

CO₂-extracted northern berries are rich sources of essential fatty acids and natural antioxidants

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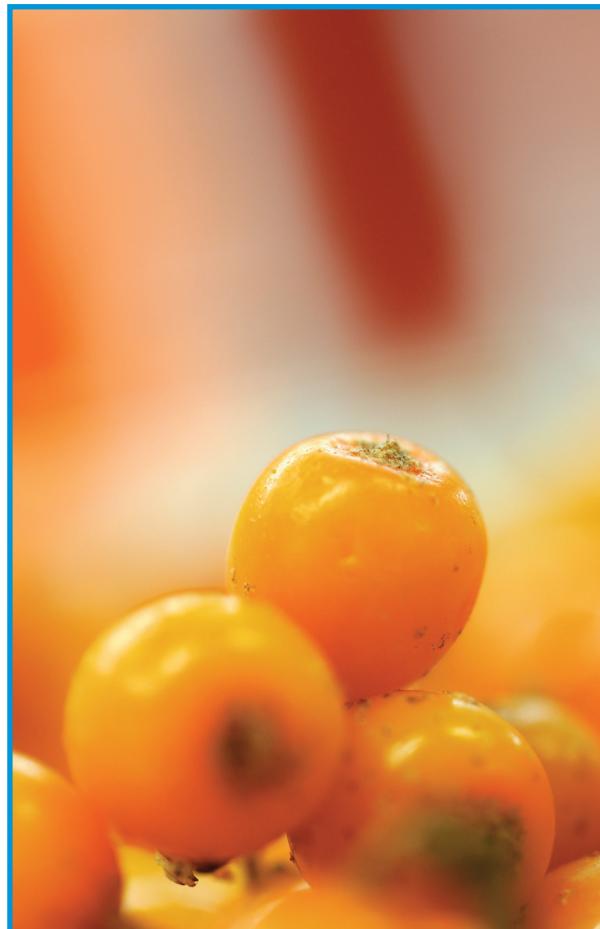
CO₂-extraction: A gentle method for recovery of lipophilic compounds

Northern berries are famous for their high content of hydrophilic bioactive compounds such as vitamin C, flavonoids, and phenolic acids. Less known is that they are also rich in lipophilic compounds with beneficial effects: fatty acids, carotenoids, tocopherols, tocotrienols and phytosterols. These are enriched in the berries' seeds and for the most part unavailable for absorption when the whole berry is consumed. Aromtech Ltd (Tornio, Finland) specialises in producing oils from Northern plants by means of supercritical CO₂ extraction, which enables gentle recovery of the berry and seed oils.

Supercritical CO₂ at high pressure and low temperature has the capacity to penetrate and elute materials efficiently. The low temperature applied, and the absence of oxygen, ensures that no bioactivity is lost. At the end of the extraction process, the CO₂ is released from the extract, producing a safe and solvent-free product without thermal stress. There is no need for refinement steps, which could affect the bioactive compounds. In comparison to the other methods of commercial oil production, organic solvent extraction (hexane) and cold pressing, higher amounts of bioactive compounds have been reported in CO₂-extracted oils¹.

Essential and unique fatty acids of Northern berries

The composition and antioxidant activity of Northern berry oils and berry seed oils extracted by supercritical CO₂ at Aromtech Ltd were recently studied by Yang et al.². The following oils were analysed: seed oils of bilberries, lingonberries, cranberries, Arctic cranberries, crowberries, cloudberry, sea buckthorn, raspberries, blackcurrants, redcurrants, snowball berries, European rowanberries, strawberries and cranberries. In sea buckthorn, unlike most other berries, also the soft part of the berries contain a high proportion of oil. The composition and activity of sea buckthorn pulp oil too were analyzed by Yang et al.².



Fresh sea buckthorn berries are first dried, seeds and soft parts are separated and the raw material for CO₂ extracted SBA24 oil is ready

All Northern berry seed oils investigated were good sources of essential linoleic (18:2n-6, LA) and α-linolenic (18:3n-3, ALA) acids. As a source of essential fatty acids, the lingonberry seed oil was exceptional. The combined proportion of LA and ALA was ~ 80 % of all fatty acids, of which the majority was ALA (~ 45 % of all fatty acids). The Western diet displays an imbalance in the intake

of n-3 vs n-6 fatty acids, with intake of n-6 fatty acids generally being higher than optimal when compared to n-3 intake³. The composition of lingonberry seed oil indicates that it is an excellent source of essential fatty acids and provides them in a ratio that would help to bring the balance closer to the recommended n-6:n-3-ratio of about 1:2:1³.

The fatty acid compositions of the currant seed oils are unique in the high content of γ-linolenic (18:3n-6, GLA) and stearidonic (18:4n-3, SDA) acids. Low GLA levels in the body have been associated with malfunction of the enzyme Δ-6-desaturase and atopic dermatitis⁴⁻⁷. When compared to redcurrant, CO₂-extracted blackcurrant seed oil is an even better source of GLA and SDA².

