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4th International Blackcurrant Conference
Bialowieza, Poland
June 2nd - 6th, 2014

Composition and Health Properties of Blackcurrant; A Nordic Perspective

Heikki Kallio
Food Chemistry and Food Development
Depertment of Biochemistry
20014 University of Turku, Finland
heikki.kallio@utu.fi

Optional Strategies of Blackcurrant processing

herbal tea
dried spice
spice extract
herbal remedies

leaves
buds
branhces

berries

nectars
jams
jellies
sauces

pressing

waste

press residue

partially pressed berries

berry wine

juice

dried residue
food fiber
cutin for plastics
phenolic extracts
bakery products
extrudates
food color
lecithin

Various technologies

residues
toxins
foreign
materials

pre-treatment, stabization, refining, milling

CO_2 extraction of seed oil

seed oil

The Presentation Includes Examples of Analysis (1), Properties of Extracts (2) and Extrudates (3), and Effects of Seedoil (4)

spice extract (1)

leaves
buds
branhces

berries

pressing

nectars
jams
jellies
sauces

waste

press residue

partially pressed berries

berry wine

juice

phenolic extracts (2)

extrudates (3)

Various technologies

residues
toxins
foreign
materials

pre-treatment, stabization, refining, milling

(4)

seed oil

CO_2 extraction of seed oil

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(1) Analysis of Phenolic Compounds of BC (Leaves)

- Buds and leaves of three varieties
- ’Mikael’, ’Mortti’, No 15
- Phenolic compounds (HPLC-DAD-ESI-MS/MS)
- Contents defined , HPLC-DAD

→

- ’Mortti’ and No 15 were alike. ’Mikael’ was different.
- Flavonolglycosides the major phenolic group
- Tannins (proanthocyanidins) and phenolic acids less abundant

Liu, Kallio, Yang
Food Chemistry
2014, *160*, 180-189

(1)

Flavonol Glycosides Form the Most Abundant group

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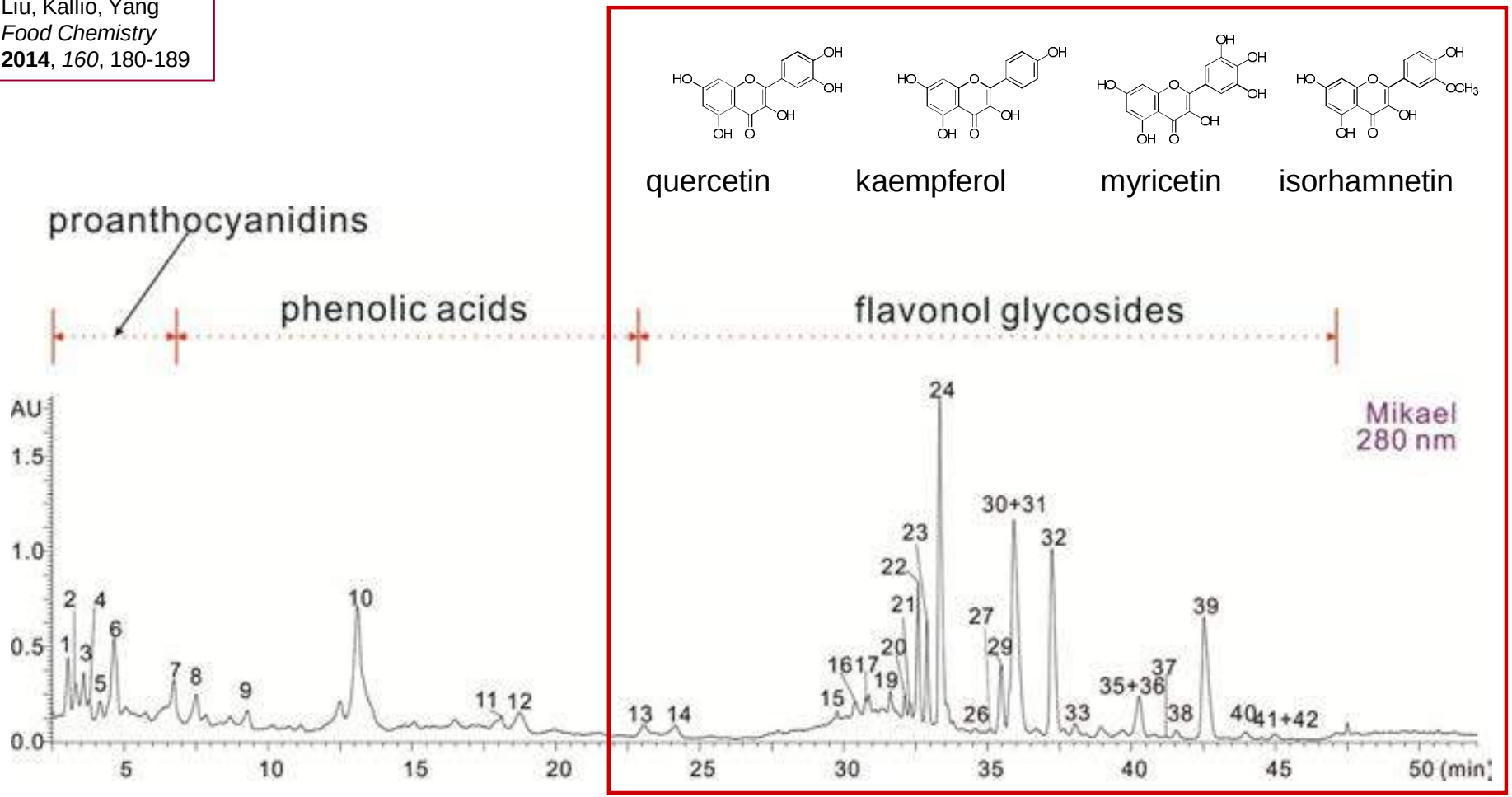

HPLC-DAD chromatogram of leaves (variety ‘Mikael’). 70 % H_2O-acetone extraction, measurd at 280 nm.

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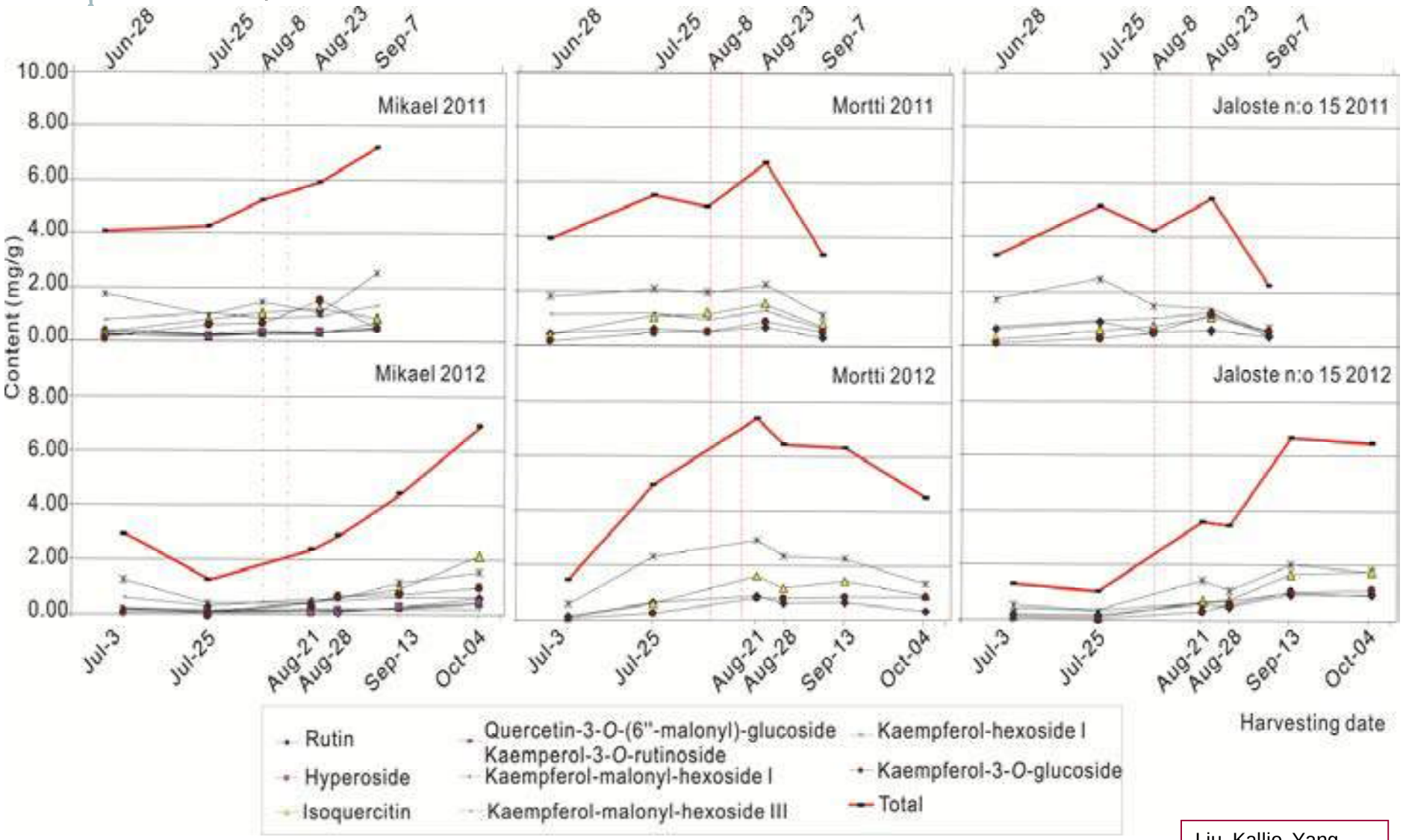

