioavailability of omega-3 fatty acids from enriched foods is as powerful as that from fish oil capsule supplements. The presented results of investigations suggest that the production of fish oil-enriched food is thus reasonable and justified. However, polyunsaturated fatty acids are highly susceptible to oxidation and additional research should be undertaken to prove the safety of food products enriched with fish oil used in human nutritional products. In addition, special conditions during production, packaging and storage must be applied to prevent the oxidation of omega-3 fatty acids, otherwise enriched food products may easily become a source of hydroperoxides and other by-products of oxidation, *e.g.* free radicals promoting cancer and atherosclerosis development.

FOOD ENRICHMENT WITH FISH OIL

The growing presence on the European market of food products enriched with fish oil as a source of polyunsaturated fatty acids (PUFA) omega-3 in the diet requires evaluation and comment. Fish oil is the main dietary source of long-chain omega-3 PUFA. It was established that long--chain omega-3 PUFA: eicosapentaenoic acid (EPA C20:5) and docosahexaenoic acid (DHA C22:6), exert beneficial effects on human health, especially due to the prevention of cardiovascular diseases (CVD), much more effective than in the case of the shorter-chain alpha linolenic acid (ALA C18:3) [Simopoulos *et al.*, 1999; Ruxton *et al.*, 2003; Stephensen, 2004]. However, the intake of long-chain omega-3 PUFA in many developed countries (average 0.15 g per day) is below the recommended level [Thautwein, 2001; Kolanowski *et al.*, 2004]. Dietary recommendations suggest that the consumption of omega-3 PUFA should be increased. The European Academy of Nutritional Sciences (EANS) and UK dietary guidelines recommend intakes of an average of 0.2 g of EPA plus DHA a day [deDeckere *et al.*, 1998; Ruxton *et al.*, 2003]. Recommendations of the International Society for the Study of Fatty Acids and Lipids (ISSFAL) suggest the adequate intake of omega-3 fatty acids to be 0.65 g of DHA plus EPA per day (0.22 g per day of each as a minimum). The ratio of omega-6 to omega-3 in the diet should be 4 to 1 [Simopoulos *et al.*, 1999]. In 2000, FDA stated that the daily intake of EPA and DHA should not exceed 3.0 g per person per day in the form of fish oil, from food and dietary supplements [FDA, 2000]. Due to the low fish consumption in developed societies, it seems reasonable to introduce several food products enriched with fish oil that can be an additional source of the desirable long-chain omega-3 PUFA in the diet.

Currently an increase in the available number of various food products enriched with omega-3 fatty acids by fish oil addition as well as fish oil capsule supplements has been observed on the market. This kind of food is often classified as the so-called functional food. The production flow of food and pharmaceutical grade fish oil is presented in Figure 1. The development of fish oil-enriched food products

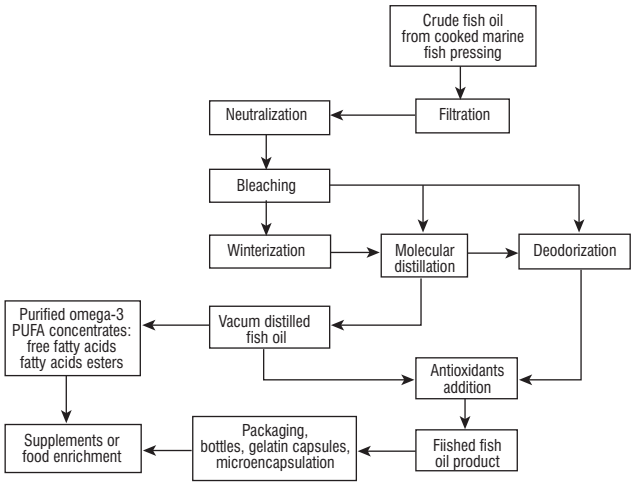

FIGURE 1. Production flow of food and pharmaceutical grade fish oil (own drawing based on Bimbo [1998]).

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as well as other functional foods must be based on the scientific knowledge of the target function in the body and show that the effects are relevant for improved health or reduction of disease risk. However, safety, bioavailability and the effects on health caused by the consumption of such enriched food products still remain to be evaluated.