The LA and ALA content of sea buckthorn pulp oil is lower than that of the seed oil². The pulp oil, however, is characterized by its roughly 40% palmitoleic acid (16:1n-7), such amounts being rare in other food sources. Consumption of high-palmitoleic-acid-oils has been associated with beneficial effects on circulating total, LDL and HDL cholesterol^{8, 9}. Recently, studies suggesting lipokine effects and modulating effects of palmitoleic acid on insulin-resistance and hepatic lipid accumulation have been published^{10, 11}.

Natural tocopherols and tocotrienols and their association with antioxidant activity

Of the CO₂-extracted oils analysed in the study of Yang et al.², the raspberry seed oil stands out in having a high total amount of tocopherols and tocotrienols. The major tocol in the raspberry seed oil was γ-tocopherol. In comparison to α-tocopherol, its vitamin E activity is low, but depending on the conditions, it can be more potent as an antioxidant. The superior protective effects against nitrosative stress are particularly well documented¹². As a source of α-tocopherol, sea buckthorn pulp oil was the richest of the oils studied by Yang et al.². Berries of the *Vaccinium* genus (lingonberry, cranberry and Arctic cranberry) were the richest sources of tocotrienols, with γ-tocotrienol being the most abundant form. Tocotrienols have been shown to possess antioxidant, neuroprotective and cholesterol-lowering properties that differ from those exhibited by tocopherols¹³.

According to current knowledge, oxidative stress, the imbalance between cells' pro-oxidant and antioxidant systems contributes to the pathogenesis of several diseases, including cancer and cardiovascular diseases¹⁴. Yang et al.² investigated the antioxidant activity of the CO₂-extracted Northern berry seed oils by several distinct methods, to capture the different aspects of antioxidant action. In the peroxy radical scavenging assay, corresponding to lipophilic

oxygen radical absorbance capacity (lipophilic ORAC), there was a positive correlation between the oils' total tocopherols and tocotrienols and the ORAC value. Accordingly, of the oils investigated, raspberry oil showed the highest *in vitro* peroxy radical scavenging activity. In the assay designed to investigate the effects of oils on the peroxidation of membrane lipids of microsomes, no correlation between the total tocol content and antioxidant activity was found.

The antioxidant activity of sea buckthorn seed and pulp oils were investigated in greater depth². Both oils were found to have an *in vitro* inhibiting effect on the copper-induced LDL oxidation, they were found to be incorporated into the LDL particles in serum and scavenge the superoxide radicals. In the above-mentioned assays, sea buckthorn pulp oil was a more potent antioxidant than the seed oil was. This is probably because the two oils' different profiles of tocopherol isomers and the higher content of carotenoids in the sea buckthorn pulp oil².

In the *in vitro* DNA oxidation assays, both sea buckthorn seed and pulp oils prevented DNA oxidation in purified DNA and in rat liver homogenate DNA. There were no great differences between the two oils in terms of DNA protection potential².

Clinically proven beneficial effects

Physiological effects of CO₂-extracted oils produced by Aromtech Ltd have been investigated in several clinical trials. Aromtech's standardised sea buckthorn oil SBA24®, containing sea buckthorn seed and pulp oils, has clinically proven beneficial effects on mucous membranes¹⁵, dry eye¹⁶, skin¹⁷ and factors associated with cardiovascular disease^{18, 19} when taken as an oral supplement. The combination of seed and pulp oils offers a balance of bioactive compounds for good effects on the mucosa and cardiovascular health: the different fatty acids in seeds and pulp; the carotenoids of pulp oil; and the tocopherols, tocotrienols, and phytosterols of both sources.

Recently, the effects of CO₂-extracted blackcurrant seed oil on skin well-being were investigated in two studies with human volunteers. The effects on atopic dermatitis in children were investigated in a double-blind, randomised, placebo-controlled study at the University of Turku, in Finland²⁰. In total, 322 pregnant women were randomised for the study, and 177 mother-child pairs completed the trial. The pregnant women consumed blackcurrant seed oil or a placebo daily from eight to 16th week of pregnancy until the end of the exclusive breast-feeding period. After that, the infants received a supplement of blackcurrant seed oil or placebo oil until reaching the age of two years.

At one year of age, the prevalence of atopic dermatitis was significantly lower among children in the blackcurrant seed oil group than in the placebo oil group. A beneficial trend was



Intake of blackcurrant seed oil has beneficial effects on skin wellbeing

observed at two years. Atopic dermatitis is associated with inflammation, low activity of Δ6-desaturase, and reduced levels of serum GLA. It is likely that the effects of black currant seed oil were due to increased GLA, dihomo-γ-linolenic acid (20:3n-6, DGLA) and DGLA-derived eicosanoids complemented with the anti-inflammatory potential of SDA and ALA (20). In an open study with 40 participants, improvement of skin hydration through intake of blackcurrant seed oil was observed in healthy volunteers from 36 to 61 years of age²¹.

In summary, the Nordic berry oils and berry seed oils produced by the gentle method of supercritical CO₂ extraction have great potential in supplying the diet with essential fatty acids and antioxidants with beneficial physiological effects.