Changes in flavonolglycosides in BC leaves in 2011 and 2012.

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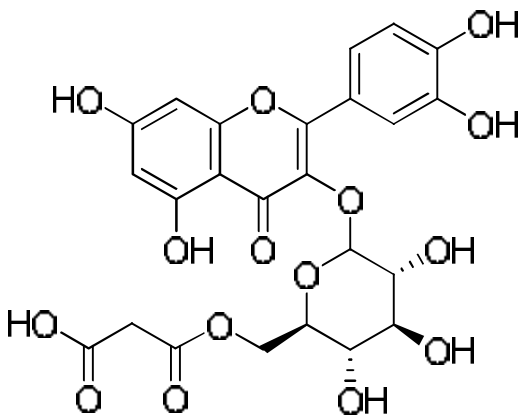
Quercetin-3-*O*-(6''-malonyl)-glucoside is a significant compound in BC



Quercetin-3-O-(6''-malonyl)-glucoside

- Qu-3-*O*-(6''-malon)-glc in BC leaves 120-230 mg/100 g fw
- High bioactivity
  - Known to lower blood glucose
  - Lowers risk of coronary clogging
  - Lowers oxidation of LDL
- Phenolic compounds have an effect on flavor and color
- Highest activities in leaves after berry harvesting!

The same methods of analysis to be applied in analysis of leaves, berries, food products, human tissues, blood, urine, feces etc.

→ Useful in food chemistry, technology, nutrition, medicine

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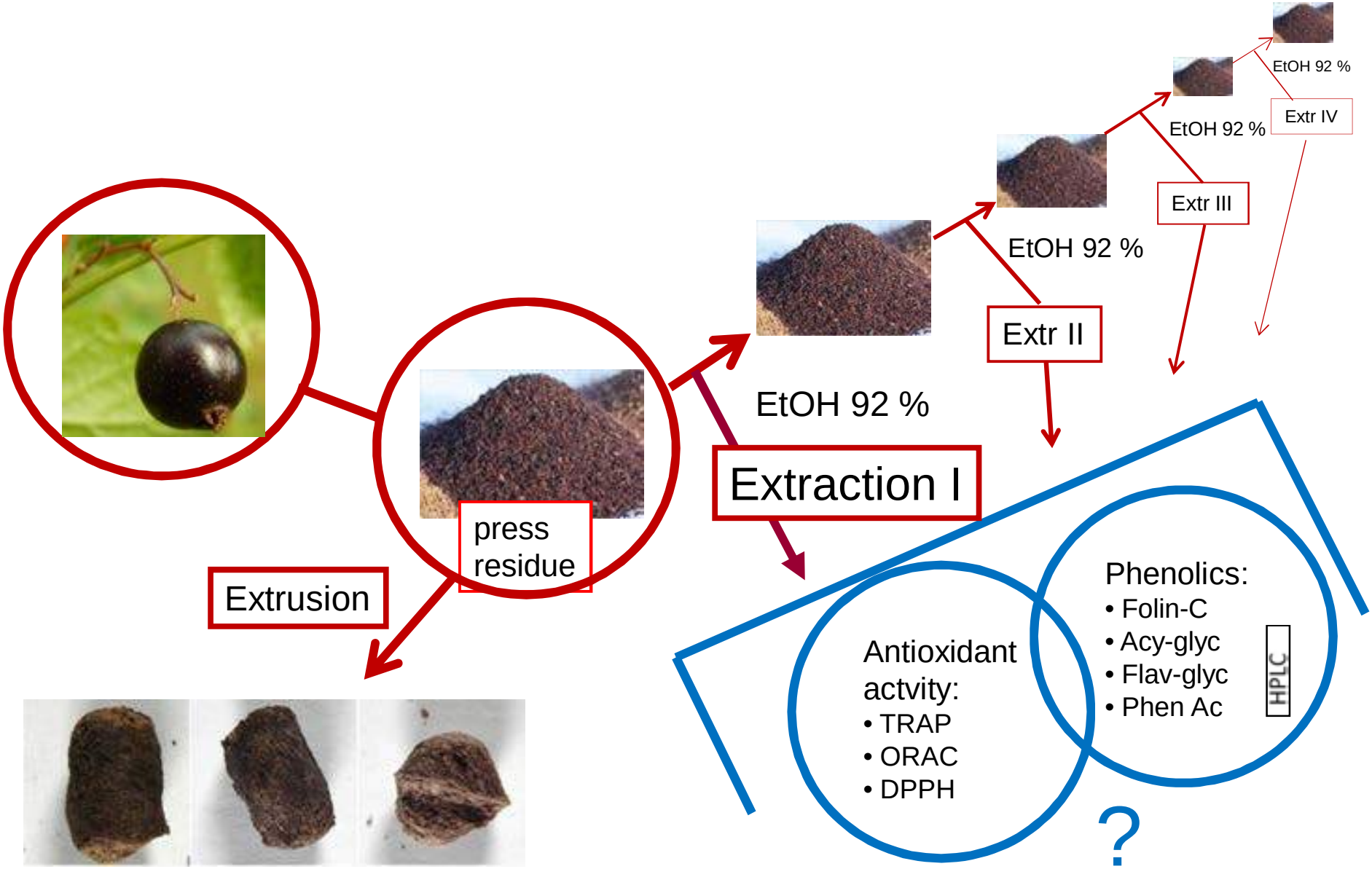
(2) Antioxidant Potential of Blackcurrant Press Residue Extracts



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Methods Applied in Analysis of the Antioxidant Potential

- **Juice pressing** by hydraulic press (no enzymes)
- **EtOH (92 %) extraction** of press residues + filtration (four consecutive extr.)
- **Filtration**, dilutions
- **Folin-Ciocalteu** reagent reducing capacity (“P.-C. phenolics”)
- **TRAP** (Total Radical-Trapping Antioxidant Parameter, luminometric)
- **ORAC** (Oxygen Radical Absorbance Capacity, fluorometric, Excitation 485 nm, emission 535 nm)
- **DPPH** (DPPH –radical trapping, reduction of violet at 515 nm)
- **HPLC phenolic profile** (anthocyanins, flavonols, phenolic acids)

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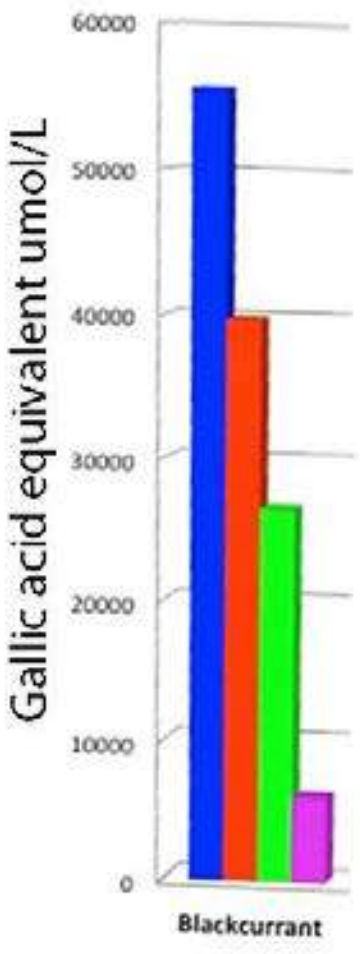
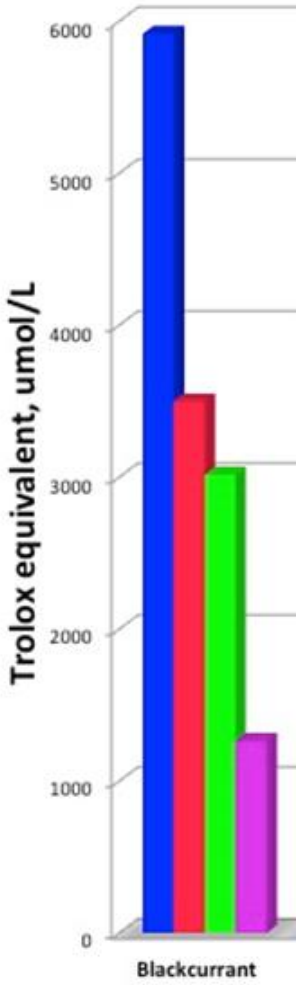
Profiles Obtained by Different Methods Give Different Results