The physiological effects of the intake of omega-3 fatty acids added to foods may differ depending on the quality of the fish oil used, the type of a product and different nutrients present in the enriched foods. Several studies have been performed to evaluate the potential health benefits caused by the consumption of foods enriched with fish oil. Some examples from current studies suggest that different amounts of long-chain omega-3 – DHA and EPA, given in capsules or added to food products, result in the same effects on the change in the blood lipid profile. Unfortunately, there is still a shortage of published experimental data which might explain and prove the beneficial influence of omega-3 PUFA-enriched food intake on human health. It should also be emphasized that animal studies provide only supportive, but not conclusive evidence. Results from such studies cannot be transferred to human conditions because of the differences in metabolism pathways between humans and animals. Consequently, studies involving humans are of essential importance.

RESEARCH ON HUMANS

Research exploring health influence of the consumption of fish oil enriched foods started in the early 1990s. Roche and Gibney [1994] investigated the influence of the intake of spreadable fats enriched with fish oil on platelet phospholipids fatty acid composition in humans during 12 weeks of testing with 33 male subjects randomly allocated into 3 groups. Each group consumed respectively 50.0 g per day of the following spreads: normal spread (control group), low fat spread and butter, both blended with 80.0 g/kg of fish oil. There was no change in the control group, whereas in groups receiving fish oil-enriched spreads, the baseline levels of platelet phospholipids omega-3 PUFA concentration increased significantly: EPA from 8.0 to 15.0 mg/kg and DHA from 23.0 to 29.0 mg/kg in the group receiving enriched low fat spread, and in the group receiving enriched butter, EPA 6.0 to 24.0 mg/kg and DHA from 23.0 to 31.0 mg/kg. The evaluation using a rating method showed that both enriched spreads were sensory acceptable. The authors concluded that in individuals with low fish intake, spreadable fats enriched with fish oil may be a useful vehicle for ensuring the adequate intake of omega-3 PUFA [Roche & Gibney, 1994]. Similarly, Sorensen *et al.* [1998] tested the effects of fish oil-enriched spreadable fat intake on the LDL-cholesterol plasma level in 47 healthy volunteers. The results showed that a 4-week consumption of enriched spread had no effect on LDL and HDL plasma concentration and increased the concentration of EPA and DHA in the LDL fraction by 3% and 7%, respectively.

The study conducted by Gustaffsonn *et al.* [1996] investigated the effects of the consumption of seafood products additionally enriched with omega-3 fatty acids. This was a double-blind, cross-over study, conducted over a 3-week period with 23 subjects with moderately high blood lipid levels. The diets including the omega-3-enriched seafood provided 3.0 g of omega-3 PUFA per day, compared with 0.3 g per day in the control group. Results showed a significant 25% reduction in plasma triacylglycerol levels, a significant 3.7% fall in systolic blood pressure and a 19% reduction in insulin secretion in the volunteers who consumed the fortified diet compared to the control diet. To match the amounts of omega-3 fatty acids provided in the test diet of the study (3.0 g per day), volunteers would have been required to consume at least 10 portions of oil-rich fish per week. It was concluded that in the context of a Western diet, these amounts were only obtainable through food supplementation [Gustaffsonn *et al.*, 1996].

Similarly, Engstrom *et al.* [2003] evaluated the effects of the consumption of caviar paste enriched with a high concentration of omega-3 PUFA. The study was a randomized, double blind, repeated measures experiment conducted over 3 weeks. In total, 16 healthy, non-smoking subjects were included in the study. Eight consumed 25 g of ordinary caviar paste daily for 3 weeks, and eight the same amount of caviar paste enriched with a very stable fish oil (70.0 g/kg). Blood lipids, plasma phospholipid fatty acids and lipid peroxidation were measured. EPA and DHA, as well as the sum of all omega-3 PUFA, increased significantly in both caviar groups, but the growth was higher in the group given fish oil caviar paste (an increase in EPA and DHA: 51% and 100%, respectively). Lipid peroxidation, measured as the thiobarbituric acid malondialdehyde adduct, was increased by 26% after the intake of ordinary caviar paste, but remained unchanged after the intake of fish oil-enriched caviar paste. The authors concluded that caviar paste can be incorporated in the diet to achieve an increase in omega-3 PUFA blood lipids. However, changing to caviar paste enriched with well stabilized fish oil led to a considerably greater increase in EPA and DHA and the consequent protection against oxidation [Ergstom *et al.*, 2003].

