Polar Lipids—Phospholipids and Glycolipids—An Enhanced Omega-3 Structure

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OMEGA-3 HEALTH BENEFITS

Omega-3 fatty acids are beneficial fats required by every cell in the body to function optimally. Thousands of studies support the health benefits of omega-3 fatty acids, particularly the long-chain omega-3s EPA (eicosapentaenoic acid) and DHA (docosahexaenoic acid).

- **Cardiovascular health:** Omega-3 fatty acids work through multiple modes to support cardiovascular health. Specifically, research shows these fatty acids help maintain triglyceride and blood pressure levels already within a healthy range, support the heart's natural rhythm and pumping ability, improve cardiac performance, and support a healthy inflammation response. Even the conservative FDA has acknowledged that "Supportive but not conclusive research shows that consumption of EPA and DHA omega-3 fatty acids may reduce the risk of coronary heart disease."
- **Brain health:** Because EPA and DHA are incorporated into cell membranes, providing both durability and flexibility, they support intracellular communication—which is particularly important for brain function. Omega-3s have been found to support infant brain development, promote a positive mood, support memory and cognition, and promote neuronal health.
- **Eye health:** Nowhere in the body is DHA more concentrated than the retina of the eye, which points toward the importance of omega-3s to healthy vision. Indeed, researchers have discovered that omega-3s support infant visual development, preserve healthy vision in aging adults, and promote the normal tearing function of the eyes.
- **Joint health:** Several studies have found that omega-3s support joint comfort and flexibility, most likely by converting into powerful inhibitory substances called resolvins.

The American Heart Association states that "Large-scale epidemiologic studies suggest that people at risk for coronary heart disease benefit from consuming omega-3 fatty acids from marine and plant sources." The organization recommends that:

- Patients with documented CHD: Consume about 1 g of EPA+DHA per day, preferably from fatty fish.
- Patients concerned with triglycerides: 2 to 4 g of EPA+DHA per day provided as capsules under a physician's care.

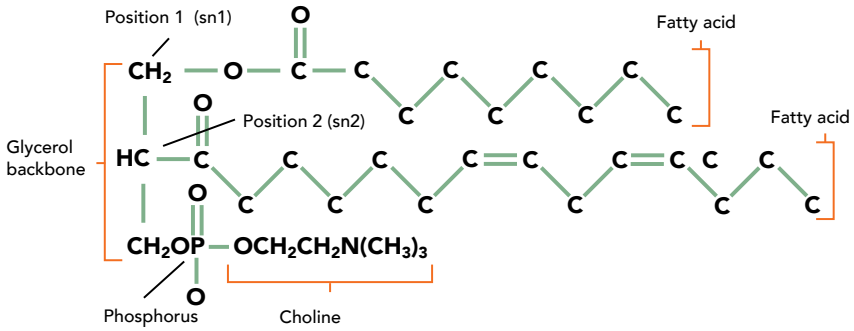
WHAT ARE POLAR LIPID OMEGA-3S?

Omega-3 fatty acids are polyunsaturated fats (PUFAs) that are crucial for maintaining and improving cellular health. PUFAs contain more than one double bond that is "unsaturated" with hydrogen atoms. Long chain (LC) PUFAs, including EPA, DHA and n-3 DPA, have more double bonds than short chain PUFAs, and are necessary for the body to perform a wide range of vital functions. Polar lipid omega-3s are a special type of omega-3 with a unique structure that provides many benefits. Following is more information on different lipid structures of omega-3s and how polar lipids are digested and absorbed into the body.

CLASSES OF LIPIDS

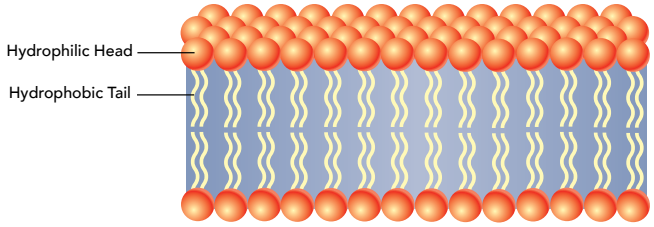
Omega-3s LC PUFAs are components of lipids, which are typically organized by the type of fat and the type of molecule they are esterified (bound) to. This compositional difference is significant because it dictates how these fats are incorporated in tissues and used by the body.