(2)

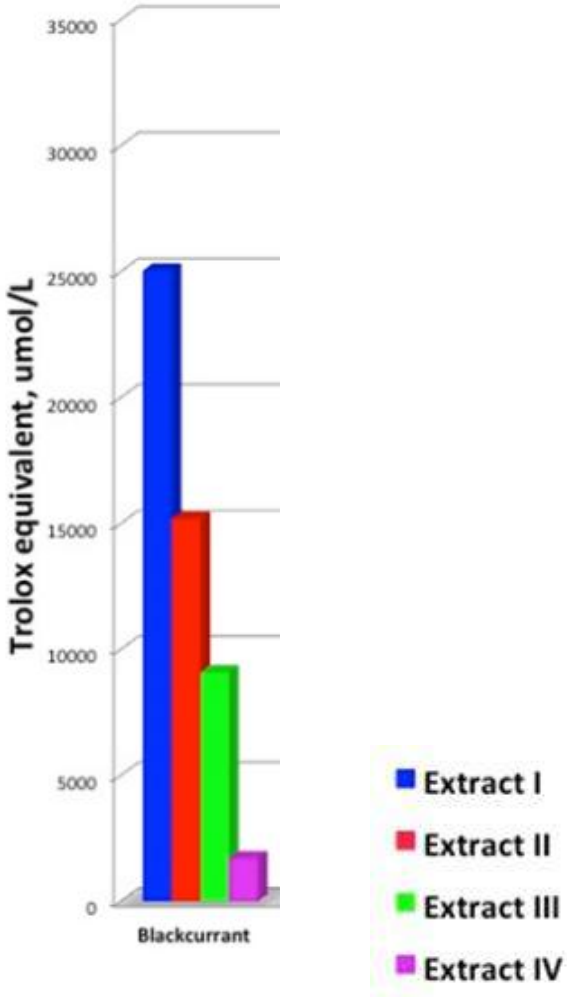
FOLIN-CIOC.



