Minor Omega-3 and Omega-6 Polyunsaturated Fatty Acids in Krill Oil

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Polyunsaturated Fatty Acids

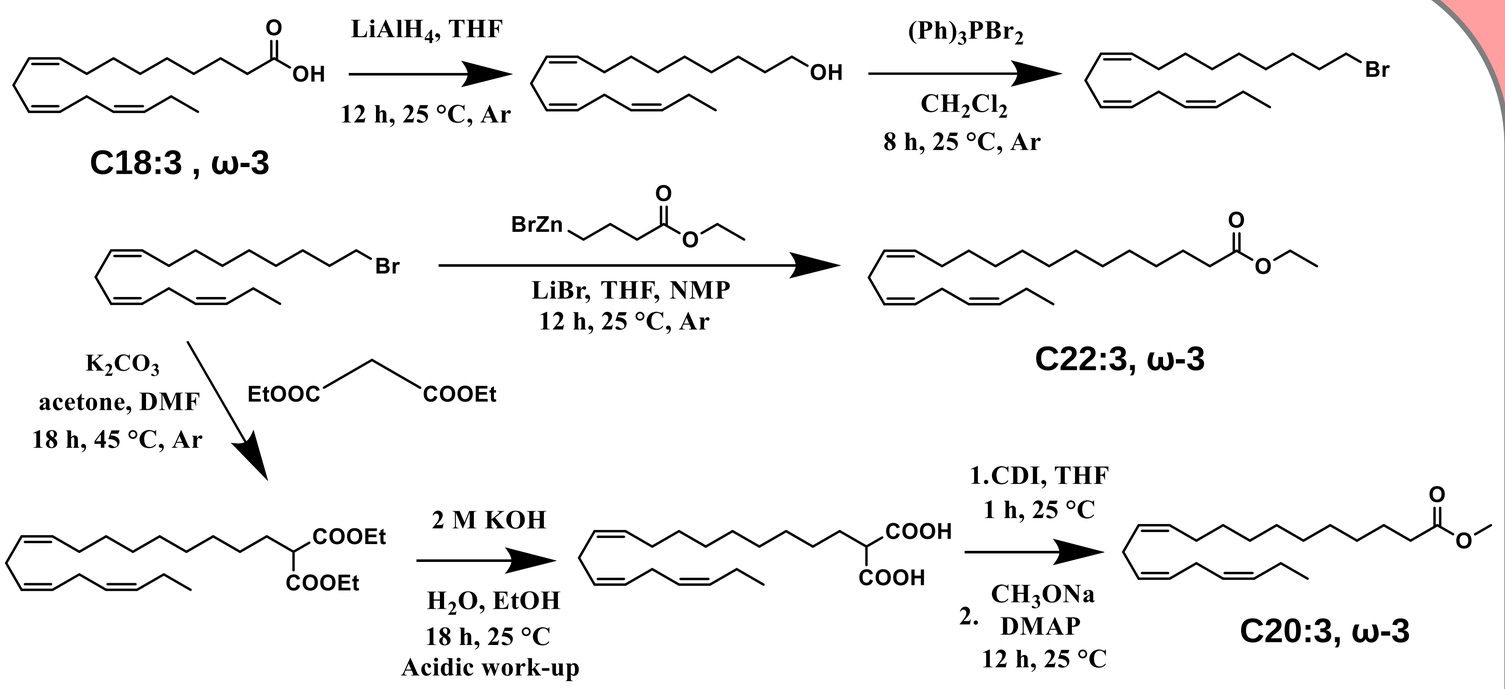
- Conditionally essential ω-3 and ω-6 polyunsaturated fatty acids (LC-PUFA) play a critical role in human nutrition and generally have beneficial health effects.
- The Western diet tends to have an unbalanced ratio of ω-6 / ω-3 fatty acids, and their intake is insufficient, which can promote the pathogenesis of some diseases. To prevent this problem, people often use dietary supplements.
- The most important supplement of both ω-3 and ω-6 series is considered fish oil. Krill oil has also been receiving attention recently.
- While current research focuses mainly on eicosapentaenoic (EPA), docosahexaenoic (DHA), and arachidonic (ARA) acids, minor ω-3 and ω-6 fatty acids such as *all-cis*-11,14,17-eicosatrienoic acid (C22:3 n-3) are neglected.