- **Polar (PL)**: Polar lipids can hold two fatty acid molecules, bound to a glycerol or sphingosine backbone, which combine with either a phosphorus (phospholipid) or sugar (glycolipid) molecule to form the "head."



PL structural formula: A glycerol or sphingosine backbone binds to either a phosphorus or sugar molecule. In this example, a glycerol backbone is bound to both a phosphorus and a choline molecule, forming the common polar lipid phosphatidylcholine.

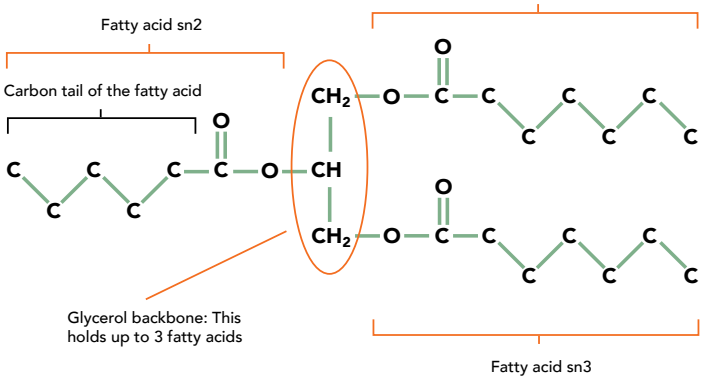
Both phospholipid and glycolipid polar lipids are amphipathic; that is, they have a hydrophilic (water-loving) head and a hydrophobic (water-fearing) tail that help them self-orient to form a double layer in which the polar end points outward and the nonpolar end points inward. This lipid bilayer forms the protective, permeable membrane of cells and is critical to cell function. Phospholipids form the cell bilayer in humans and animals and glycolipids form the cell bilayer in plants.



Polar lipids amphipathic properties form a lipid bilayer, which forms the protective permeable membrane of cells

Polar lipids are easily digested and incorporated into tissues in mammals. In aqueous environments such as the stomach and intestines, the hydrophilic heads turn out to face the water and the hydrophobic tails turn inward, forming a micelle structure. The micelle structure can deliver oils, such as omega-3 fatty acids, into cells without the gastrointestinal distress, such as fishy burps, that can occur with other lipids.

- **Triacylglycerol (TAG):** This lipid molecule can hold three fatty acids that are esterified to a glycerol backbone. As a large, complex molecule, TAG requires the assistance of both bile salts and multiple pancreatic enzymes to remove the fatty acids, making them Free Fatty Acids (FFAs), from the glycerol backbone prior to absorption and use. TAG lipids are abundant in most fish oils. TAG fatty acids are well-digested, absorbed, and transported to key tissues for storage and metabolism in mammals.



TAG structural formula: A glycerol backbone that binds up to three fatty acids

- **Ethyl ester (EE):** This omega-3 structure is synthetic (man-made) and found in highly concentrated fish oils. To concentrate the omega-3s, the glycerol backbone is replaced with an ethanol backbone. Ethyl ester forms are not considered "natural" by many and studies have generally shown poorer bioavailability than TAG or polar lipid forms of omega-3s[1].

- **Free fatty acids:** After omega-3s have been cleaved from the backbone by the digestive process, they function as free fatty acids, which are available to be absorbed and used by the body.