Lovegrove *et al.* [1997] studied the effect of foods enriched with fish oil on both fasting and postprandial lipaemia in subjects with normal blood lipid levels. The following food items were used: bread, biscuits, cake, ice-cream, orange drink, low-fat spread, pasta, mayonnaise, vinaigrette, and milkshakes. All products were enriched by incorporating fish oil either directly or in a microencapsulated form. During the 22-day intervention period, the average daily intake of DHA plus EPA in the test diet was 1.4 g per day, compared to 0.4 g per day in the control diet. The volunteers had free access to all the enriched foods until the 1.8 g per day limit was reached, and fish consumption was not allowed during the study period. Surprisingly, in spite of the amounts of omega-3 PUFA provided, there was no reduction in fasting or postprandial blood triacylglycerols, cholesterol or insulin levels, although there was a significant 16% increase in plasma values of HDL-cholesterol in the supplemented group compared to the control group. While these results showed that none of the tested foods produced a desirable reduction in the plasma concentration of triacyloglycerols, the authors suggested that the duration of the study was too short for them to be able to obtain the expected effect [Lovegrove *et al.*, 1997].

Saldeen *et al.* [1998] investigated the effects of a small dose of stable fish oil as a substitute for margarine on bread on plasma phospholipid fatty acids and serum triacylglycerols in a parallel, single-blinded, randomized study in 17 healthy subjects for 4 weeks. The enriched bread contained 3.0 g/kg of omega-3 PUFA. They found that the daily intake of 1.0 g of fish oil (containing 380.0 g/kg of omega-3 PUFA) in the form of enriched bread increased the concentration of EPA and DHA plasma phospholipids by 50% and decreased serum triacylglycerols by 17%. In addition, in the blinded consumer sensory test only 2 out of 195 subjects perceived any fishy taste in the enriched bread [Saldeen *et al.*, 1998]. Results obtained by Newton and Syndler [1993] or Liu *et al.* [2001] and Yep *et al.* [2002] also showed that the consumption of bread enriched with microencapsulated fish oil resulted in increased plasma DHA acid and total omega-3 PUFA, and decreased triacylglycerol plasma level.

Another alternative to fortifying foods with omega-3 PUFA is the addition of fish meal or seed oils (flax or rapeseed) to animal feeds, which resulted in meat and eggs enrichment with omega-3 [Grune *et al.*, 2001; Lopez-Ferrer *et al.*, 2001]. Several controlled studies using eggs enriched with omega-3 fatty acids have been carried out in healthy volunteers. The study by Ferrier *et al.* [1995] showed that regular consumption of the enriched eggs did not produce adverse effects on blood cholesterol levels produced by normal eggs. In other studies, however, the increase in plasma levels of long-chain omega-3 fatty acids derived from egg consumption did not correlate with any decrease in plasma concentration of triacylglycerols [Farrell, 1998; Surai *et al.*, 2000]. Moreover, Cherian and Sim [1996] demonstrated that the consumption of omega-3 fatty acids-enriched eggs resulted in a significant increase in the concentration of omega-3 PUFA in the milk of breast-feeding mothers, without any further effect in blood lipids and with important beneficial effects for breastfed children [Cherian & Sim, 1996]. Along with poultry and eggs, pig meat may be enriched with omega-3 PUFA. Some studies were performed on the effect of dietary omega-3 PUFA administration on fatty acid composition and sensory characteristics of pig meat and lard. Results showed possibilities of a nutritional improvement in the composition of pig meat [Leskanich *et al.*, 1997].

The study of Mantzioris *et al.* [2000] examined the effectiveness of a diet incorporating foods rich in omega-3 fatty acids in elevating the tissue concentration of DHA, EPA and ALA and in suppressing the production of inflammatory mediators. Volunteers had free access to oily fish and a number of omega-3-enriched food items (cooking oil, margarine, salad dressing, mayonnaise, sausages, onion dip) to obtain an average daily intake of 1.8 g per day of DHA plus EPA and 9.0 g per day of ALA. During the 4-week intervention period, the synthesis of thromboxane A2, prostaglandin E2 and interleukin l beta decreased by 36%, 26% and 20%, respectively. In this study, no indications of the effect of the omega-3-enriched diet on lipid plasma values were given [Mantzioris *et al.*, 2000].