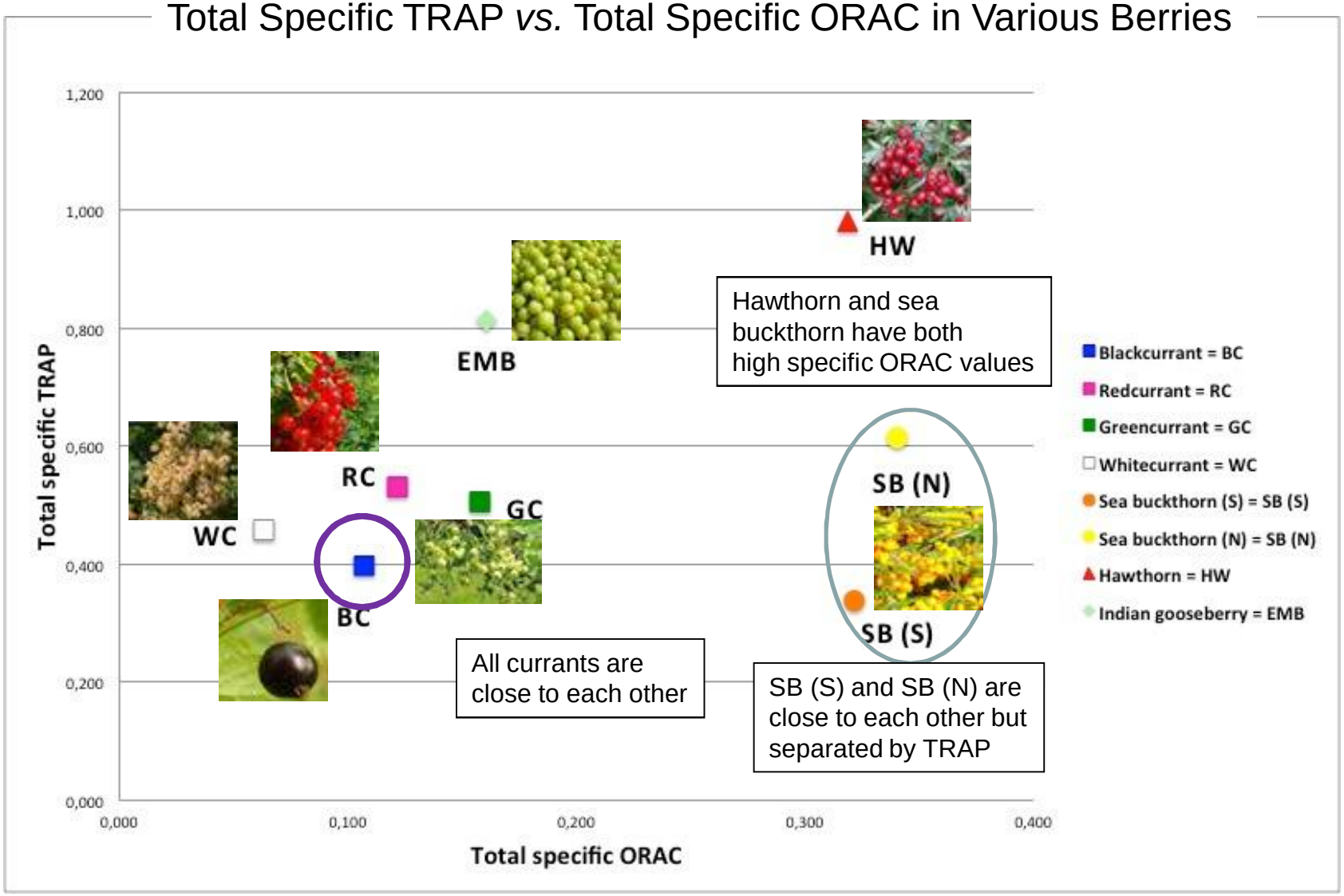
ORAC



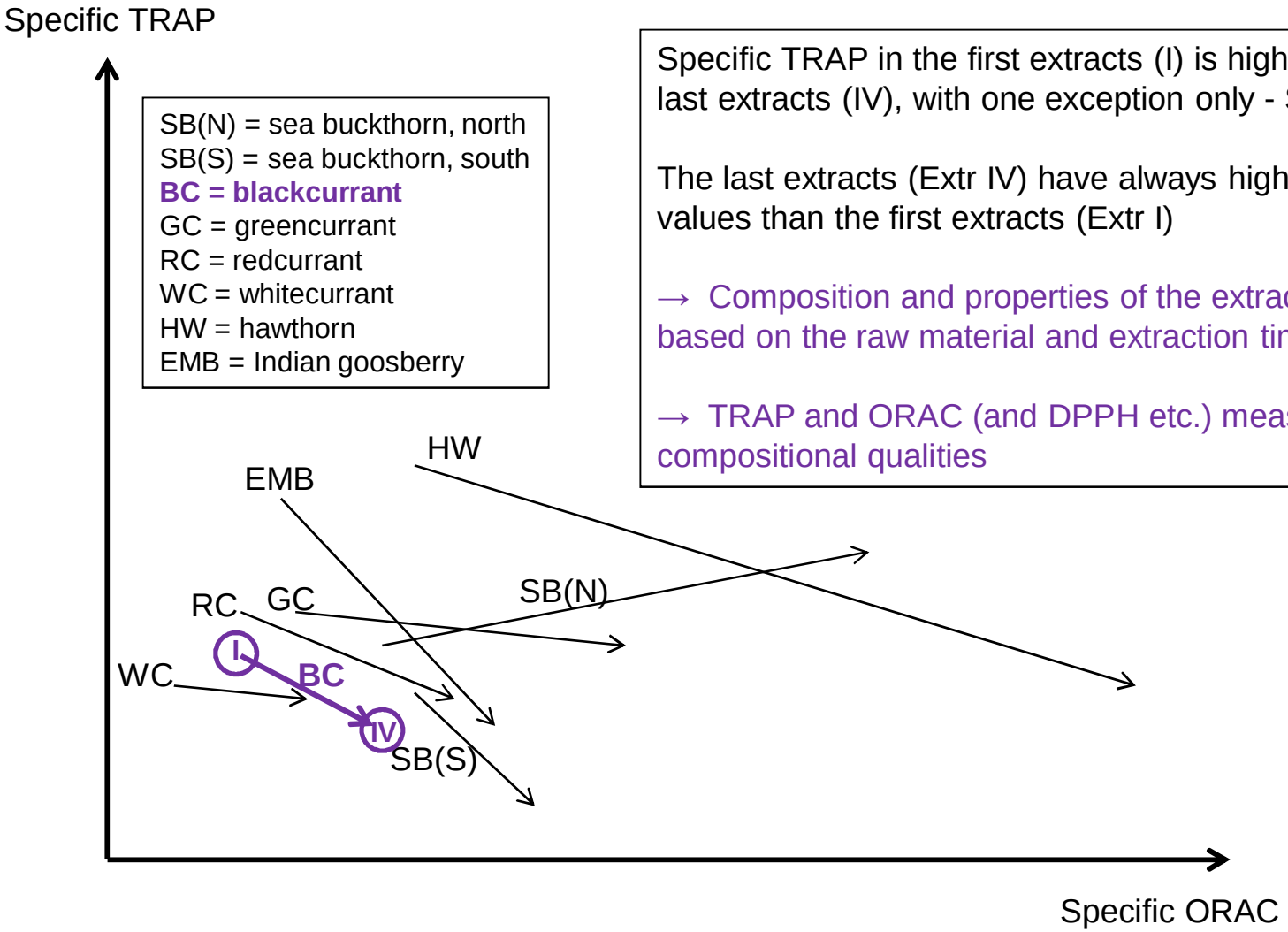
TRAP



Total Specific TRAP *vs.* Total Specific ORAC in Various Berries



Differences between Extract I and Extract IV in Various Berries



Specific TRAP in the first extracts (I) is higher than in the last extracts (IV), with one exception only - SB(N)

The last extracts (Extr IV) have always higher specific ORAC values than the first extracts (Extr I)

→ Composition and properties of the extracts vary based on the raw material and extraction time.

→ TRAP and ORAC (and DPPH etc.) measure different compositional qualities

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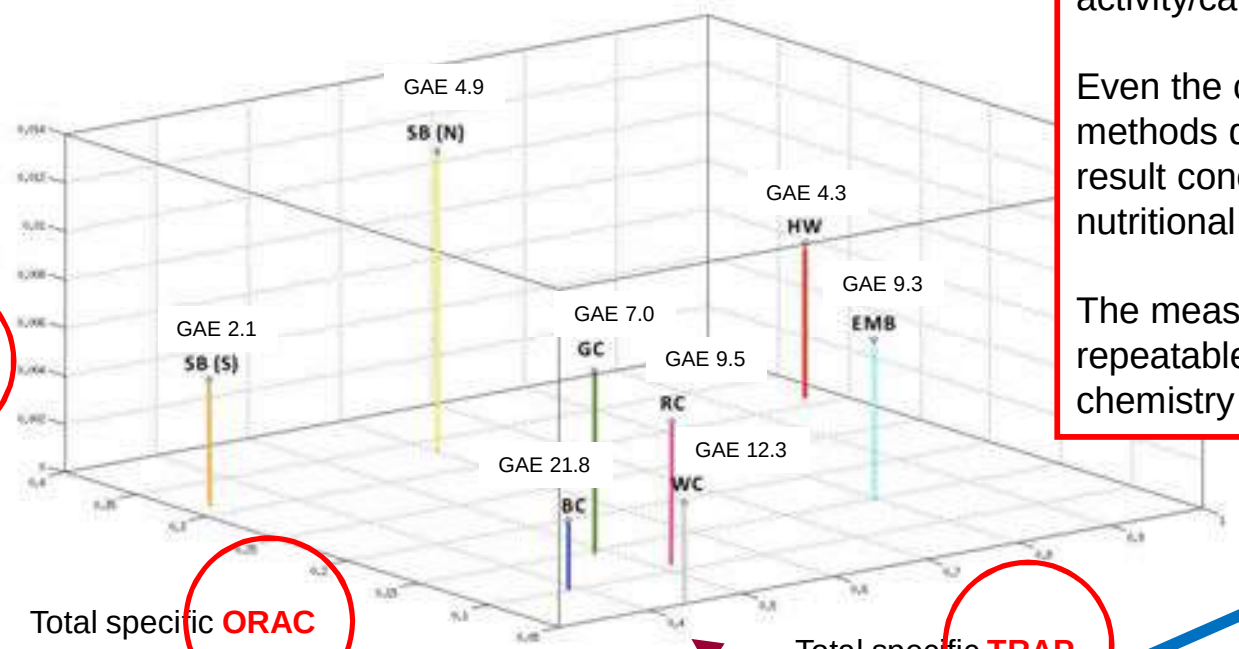
Interpretations?

(2)

No single method of analysis gives a universal answer of the antioxidative activity/capacity of a product

Even the combination of three methods does not give reliable result concerning the biological/ nutritional effects of the product

The measurements are rliable and repeatable – let us investigate the chemistry behind these values!



Total specific **DPPH**

Total specific **ORAC**

Total specific **TRAP**

SB(N) = sea buckthorn, north
SB(S) = sea buckthorn, south
BC = blackcurrant
GC = greencurrant
RC = redcurrant
WC = whitecurrant
HW = hawthorn
EMB = Indian goosberry

Antioxidant actvity:
- TRAP
- ORAC
- DPPH

?

Phenolics:
- Folin-C
- Acy-glyc
- Flav-glyc
- Phen Ac

HPLC

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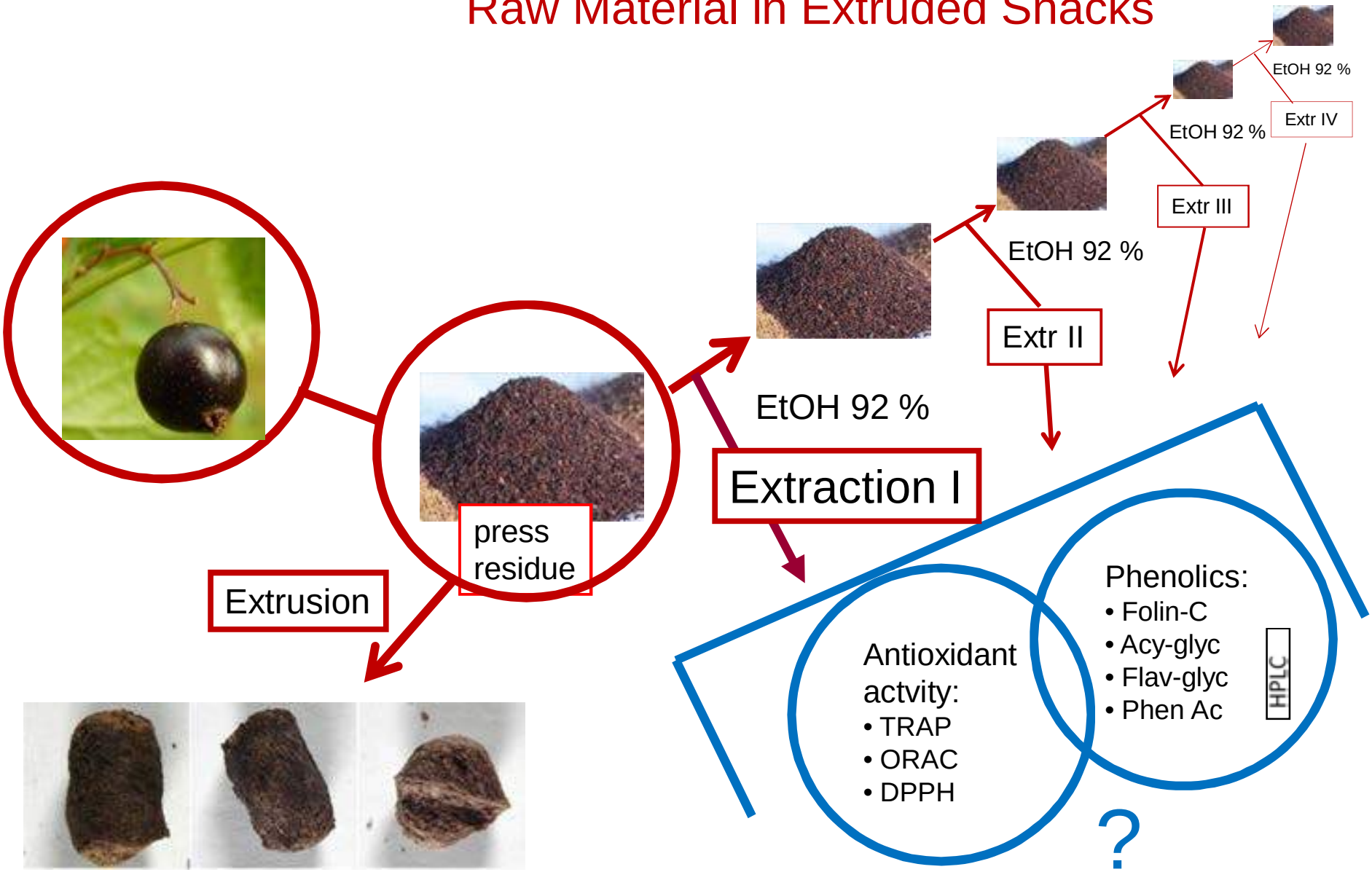
(3) Blackcurrant Press Residue as Raw Material in Extruded Snacks