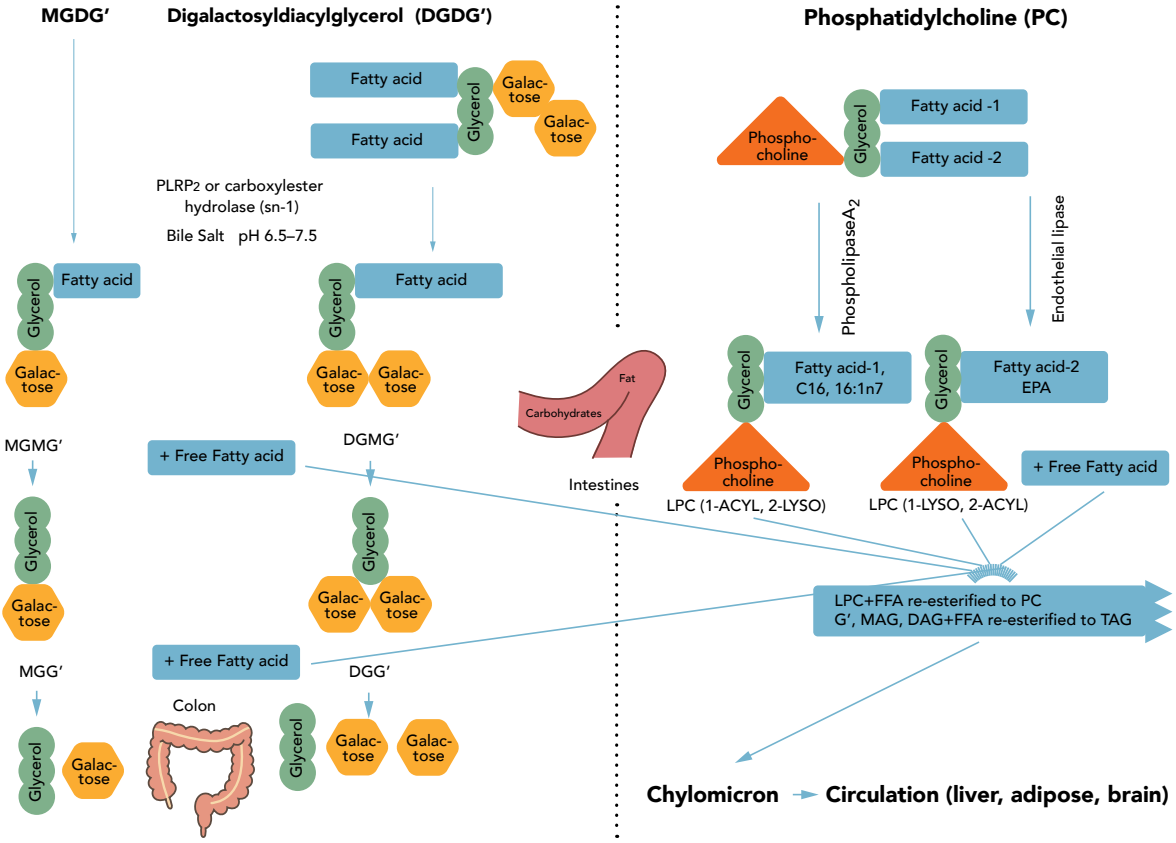
STRUCTURE AND DIGESTION OF POLAR LIPIDS

Almega PL™ is a unique omega-3 from the non-GMO microalgae *Nannochloropsis oculata* containing ≥15% polar lipids, including phospholipids and glycolipids. **It is the only omega-3 ingredient available that contains both phospholipids and glycolipids.**

The phospholipid content of Almega PL is around 5%, with phosphatidylcholine (PC) constituting the most abundant phospholipid. PC has been shown to support brain health, healthy cell function, healthy liver function and intestinal health in multiple studies.

The glycolipid content of Almega PL™ totals roughly 10%, as both monogalactosyldiacylglycerols (MGDG) and digalactosyldiacylglycerols (DGDG). MGDG and DGDG are the most abundant lipids in nature[2] and are important components of plant leaves and chloroplast membranes[3]. MGDG and DGDG contain one or two molecules of galactose (a monosaccharide), respectively, attached to the common neutral lipid, diacylglycerol (DAG). Glycolipids are plant membrane lipids that can substitute as the phosphate head in a phospholipid, conserving the phosphate for other essential processes.