In the study of Visioli *et al.* [2000], semi-skimmed milk containing omega-3 fatty acids from fish oil intake was tested in a controlled trial. In this study, volunteers consumed omega-3-enriched milk as the only source of omega-3 fatty acids for 6 weeks, supplying 0.3 g of DHA plus EPA daily. Results at the end of the dietary period showed an expected increase in plasma concentrations of DHA plus EPA (30% on average) together with a significant 19% reduction in fasting plasma triacylglycerols levels. Although levels of total cholesterol remained unchanged throughout the study, a 19% increase in HDL-cholesterol concentration was found [Visioli *et al.*, 2000]. In the study by Cobiac *et al.* [1991] a similar effect on the reduction of plasma triacylglycerols and HDL-cholesterol increase was only reached over a 5-week period of daily administration of encapsulated 4.5 g DHA plus EPA (4.5 g compared to 0.3 g in the study of Visioli *et al.* [2000]) in mildly hyperlipidemic males [Cobiac *et al.*, 1991]. This example indicates that the food matrix and other nutrients present in the food may positively affect the bioavailability of omega-3 PUFA and therefore, their physiological effects.

Baró *et al.* [2003] prepared a semi-skimmed milk enriched with long-chain omega-3 fatty acids, oleic acid, vitamins E and folic acid, and studied the effects of the consumption of this product on the risk factors for CVD in healthy, young subjects. The amounts of DHA and EPA in 500.0 mL of the omega-3-enriched milk were 0.13 g and 0.2 g respectively, whereas the control semi-skimmed milk contained no detectable levels of DHA and EPA. Results showed that consumption of 500.0 mL of the omega-3--enriched milk for 2 months produced a significant average 30% increase in plasma levels of DHA and EPA. Milk consumption also produced a decrease in the total plasma concentration (6%) and LDL-cholesterol (16%), accompanied by a reduction in plasma levels of homocysteine (13%) – all well known risk factors for CVD. In this study, the authors showed that small doses of omega-3 fatty acids might also produce desirable health effects [Baró *et al.*, 2003]. Milk fat is highly dispersed in micelles and is known to be very efficiently absorbed in the gut. The use of milk as a vehicle for the administration of omega-3 PUFA may be responsible for the relatively high levels of EPA and DHA found in plasma and the health benefits derived from regular consumption. However, other studies showed that milk enrichment strongly decreased sensory acceptance of milk and only a small amount of fish oil (up to 1 g/kg) may be added to milk in natural form [Kolanowski *et al.*, 1999].

Metcalf *et al.* [2003] evaluated the effects of providing a wide range of foodstuffs containing omega-3 PUFA, occurring naturally or from fortification, on the intake and blood and tissue proportions of ALA, EPA and DHA. Subjects were provided with a range of foods containing natural omega-3 PUFA (fresh fish, canned fish, flaxseed meal, rapeseed oil) and items fortified with fish oil (spreadable fat, milk, sausages, luncheon meat, onion dip). Food choices were left to the discretion of each subject. Intake was noted in a diet diary. The consumption of omega-3 PUFA increased significantly: ALA from 1.4 to 4.1 g per day, EPA from 0.03 to 0.51 g per day, and DHA from 0.09 to 1.01 g per day. Omega-6 linoleic acid intake decreased from 13.1 to 9.2 g per day. The authors concluded that incorporation of fish oil into a range of commercial foods provided the opportunity for wider public consumption of omega-3 PUFA with its associated health benefits [Metcalf *et al.*, 2003].

Higgins *et al.* [1999] evaluated the bioavailability of omega-3 PUFA from the intake of microencapsulated fish oil-enriched food compared with a fish oil capsules supplement. Twenty eight healthy subjects took part in the randomized, controlled trial and were supplemented with 0.9 g of omega-3 PUFA per day for 4 weeks, delivered either as microencapsulated fish oil in a milkshake or as a fish oil capsules. Plasma fatty acids composition and plasma total cholesterol levels were measured at baseline and after supplementation. Plasma omega-3 PUFA concentrations were raised significantly at a similar level by both fish oil supplements. Plasma total cholesterol levels were not altered by supplementation in either group. The results of this study indicated that there was no significant difference in the bioavailability and health effect of omega-3 PUFA given as enriched milkshakes in comparison to omega-3 PUFA delivered as capsule supplements [Higgins *et al.*, 1999].

Similarly, Wallace *et al.* [2000] evaluated the bioavailability of omega-3 PUFA from microencapsulated fish oil-enriched foods (bread, biscuits and soup) compared with equal amounts of omega-3 PUFA contained in fish oil capsules. Twenty five healthy female volunteers were randomly assigned to one of two groups for the 4-week investigation. Both groups received 0.9 g per day of omega-3 PUFA, one group in the form of fish oil capsules, the other – with enriched foods. The composition of platelet fatty acids was measured. EPA and DHA in platelet increased considerably in both groups, but there was no significant difference between the two groups following the investigation. The results of this study showed that omega-3 PUFA from enriched foods were as bioavailable as from capsule supplements [Wallace *et al.*, 2000].