References

- Li TSC, Beveridge THJ, Droyer JCG. Phytosterol content of sea buckthorn (*Hippophae rhamnoides* L.) seed oil: Extraction and identification. *Food Chem.* 2007;101(4):1633-9.
- Yang B, Ahotupa M, Määttä P, Kallio H. Composition and antioxidative activities of supercritical CO₂-extracted oils from seeds and soft parts of northern berries. *Food Res Int.* 2011; 44:2009-2017
- Simopoulos AP. The importance of the ratio of omega-6/omega-3 essential fatty acids. *Biomed Pharmacother.* 2002;56(8):365-79.
- Schmitz G, Ecker J. The opposing effects of n-3 and n-6 fatty acids. *Prog Lipid Res.* 2008;47(2):147-55.
- Horrobin D. Fatty-acid metabolism in health and disease - the role of delta-6-desaturase. *Am J Clin Nutr.* 1993;57(5):S732-7.
- Laitinen K, Sallinen J, Linderborg K, Isolauri E. Serum, cheek cell and breast milk fatty acid compositions in infants with atopic and non-atopic eczema. *Clin Exp Allergy.* 2006;36(2):166-73.
- Guillou H, Zadravec D, Martin PGP, Jacobsson A. The key roles of elongases and desaturases in mammalian fatty acid metabolism: Insights from transgenic mice. *Prog Lipid Res.* 2010;49(2):186-99.
- Garg M, Blake R, Wills R. Macadamia nut consumption lowers plasma total and LDL cholesterol levels in hypercholesterolemic men. *J Nutr.* 2003;133(4):1060-3.
- Yang B, Kalimo KO, Mattila LM, Kallio SE, Katajisto JK, Peltola OJ, Kallio HP. Effects of dietary supplementation with sea buckthorn (*Hippophae rhamnoides*) seed and pulp oils on atopic dermatitis. *J Nutr Biochem.* 1999;10(11):622-30.
- Yang ZH, Miyahara H, Hatanaka A. Chronic administration of palmitoleic acid reduces insulin resistance and hepatic lipid accumulation in KK-ay mice with genetic type 2 diabetes. *Lipids Health Dis.* 2011;10:120.
- Cao H, Gerhold K, Mayers JR, Wiest MM, Watkins SM, Hotamisligil GS. Identification of a lipokine, a lipid hormone linking adipose tissue to systemic metabolism. *Cell.* 2008;134(6):933-44.
- Wagner KH, Kamal-Eldin A, Elmadafa I. Gamma-tocopherol—an underestimated vitamin? *Ann Nutr Metab.* 2004;48(3):169-88.
- Sen CK, Khanna S, Roy S. Tocotrienols: Vitamin E beyond tocopherols. *Life Sci.* 2006;78(18):2088-98.
- Thomas JA. Oxidant defense in oxidative and nitrosative stress. In: Shih ME, Shike M, Ross CA, Cabarello B, Cousins R, editors. *Modern Nutrition in Health and Disease.* 10th Edition ed. Philadelphia (PA): Lippincott Williams & Wilkins; 2006. p. 685-94.
- Yang B, Erkkola R. Sea buckthorn oils, mucous membranes and Sjögren's syndrome with special reference to latest studies. In: Singh et al. (eds.) 2006. *Seabuckthorn (*Hippophae L.*). A Multipurpose Wonder Plant. vol. III: Advances in research and development.* Dya Publishing House, New Delhi, India; 2006 p. 254-267.
- Larmo P, Järvinen R, Setälä N, Yang B, Viitanen M, Engblom J, Tahvonen R, Kallio H. Oral sea buckthorn oil attenuates tear film osmolarity and symptoms in individuals with dry eye. *J Nutr.* 2010;140:1462-8.
- Yang B, Bonfigli A, Pagani V, Isohanni T, von Knorring Å, Jutila A, Judin V-. Effects of oral supplementation and topical application of supercritical CO₂ extracted sea buckthorn oil on skin ageing of female subjects. *J Appl Cosmetol.* 2009;27:1-13.
- Johansson AK, Korte H, Yang B, Stanley JC, Kallio HP. Sea buckthorn berry oil inhibits platelet aggregation. *J Nutr Biochem.* 2000;11(10):491-5.
- Lehtonen H, Suomela J, Tahvonen R, Yang B, Venöjärvi M, Viikari J, Kallio H. Different berries and berry fractions have various but slightly positive effects on the associated variables of metabolic diseases on overweight and obese women. *Eur J Clin Nutr.* 2011;65(3):394-401.
- Linnamaa P, Savolainen J, Koulu L, Tuomasjukka S, Kallio H, Yang B, Vahlberg T, Tahvonen R. Blackcurrant seed oil for prevention of atopic dermatitis in newborns: A randomized, double-blind, placebo-controlled trial. *Clinical and Experimental Allergy.* 2010;40(8):1247-55.
- Larmo P, Rintala K, Ulinnen T, Judin V-. Intake of CO₂-extracted black currant seed oil improves skin hydration. *NutraCos Cosmetics.* 2011(3)

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