Absorption of dietary phospholipids and glycolipids



M=Mono; D=Di; G=Galactosyl; L=Lyso; PLRP2=Pancreatic lipase-related protein 2, galactolipase, G'=Glycerol

Digestion of phospholipids

Almega PL is a natural source of EPA, which is bound to phospholipids on either the sn-1 or sn-2 position on a glycerol backbone. During transit through the intestine, pancreatic enzymes separate the fatty acids at either connection point to form molecules with different bioactive properties.

In one instance, intestinal phospholipase A2 (PLA_2) cleaves a single EPA fatty acid from the sn-2 position on the backbone, generating a 1-acyl 2-lyso phospholipid and a corresponding free fatty acid of EPA. The free fatty acid is then available to be absorbed into tissues. Almega PL also contains palmitate (16:0) and bioactive palmitoleate (16:1n7) components, which remain intact following PLA_2 cleavage. The lysoPL generated during digestion likely facilitates micelle formation in the intestinal lumen, further increasing bioavailability of these dietary lipids and possibly playing a role in intestinal signaling.

Importantly, recent studies demonstrate that the sn-2 position of phospholipids may also be acted upon by an intestinal endothelial lipase (also known as carboxyl ester hydrolase or carboxy ester lipase) with PLA_1 activity[3,4]. This reaction generates a 1-lyso 2-acyl phospholipid molecule that is highly bioavailable and bio-accretible for uptake by brain and other tissues[5,6]. In this instance, the EPA on the sn-2 position of phosphatidylcholine (PC) in Almega PL remains with the starting source (rather than being released as a free fatty acid); this may contribute to the enhanced bioavailability of phospholipid-bound EPA.

Digestion of glycolipids

MGDG and DGDG glycolipids are digested in the gastrointestinal tract by a galactolipase enzyme known as pancreatic lipase-related protein 2 ($PLRP_2$)[7]. The enzyme has surface loops surrounding the active site exposing a large hydrophobic surface. This larger surface area (referred to as a "lid") increases acyl chain binding of the glycolipid and stabilization of acyl intermediates[8]. The lid interacts with colipase[9] and is particularly important for binding the longer chain acyl subtrates present in Almega PL. Hydrolysis of glycolipids is dependent on bile salt, with an optimum pH of 6.5–7.5[10,11].

Model systems show that human recombinant $PLRP_2$ is able to digest glycolipids such as MGDG with similar magnitude as TAG lipase digestion[12]. Recombinant human carboxyl ester hydrolase (acting on the sn-1 position) also digested MGDG (at $\frac{1}{10}$ the activity of recombinant $PLRP_2$)[12] and DGDG and SQDG[10]. Taken together, the fact that multiple enzymes digest various types of glycolipids efficiently implies that humans are well adapted to glycolipid digestion.

Model systems also suggest that $PLRP_2$ hydrolyzes fatty acids at the sn-1 position, generating mono and di-galactosylated monoglycerides (MGMG and DGMG) and free fatty acids (which can then be absorbed and used) in the gut[7].

BENEFITS OF POLAR LIPIDS

The unique structure of polar lipids is more easily digested and absorbed than TAG lipids, and research shows that Almega PL's unique phospholipid and glycolipid structure provides superior EPA absorption and other health-promoting features. These features are translated to clear consumer benefits, especially smaller, easy-to-swallow capsules without fishy burps.

Polar lipids improve bioavailability of omega-3s

The polar structure allows for easier digestion and absorption of omega-3 fatty acids. Many studies have shown that krill oil and other polar lipid omega-3s are better absorbed into the bloodstream and consequently into tissues. This improved efficacy allows for a smaller dosage to increase in *invivo* omega-3 levels (blood plasma fatty acids, red-blood cell phospholipids and fatty acid levels in tissues).

Phospholipids and glycolipids may reduce burping

Burping (fishy burps) is a key reason consumers avoid consuming fish oil capsules. Enteric coating of fish oils is a means to avoid burping, but can bypass or perturb normal fat digestion processes. Burping is reduced when phospholipids (and potentially glycolipids) are consumed because fats become better emulsified in the gut. This is particularly important in the fasted state when bile salts are not actively being released and recirculated enterohepatically (i.e. from the liver to the intestine and back to the liver again). Almega PL may reduce burping in a natural way that does not rely on enteric coatings.