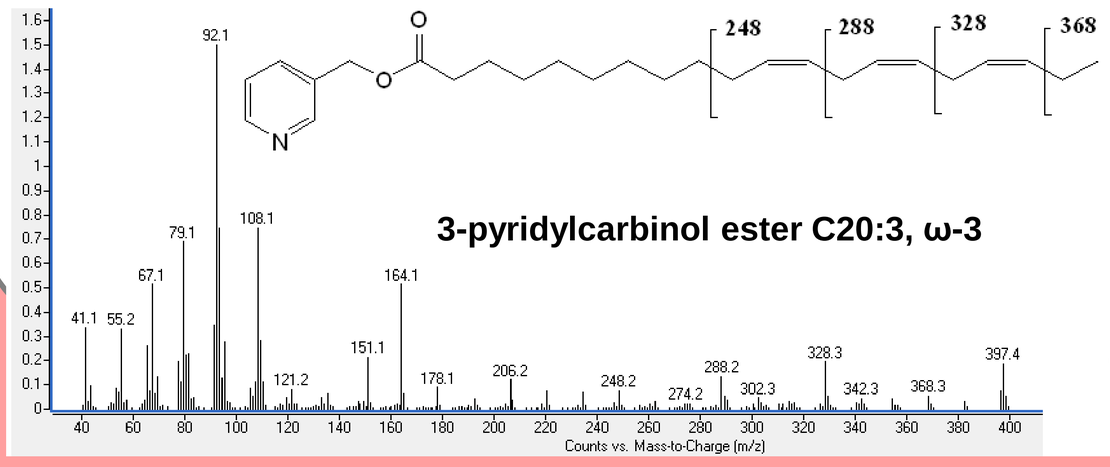
Krill oil

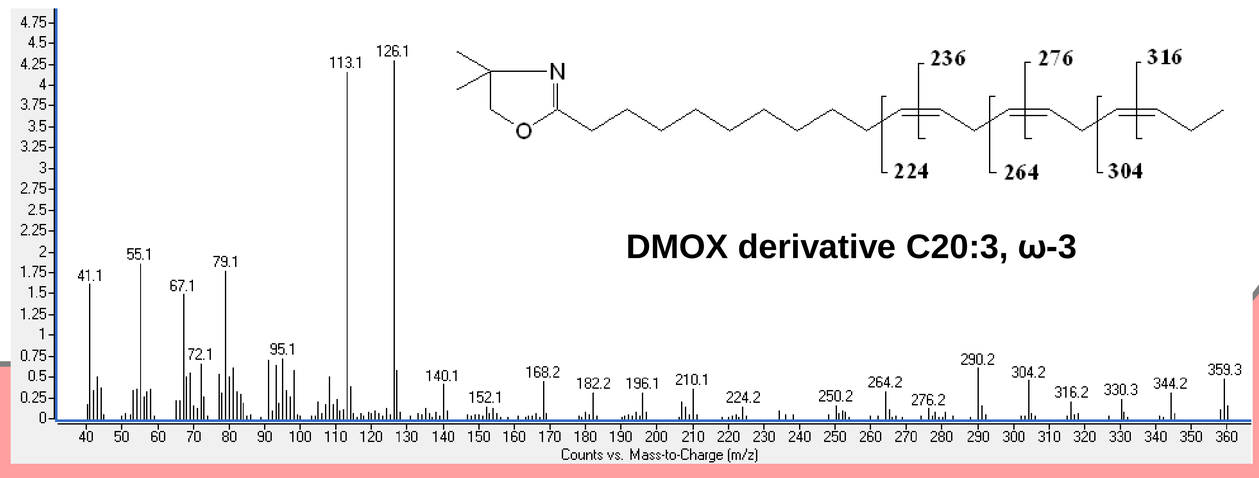
- Krill oil (KO) is extracted using polar solvents from the abundant Antarctic krill (*Euphausia superba*) native to the Southern Ocean around Antarctica. Like fish, krill consume a diet rich in ω-3 fatty acids and are, therefore, a natural source of EPA and DHA with their content similar to fish oil.
- However, unlike fish oil, EPA and DHA are incorporated into glycerophospholipids (mostly phosphatidylcholine) instead of triacylglycerols, which might contribute to enhanced absorption and better bioavailability.
- KO also contains potent antioxidant astaxanthin, a red carotenoid pigment, which may have possible health benefits.

Elongation of linoleic acid (C18:2, ω-6) and α-linolenic acid (C18:3, ω-3)

- Linoleic acid (C18:2, ω-6) and α-linolenic acid (C18:3, ω-3) were elongated by two and four carbon atoms to obtain *all-cis*-11,14-eicosanedienoic acid (C20:2, ω-6), *all-cis*-13,16-docosadienoic acid (C22:2, ω-6), *all-cis*-11,14,17-eicosatrienoic acid (C20:3, ω-3), and *all-cis*-13,16,19-docosatrienoic acid (C22:3, ω-3), which served as analytical standards.
- We confirmed the positions of *all-cis* double bonds in the synthesized fatty acids by converting them into 3-pyridylcarbinol esters and DMOX derivatives.
- Overall, three (C4) and five (C2) step elongations provided decent overall yields, and mild conditions minimalized isomerization/migration of the *all-cis* double bond system.