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(3)

LWT - Food Science and Technology 57 (2014) 618–627

Contents lists available at ScienceDirect

LWT - Food Science and Technology

journal homepage: www.elsevier.com/locate/lwt

Exploiting blackcurrant juice press residue in extruded snacks

Leenamaija Mäkilä [a], Oskar Laaksonen [a], Jose Martin Ramos Diaz [b], Marjatta Vahvaselkä [c], Olavi Myllymäki [c], Ilkka Lehtomäki [c], Simo Laakso [c], Gerhard Jahreis [d], Kirsi Jouppila [b], Petra Larmo [a,1], Baoru Yang [a], Heikki Kallio [a,*]

[a] Food Chemistry and Food Development, Department of Biochemistry, University of Turku, FI-20014 Turku, Finland
[b] Department of Food and Environmental Sciences, University of Helsinki, FI-00014 Helsinki, Finland
[c] Department of Biotechnology and Chemical Technology, School of Chemical Technology, Aalto University, FI-00076 AALTO, Finland
[d] Department of Nutritional Physiology, Friedrich Schiller University of Jena, D-07743 Jena, Germany

ARTICLE INFO

Article history:
Received 8 August 2013
Received in revised form
11 November 2013
Accepted 4 February 2014

Keywords:
Blackcurrant
Extrusion
Liking
Physical properties
Press residue

ABSTRACT

Extrusion process was developed to exploit blackcurrant juice press residues from industrial side-streams. Press residues obtained from conventional enzymatic pressing, with high content of fiber and seed oil, and novel non-enzymatic juice processing, with high content of sugars, fruit acids and anthocyanins, were extruded with barley flour, oat flour or oat bran. The recipes consisted of blackcurrant press residues (30%), cereal materials (40%) and potato starch (30%) and small amount of sugar and salt. When compared to enzymatic press residue and oat bran, the novel non-enzymatic press residue extruded with barley or oat flour had higher expansion, lower hardness and density, higher redness (a^*), lower pH, and higher contents of fructose, glucose and fruit acids, all contributing positively to liking of texture, appearance, and flavor as well as berry-like experience. These characteristics were obtained with more gentle processing parameters, consisting of a lower total mass flow, screw speed and barrel temperature. Female consumers gave lower ratings in flavor, appearance and overall pleasantness for blackcurrant snacks than males. The study presented a sustainable way of utilizing industrial press residues from different processes of berry juice pressing for production of healthy snacks and breakfast cereals.

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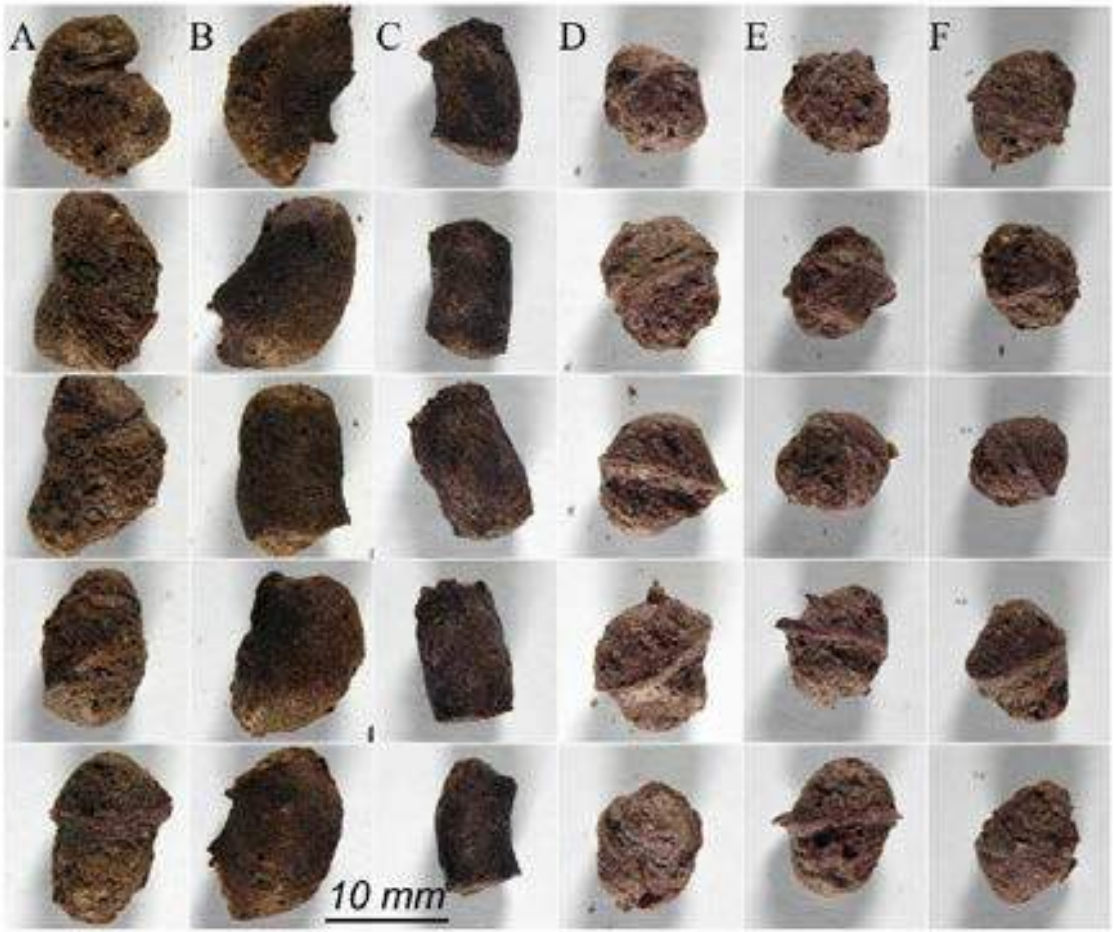

Fig. 1. Five parallel pictures of the samples: enzymatic residue (ER) extrudates A. ER-barley, B. ER-oat and C. ER-oat bran; non-enzymatic residue (NR) extrudates D. NR-barley, E. NR-oat and F. NR-oat bran.