Phospholipids and glycolipids may increase digestibility of other lipids

Dietary lipids are primarily consumed as TAG. Phospholipids may increase the bioavailability of EPA and DHA (and other fatty acids)

A LOOK AT OTHER OMEGA ISSUES

The pioneering studies by Dyerberg in the 1970s[20] reveal most omega-3 dietary sources have come from marine animals, mainly fish and recently krill. However, there are two significant issues with omega-3s sourced from fish and krill—sustainability and availability of a vegetarian source.

Sustainability

Demand for omega-3s for nutrition and pharmaceutical products is rising. GOED, the Global Organization for EPA and DHA, estimates that the demand for omega-3s far outstrips the potential supply available from marine animal sources. In addition, humans have not been very adept at managing marine natural resources. More than 70% of the omega-3 supply comes from a single fishery—the Peruvian anchoveta. Thus, much of the supply of fish oil-based omega-3s depends on the health and longevity of this one fishery. A different source of omega-3s, krill, also raises sustainability questions. Krill harvesting occurs mainly in remote Antarctica, and krill are at the base of the food chain, as the source of food for many other marine animals such as whales, seals and fish. As demand for krill rises, it raises a question how increased harvesting might affect this fragile eco-system.

Vegetarian source

Vegetarianism and veganism are significant and growing consumer trends. Most vegetarians and vegans do not consume enough health-promoting omega-3s. This is mainly because there are very few sources of LC-PUFA omega-3s that are suitable for vegetarians. Fish, krill and other marine animals are not acceptable, while flax, chia, and canola (rapeseed) oils only provide the shorter-chain alpha-linolenic acid (ALA) omega-3. The short-chain ALA does not have the same health benefits that are associated with the longer-chain EPA and DHA omega-3s.

Almega PL provides both sustainable and vegetarian LC-PUFA omega-3s

Almega PL provides an environmentally sustainable and vegetarian alternative source for omega-3s. This EPA-rich, polar-lipid omega-3 is sourced from sustainable algae. The production of Almega PL does not affect complex marine ecosystems and has sustainability built into the manufacturing process. The algae are grown in ponds on a Texas farm, using sunlight as the main energy input; as well as utilizing non-fertile desert land and non-potable brackish water. The hexane-free extraction process employs a patented "wet-extraction" technology that saves energy by reducing a drying step.

present as TAG. This is expected, since phospholipids are already established to increase digestibility of the fatty acids in TAG, probably by promoting chylomicron production[13] (droplets of fat in the blood or lymph after being absorbed from the small intestine). Furthermore, emulsified TAG sources of EPA and DHA are generally more bioavailable than non-emulsified forms of EPA and DHA[14-16], and phospholipids are good emulsifiers.

Phospholipids and glycolipids may increase digestibility of fat soluble vitamins

Some fat-soluble vitamins and other nutraceuticals are poorly absorbed. Dietary phospholipids and glycolipids are known to increase the digestibility of fat-soluble vitamins such as carotene, lycopene, alpha-tocopherol[17] and lutein[18]. This ability to increase digestibility is particularly important when fats are consumed in the fasted state[19]. In mice, glycolipids increased the in vitro micellization of lutein relative to neutral lipids, probably by reducing the micellar size[18]. Both glycolipids and phospholipids increased the rate at which lutein appeared in the plasma relative to neutral lipids. Phospholipids increased lutein levels in the liver, whereas glycolipids increased lutein levels in the eye more than neutral lipids.

Glycolipids may inhibit COX-2 and help modulate inflammation

Multiple published studies indicate the role of glycolipids in supporting a healthy inflammation response.