Kew *et al.* [2003] evaluated the effects of enriching the diet with a spread containing ALA or EPA and DHA on immune results representing the key functions of human neutrophils, monocytes, and lymphocytes. In a placebo-controlled, double-blind, parallel study, 150 healthy men and women were randomly assigned to 1 of 5 interventions: placebo (no additional omega-3 PUFA), 4.5 or 9.5 g ALA per day and 0.77 or 1.7 g EPA plus DHA per day for 6 months. The omega-3 PUFA were provided in 25 g fat spread plus 3 fish oil capsules. PUFA composition regarding peripheral blood mononuclear cell phospholipids was significantly different in the groups with the higher intakes of ALA or EPA and DHA. Results showed that an intake of up to 9.5 g ALA or up to 1.7 g EPA and DHA per day did not alter the functional activity of neutrophils, monocytes, or lymphocytes, but it changed the fatty acid composition of mononuclear cells. The authors concluded that there was no effect of foods enriched with plant- or marine-derived omega-3 PUFA on human immune function [Kew *et al.*, 2002].

FINAL REMARKS

The results of investigations on the use of food products enriched with fish oil, as an easy delivery system of long-chain omega-3 PUFA into the human organism, might encourage efforts towards the production of such enriched foods. It is well known that an increase in long-chain omega-3 PUFA intake with these enriched foods can have a desirable effect on human health, especially by decreasing the risk of the so-called life-style diseases, particularly CVD. In the currently available published studies the bioavailability of omega-3 PUFA from enriched food intake was demonstrated to be comparable with the bioavailability of the acids from capsule supplements. In conclusion and with reference to the presented data, it can be stated that the consumption of food enriched with unhydrogenated fish oil as a source of long-chain omega-3 PUFA significantly improves the level and profile of PUFA in the diet and in human body tissues. However, the current data do not fully justify the production of fish oil-enriched foods, particularly because of the high susceptibility of fish oil to rapid oxidation. For potential producers of such enriched foods special types of packaging and storage conditions are strongly advised, especially ensuring the elimination of contact with the air. It is advisable to store fish oil-enriched food products for short periods of time, preferably packaged in one-portion units eliminating all factors promoting the oxidation of PUFA. Otherwise the desirable health value of omega-3 fatty acids may very easily transform into highly toxic peroxides and other by-products of oxidation, which promote harmful effects, especially the development of cancer [Grune *et al.*, 2001; Grundt *et al.*, 2003]. Special attention should be paid in the case of powdered forms (microencapsulated) of fish oil preparations, where the surface is enormously developed. However, it is expected that frequent consumption of properly prepared food products enriched with well refined, stabilised, food grade fish oil might increase the amount of long-chain omega-3 PUFA in the diet, thus significantly improving its nutritional quality, which might result in better protection against cardiovascular and many other life-style related diseases. However, due to the high susceptibility of fish oil to oxidation, additional research is needed which will provide adequate measures for the evaluation of the safety of fish oil-enriched food during production, storage and consumption.

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BIODOSTĘPNOŚĆ WIELONIENASYCONYCH KWASÓW OMEGA-3 Z ŻYWNOŚCI WZBOGACONEJ OLEJEM RYBIM – MINI ARTYKUŁ PRZEGLĄDOWY

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W pracy przedstawiono przegląd wyników badań eksperymentalnych prowadzonych na ludziach dotyczących wpływu zdrowotnego wielonienasyconych kwasów tłuszczowych omega-3 spożywanych z żywnością wzbogaconą olejem rybim. Z przedstawionych wyników można wnioskować, że biodostępność kwasów omega-3 ze wzbogaconej żywności jest równie wysoka jak z suplementów diety i preparatów leczniczych. Obserwacje te potwierdzają zasadność produkcji żywności wzbogacanej olejem rybim. Jednak wielonienasycone kwasy tłuszczowe są wysoce podatne na zmiany oksydacyjne i wymagane są dodatkowe badania w pełni potwierdzające bezpieczeństwo zdrowotne tak wzbogaconych produktów. Brak odpowiednich zabiegów technologicznych i pakowania hamującego zmiany oksydacyjne prowadzi do szybkiej przemiany prozdrowotnych kwasów omega-3 w rakotwórcze nadtlenki, a wzbogacona w te kwasy żywność może stać się niebezpieczna dla zdrowia konsumenta.