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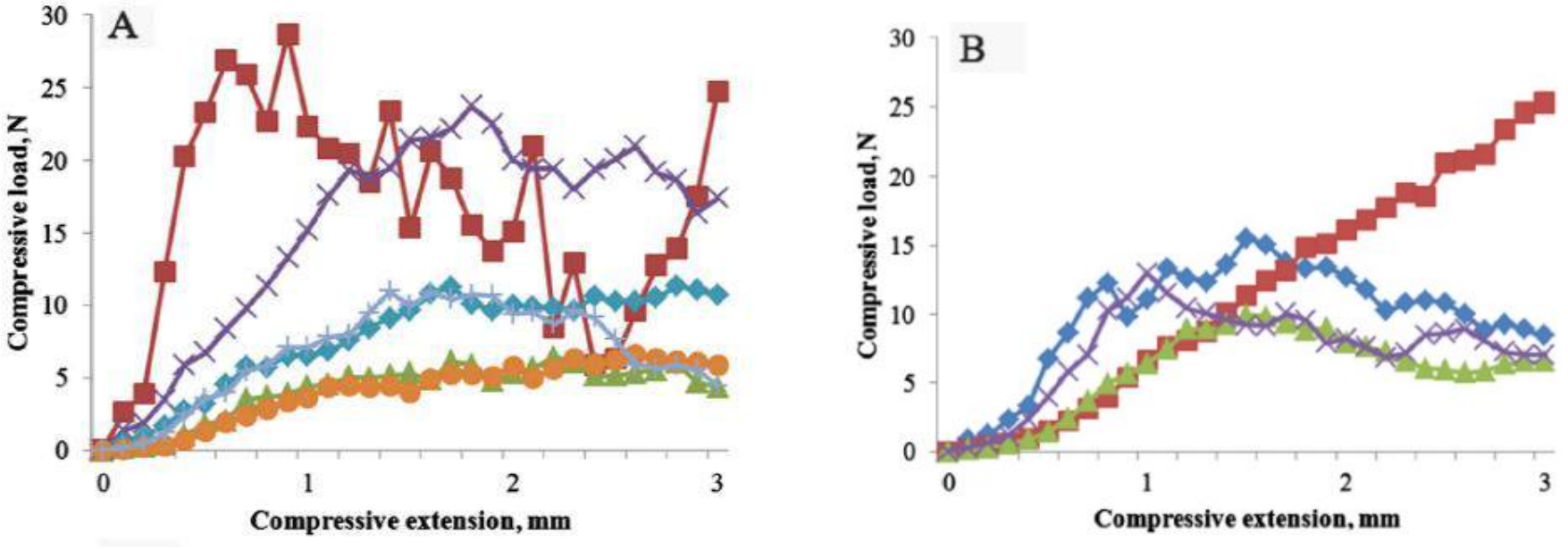

Fig. 2. Hardness values (N/mm: averages of triplicate measurements, 9–10 replicates) of the extrudates: (♦) ER-barley, (×) ER-oat, (■) ER-oat bran, (▲) NR-barley, (●) NR-oat and (+) NR-oat bran (A) and reference products: (♦) Reference 1, (■) Reference 2, (▲) Reference 3, (×) Reference 4 B). The force (compressive load; N) needed for probe to penetrate to certain depth (compressive extension; mm) into the extrudate. ER-oat bran had 2–9 replicates in compressive extensions 0.8–3.0 mm.

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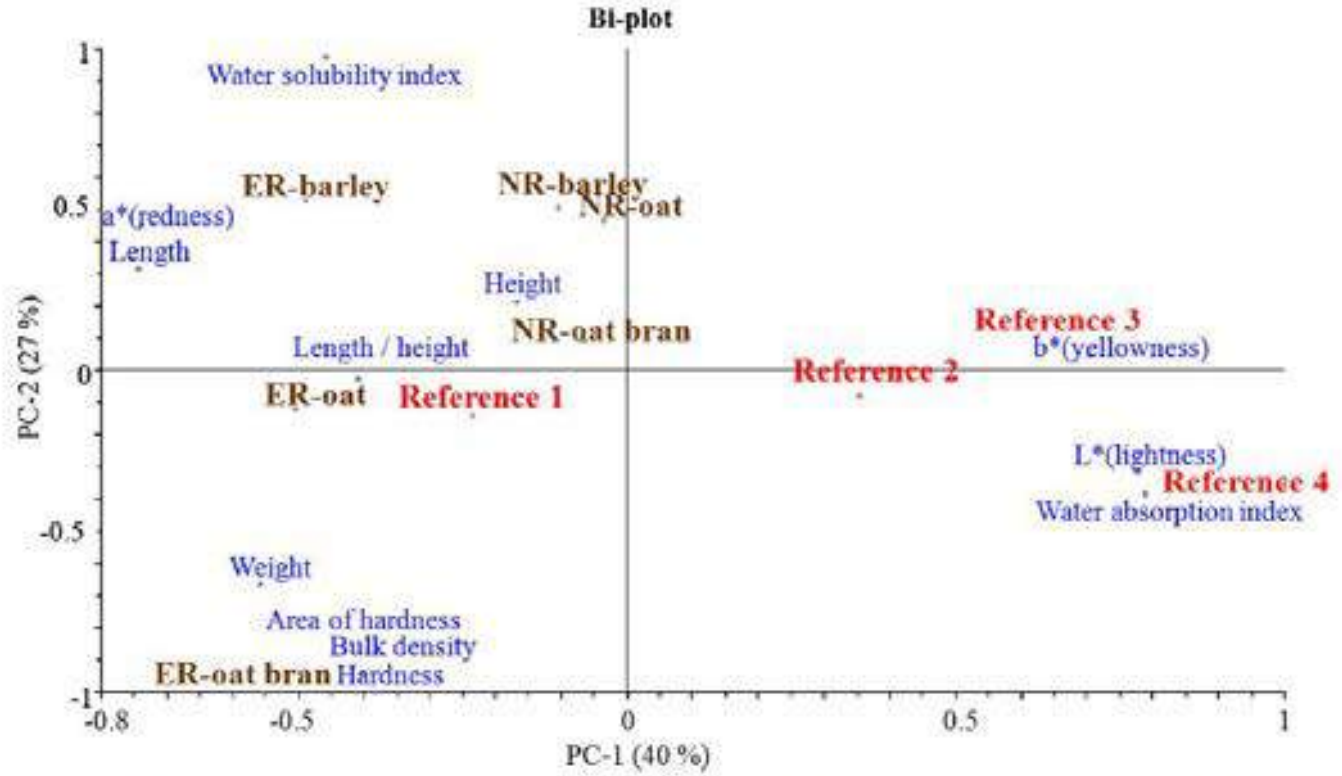

Fig. 3. PCA bi-plot of physical properties of the extrudates and reference products (References 1–4). The physical properties are presented in blue, reference products in red and extrudates in brown font. Abbreviations of physical properties refer to Table 3. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

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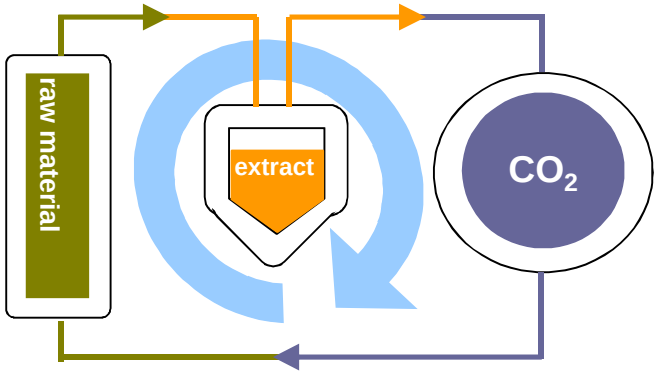
(4) Examples of Biological Effects of BC Seed Oil Extracted by Supercritical CO2



- No organic solvents
- No heat stress of raw materials or products
- Aseptic extracts possible
- CO_2 atmosphere during processing
- Environmental-friendly
- Often no further refining of the products needed
- Major applications are natural lipids and natural aroma compounds / perfumes

- Heavy investments needed
- If small facilities, high production costs
- Limited selectivity
- Technically most feasible modifiers not always practical
- Oils comparable with “non-refined” or “virgin”
- International growth has been slow – why?