- MGDG enriched with omega-3 fatty acids was applied to adult avian articular cartilage cells following an inflammatory insult[21]. Treatment with MGDG prior to the insult suppressed expression of inflammation-induced proteins (lipocalin extracellular fatty acid binding protein, avidin and serum amyloid A).
- In later in vitro experiments[22], MGDG from the same source was shown to support healthy inflammation activity in human articular cartilage by activating an inflammation modulation loop triggered by COX-2 synthesis of prostaglandin production. This indicates a possible role of MGDG in inflammation resolution.
- MGDG and DGDG inhibited croton-oil-induced ear edema in mice in a dose-dependent manner[23]. Inhibition by MGDG was greater than that of the reference treatment and was largely abrogated following acyl group saturation, indicating the PUFAs linked to MGDG are important for bioactivity. In the mouse carrageenan-induced paw edema model, inhibitory effects were also dose-dependent.
- The inflammation modulation potential of MGDGs was tested[24]. Both compounds inhibited nitric oxide (NO) production in macrophage cells. This indicates inflammation modulation, since controlling NO production by inducible nitric oxide synthase (iNOS) in macrophages is important for supporting bacterial balance, tissue integrity, endothelial function, organ function, and healthy cell division.

Taken together, these studies indicate that Almega PL™ glycolipids have a protective role in cases of acute and recurring inflammation.

ANIMAL AND CLINICAL TRIALS WITH ALMEGA PL™

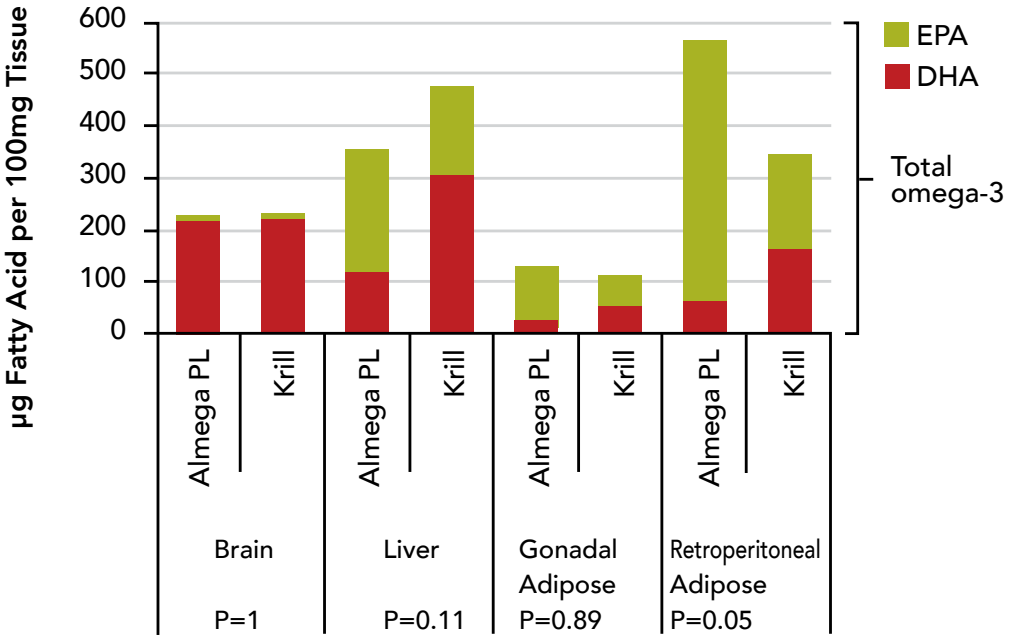
Almega PL versus krill oil tissue uptake study in rats[25]

Study parameters

	Krill Oil	Almega PL
Feeding period	7 days	7 days
Omega-3s dosage	7.24 g of combined EPA+DHA (4.8 g EPA, 2.4 g DHA)	7.3 g EPA
Polar lipid content	40% phospholipids	15% polar lipids (phospholipid/ glycolipid combination)

Study results: For most of the tissues, the uptake of total omega-3s was similar between Almega PL and krill oil. Levels of EPA were statistically significantly higher in retroperitoneal adipose tissue in the Almega PL group as compared to the krill oil group[25]. Although the amounts of EPA provided were higher in the algal group, these results are important because when comparing equi-doses of algal and krill oil, the former will lead to higher plasma and adipose levels. Adipose tissue may serve as a storage form of DHA and possibly EPA for delivery to the brain[27].