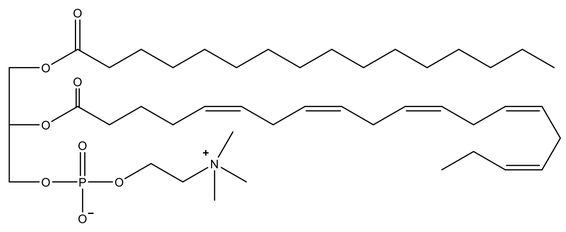






The distribution of fatty acyl moieties in the lipids of krill oil

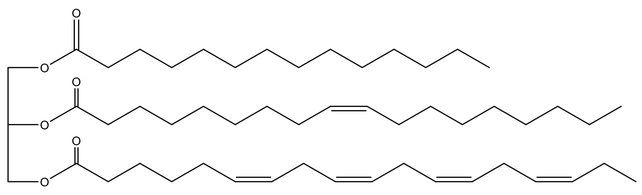
- A commercial sample of krill oil was fractionated on silica gel column chromatography. GPLs were the main lipid class of commercial krill oil, followed by TAGs. The fraction containing DAGs and MAGs was surprisingly large, while the content of the FFAs was low (< 2%), which indicates phospholipase activity since the fatty acid composition of GPLs with DAGs and MAGs fraction was nearly identical.



Glycerophospholipids (47.04 wt. %)

DAGs + MAGs (12.20 wt. %)

Sterol esters (0.66 wt. %)



Triacylglycerols (38.30 wt. %)

Free fatty acids (1.48 wt. %)

Hydrocarbons (0.33 wt. %)

- As can be seen in the table, the fatty acid composition of krill oil is quite unique, containing unusual fatty acids such as polyunsaturated C16 series, stearidonic acid (C18:4, ω-3), and phytanic acid (3,7,11,15-tetramethyl hexadecanoic acid).
- However, three out of four of our prepared fatty acids (*all-cis*-11,14-eicosanedienoic acid, *all-cis*-13,16-docosadienoic acid, and *all-cis*-13,16,19-docosatrienoic acid) were not detected. That might be expected from ω-6 fatty acids since their content is low (around 2%), but it is surprising for the ω-3 fatty acid. *All-cis*-11,14,17-eicosatrienoic acid was present across all dominant fractions, and its content was around 0,15%.

Fatty acids (wt. %)	GPLs	TAGs	DAGs MAGs	Sterol esters	Krill oil
C12:0	0.18	0.48	0.87	4.49	0.41
C14:0	2.73	17.29	5.96	5.22	8.83
C16:0	26.53	19.62	14.23	15.73	22.23
C16:1 ω-7	1.68	8.36	4.27	4.10	4.63
C16:2 ω-4	0.00	0.81	0.00	0.48	0.32
C16:3 ω-4	0.00	0.53	0.17	0.26	0.23
C16:4 ω-1	0.00	2.59	0.86	0.80	1.12
C18:0	1.38	1.43	0.91	1.58	1.34
C18:1 ω-7	7.26	8.10	7.07	8.33	7.57
C18:1 ω-9	8.55	15.58	9.53	8.46	11.41
C18:2 ω-6	1.50	1.30	1.44	1.73	1.42
C18:3 ω-3	0.00	0.00	0.23	0.00	0.03
C18:4 ω-3	2.75	8.50	5.13	3.65	5.29
C20:0	0.48	0.05	0.06	0.12	0.26
Phytanic acid	0.11	3.78	0.70	0.16	1.61
C20:1 ω-9	0.52	1.41	0.74	1.65	0.90
C20:3 ω-3	0.19	0.10	0.12	0.01	0.14
C20:4 ω-6	0.71	0.30	1.09	0.87	0.60
C20:5 ω-3	25.60	4.25	28.18	23.94	17.58
C22:6 ω-3	16.17	2.14	16.07	13.93	10.67
Other FAs	3.67	3.36	2.38	4.47	3.39
SFA´	33.28	44.17	29.08	23.70	36.31
MUFA´	19.81	35.31	24.67	22.83	26.26
PUFA´	46.92	20.52	46.25	53.47	37.43
ω-3	44.71	14.98	41.55	49.72	33.71
ω-6	2.21	1.61	2.60	2.52	2.01

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