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Composition of Currant Seed Oils Vary Based on Species, Varieties and Growth Conditions

Table 2 Fatty acid composition (mol%) of the seed oil triacylglycerols of *Ribes* berries. Fatty acids found at less than 0.1% of the total are included with the "others"

Johansson et al., ZLUF 1997, 204, 300-307

Fatty acid	Proportion, mol% (RSD)		
	Ribes nigrum	*Ribes spicatum*	*Ribes alpinum*
14:0	0.1 (2.7)	–	0.1 (44.2)
16:0	5.2 (0.9)	3.8 (7.4)	5.2 (0.9)
16:1 ($n-7$)	–	–	0.1 (1.8)
18:0	1.8 (0.2)	1.2 (0.8)	1.1 (1.0)
18:1 ($n-9$)	10.3 (0.2)	13.5 (5.2)	18.6 (0.1)
18:1 ($n-7$)	0.5 (1.0)	0.5 (4.2)	0.8 (1.1)
18:2 ($n-6$)	48.2 (0.3)	38.5 (1.5)	36.1 (0.6)
18:3 ($n-6$)	11.3 (0.3)	16.1 (3.5)	13.3 (1.6)
18:3 ($n-3$)	17.5 (0.1)	19.1 (7.0)	18.2 (1.0)
18:4 ($n-3$)	3.0 (0.2)	6.0 (4.5)	5.1 (0.4)
20:0	0.2 (7.6)	0.2 (22.4)	0.1 (1.2)
20:1 ($n-9$)	0.9 (1.0)	0.1 (6.2)	0.1 (5.0)
20:2 ($n-6$)	0.3 (1.3)	–	–
Others	0.9	1.1	1.2

Fatty acids in acyl glycerols and other fat soluble bioactive compounds of BC seed oil are assumed to be related to the health-beneficial effects in man



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Correlations of FAs in Alpine and Red Currant Seed Oils

Johansson et al Eur Food Res Technol, 2000, 211, 277-283

Table 3 Correlation coefficients between the polyunsaturated fatty acids of alpine currant and northern redcurrant seed oil

	Alpine currant		
	Linoleic acid	γ-Linolenic acid	α-Linolenic acid
γ-Linolenic acid	−0.76*	–	0.16
α-Linolenic acid	−0.27	0.16	–
Stearidonic acid	−0.72*	0.65	0.73*
	Northern redcurrant		
	Linoleic acid	γ-Linolenic acid	α-Linolenic acid
γ-Linolenic acid	0.72*	–	−0.92**
α-Linolenic acid	−0.89**	−0.92**	–
Stearidonic acid	−0.98**	−0.74*	0.93**

* Significant at P %<0.05; ** significant at $P<0.001$

(4)

Linnamaa et al., Clin Exp Allergy 2010, 40, 1247-1255

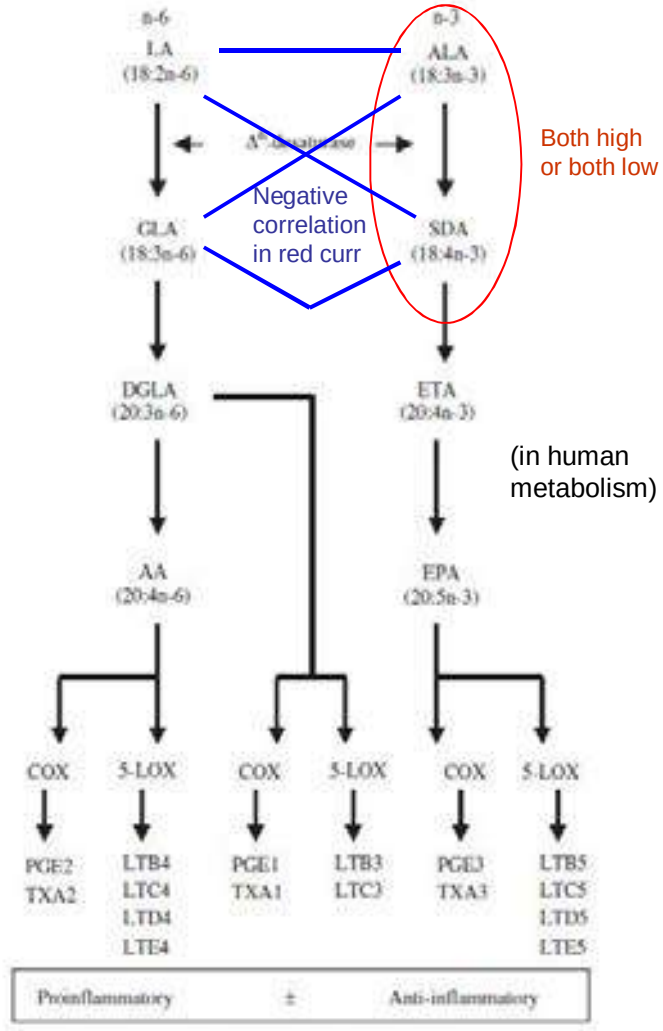

Fig. 1. The metabolism of n-6 and n-3 fatty acids and the synthesis of their eicosanoids presented together with the effects of different eicosanoids on inflammatory response [LA, linoleic acid; ALA, Δ-linolenic acid; GLA, γ-linolenic acid; SDA, stearidonic acid; DGLA, dihomo-γ-linolenic acid; ETA, eicosatetraenoic acid; AA, arachidonic acid; EPA, eicosapentaenoic acid; COX, cyclo-oxygenase pathway; 5-LOX, 5-lipo-oxygenase pathway; PGE, prostaglandin E; TXA, thromboxane; LTB, leukotriene B; LTC, leukotriene C; LTD, leukotriene D and LTE, leukotriene E]. Modified from Calder et al. 2002 [6].

BC oil prevents atopic dermatitis in newborns

Clinical & Experimental Allergy, 1–9 doi: 10.1111/j.1365-2222.2010.03540.x

ORIGINAL ARTICLE

Blackcurrant seed oil for prevention of atopic dermatitis in newborns: a randomized, double-blind, placebo-controlled trial

P. Linnamaa[1], J. Savolainen[2], L. Koulu[1], S. Tuomasjukka[3], H. Kallio[3], B. Yang[3], T. Vahlberg[4] and R. Tahvonen[3,5]

[1]Department of Dermatology, [2]Department of Pulmonary Diseases and Clinical Allergology, [3]Department of Biochemistry and Food Chemistry, [4]Department of Biostatistics, University of Turku, Turku, Finland and [5]MTT Agrifood Research Finland, Jokioinen, Finland

Summary

Background The present increased incidence of atopic diseases has been associated with an altered intake of essential fatty acids (EFAs). The composition of blackcurrant seed oil (BCSO) corresponds to the recommended dietary intake of EFAs, and as a dietary supplement could, in small doses, modify the imbalance of EFAs in an efficient way.

Objective To assess the effect of dietary supplementation with BCSO on the prevalence of atopy at 12 months of age.

Methods Three hundred and thirteen pregnant mothers were randomly assigned to receive BCSO (151) or olive oil as placebo (162). The first doses were administered at 8th–16th weeks of pregnancy and were continued until the cessation of breastfeeding, followed by supplementation to the infants until the age of 2 years. Atopic dermatitis and its severity (SCORAD index) were evaluated, serum total IgE was measured and skin tests were performed at the age of 3, 12 and 24 months.

Results Parental atopy was common (81.7%) among study subjects, making them infants with increased atopy risk. There was a significantly lower prevalence of atopic dermatitis in the BCSO group than in the olive oil group at the age of 12 months (33.0% vs. 47.3%, $P = 0.035$). SCORAD was also lower in the BCSO group than in the olive oil group at 12 months of age ($P = 0.035$). No significant differences in the prevalence of atopic dermatitis were observed between the groups at the age of 24 months ($P = 0.18$).

Conclusion Dietary supplementation with BCSO was well tolerated and it transiently reduced the prevalence of atopic dermatitis. It could therefore be one potential tool in the prevention of atopic symptoms when used at an early stage of life.

(Registration number SRCTN14869647, http://www.controlled-trials.com)

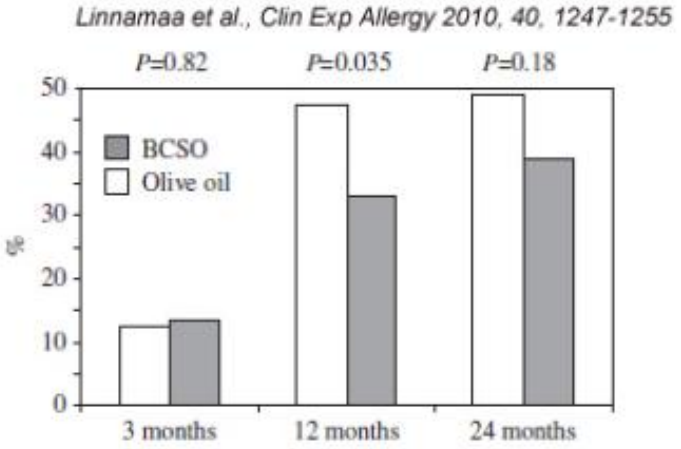

Fig. 3. Prevalence of atopic dermatitis for 1 month or longer in the blackcurrant seed oil (BCSO) intervention group (grey bar) and the placebo group (white bar) at three different study time-points: 3, 12 and 24 months. Statistical comparison with the χ^2-test.