Distribution of EPA and DHA into tissues

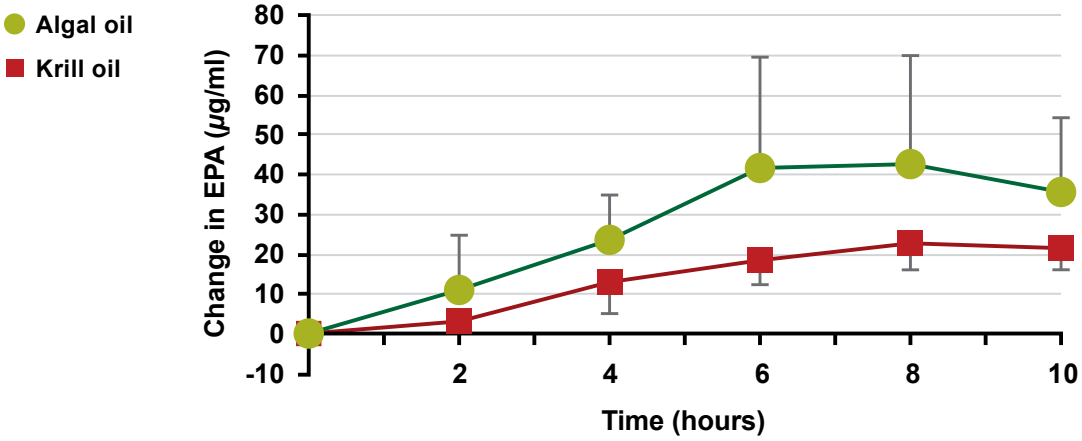


ALMEGA PL™ VERSUS KRILL OIL: ACUTE PLASMA BIOAVAILABILITY STUDY IN HUMANS

Study parameters: In a crossover study, the bioavailability of a single dose of AlmegaPL was compared to krill oil in 10 healthy males aged 18-45 years, with plasma fatty acid levels monitored for 0.5-10 hours[28]. Subjects consumed a standard high fat (55 g) breakfast followed by either algal oil (providing 1.5 g of omega-3: all EPA and no DHA) or krill oil (providing 1.56 g of omega-3: 1.02 g EPA and 0.54 g DHA).

Study results: Total omega-3 absorption into blood plasma was similar between Almega PL and krill oil, without a statistically significant difference. However, when looking only at EPA, the plasma concentration of EPA was higher with algal oil than with krill oil at all time points. The maximum concentration of EPA was also higher with algal oil and both the area under the concentration curve (AUC) and the incremental AUC (IAUC) for EPA were greater and highly statistically significant with Almega PL (p=0.006). Even when taking into account the different dosages of EPA, this increase was disproportionally high for Almega PL (dosage of 50% more EPA; but 100% higher increase in EPA). These results suggest that the combination of phospholipids plus glycolipids in Almega PL may be a more effective carrier for EPA/omega-3s than the phospholipids in krill.

Plasma EPA absorption



CONCLUSION

Long chain omega-3 PUFAs are important dietary constituents in maintaining good health. The lipid structure of the PUFA plays a significant role in how effectively omega-3s are delivered to the body. Fatty acids in Polar Lipid structures (both phospholipid and glycolipid PLs) provide desirable benefits—including easier digestion, enhanced bioavailability and uptake into tissues than other lipids. Almega PL™, a new omega-3 source from a natural algae strain, contains both phospholipids and glycolipids. Scientific studies have shown that the total omega-3 uptake into tissues and into plasma is equivalent between krill oil and EPA-rich Almega PL. **They have also shown that the EPA has a disproportionally higher uptake into plasma, which suggests that the combination of phospholipids plus glycolipids may be a more effective carrier for omega-3s than phospholipids alone.**

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