Linnamaa et al.
Clin. Exp. Allergy **2010,** *40*, 1247-1255

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(4)

BC oil enhances innate and adaptive immunity *via* breast milk cytokines

Black currant seed oil supplementation of mothers enhances IFN-γ and suppresses IL-4 production in breast milk

Pia Linnamaa[1], Kaisa Nieminen[2], Leena Koulu[1], Saska Tuomasjukka[3], Heikki Kallio[3], Baoru Yang[3], Raija Tahvonen[3,4] & Johannes Savolainen[2]

[1]Department of Dermatology, University of Turku, Turku, Finland; [2]Department of Pulmonary Diseases and Clinical Allergology, Turku University Hospital, University of Turku, Turku, Finland; [3]Department of Biochemistry and Food Chemistry, University of Turku, Turku, Finland; [4]MTT Agrifood Research Finland, Jokioinen, Finland

To cite this article: Linnamaa P, Nieminen K, Koulu L, Tuomasjukka S, Kallio H, Yang B, Tahvonen R, Savolainen J. Black currant seed oil supplementation of mothers enhances IFN-γ and suppresses IL-4 production in breast milk. *Pediatr Allergy Immunol* 2013: **24**: 562–566.

Keywords
atopic dermatitis; breast milk; cytokines; fatty acids; intervention

Correspondence
Johannes Savolainen, Department of Pulmonary Diseases and Clinical Allergology, University of Turku, FI-20520 Turku, Finland
Tel.: +358 2 3132564
Fax: +358 2 3133328
E-mail: johannes.savolainen@utu.fi

Accepted for publication 1 July 2013

DOI:10.1111/pai.12110

Abstract

Background: The first year of infancy is crucial for the development of atopic immune response. Inadequate early Th1 and Treg responses and increased production of Th2 cytokines are associated with atopy. Breast milk contains several immunomodulatory cytokines and other factors that might influence the maturation of the infant's immune system. We assessed the cytokines in breast milk of mother of newborn infants and their associations with black currant seed oil (BCSO) supplementation during pregnancy, mother's atopic status and the development of infant's atopic dermatitis.

Methods: Mothers and infants from an intervention study by black currant seed oil (n = 31) or olive oil as placebo (n = 30) were included in the study. Breast milk samples were collected during the first 3 months of breastfeeding. Breast milk levels of IL-4, IL-5, IL-10, IL-12, IFN-γ and TNF were measured by Luminex technology.

Results: BCSO intervention group had decreased level of IL-4 ($p = 0.044$) and elevated level of IFN-γ ($p = 0.014$) in breast milk as compared to olive oil group. No significant differences were observed in IL-5, IL-10, IL-12 and TNF levels between the BCSO and olive oil groups. Mothers who had atopic dermatitis had significantly decreased levels of IL-10 ($p = 0.044$) in breast milk. Breast milk of the mothers of the children who developed atopic dermatitis had lower levels of IFN-γ ($p = 0.039$) as compared to the breast milk of the mothers of the children without dermatitis.

Conclusion: Dietary intervention with BCSO had immunomodulatory effects on breast milk cytokine production towards Th2 to Th1 immunodeviation.

Linnamaa et al.
Pediatric Allergy and Immunology
2013, *24,* 562-566

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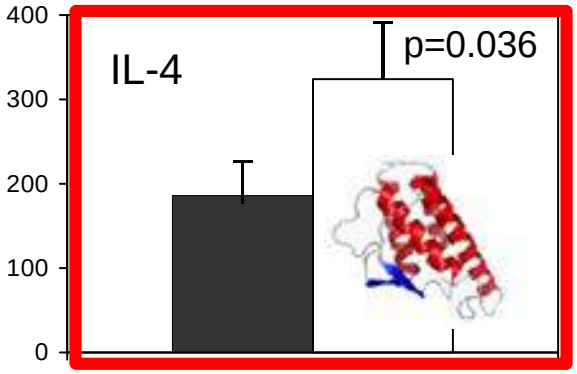
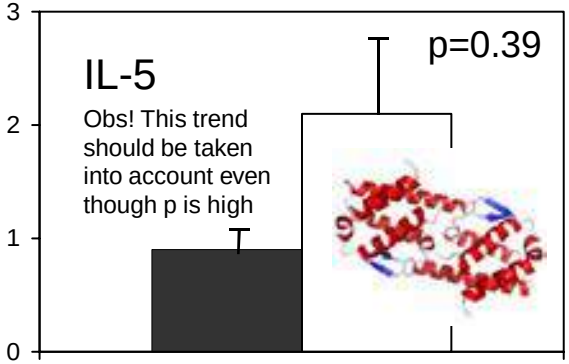
Breast milk concentrations (pg/ml) of mothers undergoing BSCO (black, n=31) and olive oil (white, n= 30) intervention

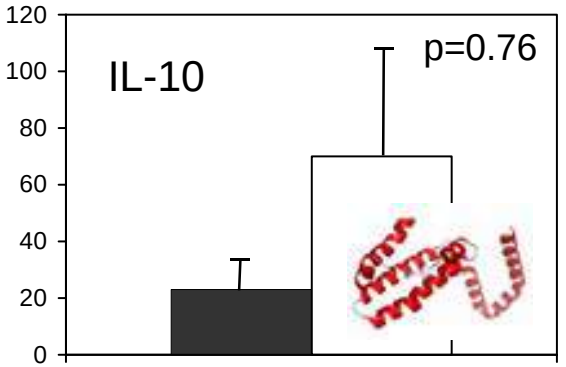
Interleukin 4 is a cytokine regulator in immunity
Decreases production of IFN-gamma
Overproduction associated with allergies

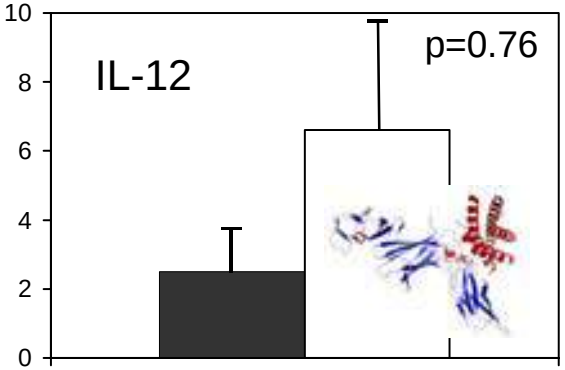
Associated with allergic rhinitis (jnflammation of nose mucose membranes) and asthma

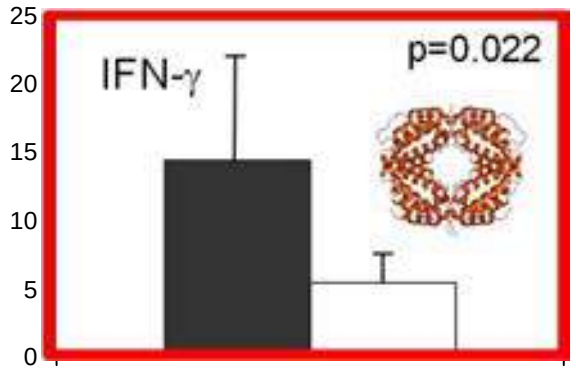
Pleiotropic effects in immunoregulation and inflammation
Inhibits synthesis of IFN-gamma

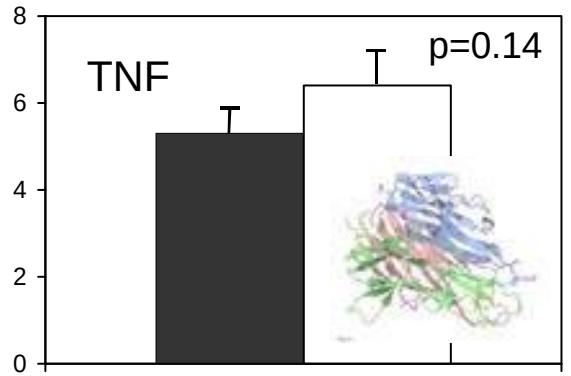












Stimulates production of IFN-gamma
Reduces IL-4 mediated suppression of IFN-gamma
Anti-angiogenic activity

Interferon γ is a dimerized soluble cytokine
Secreted by T helper cells
Antiviral, immunoregulatory, anti-tumor properties
Pro-inflammatory
Leds to cellular immunity
Critical for innate and adaptive immunity

Group of apoptosis-indicing cytokines

Modified from:
Linnamaa et al.
Pediatric Allergy and Immunology
2013, *24,* 562-566

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Not Only Blackcurrants!

There is an Extreme Selection of Berries to be Investigated

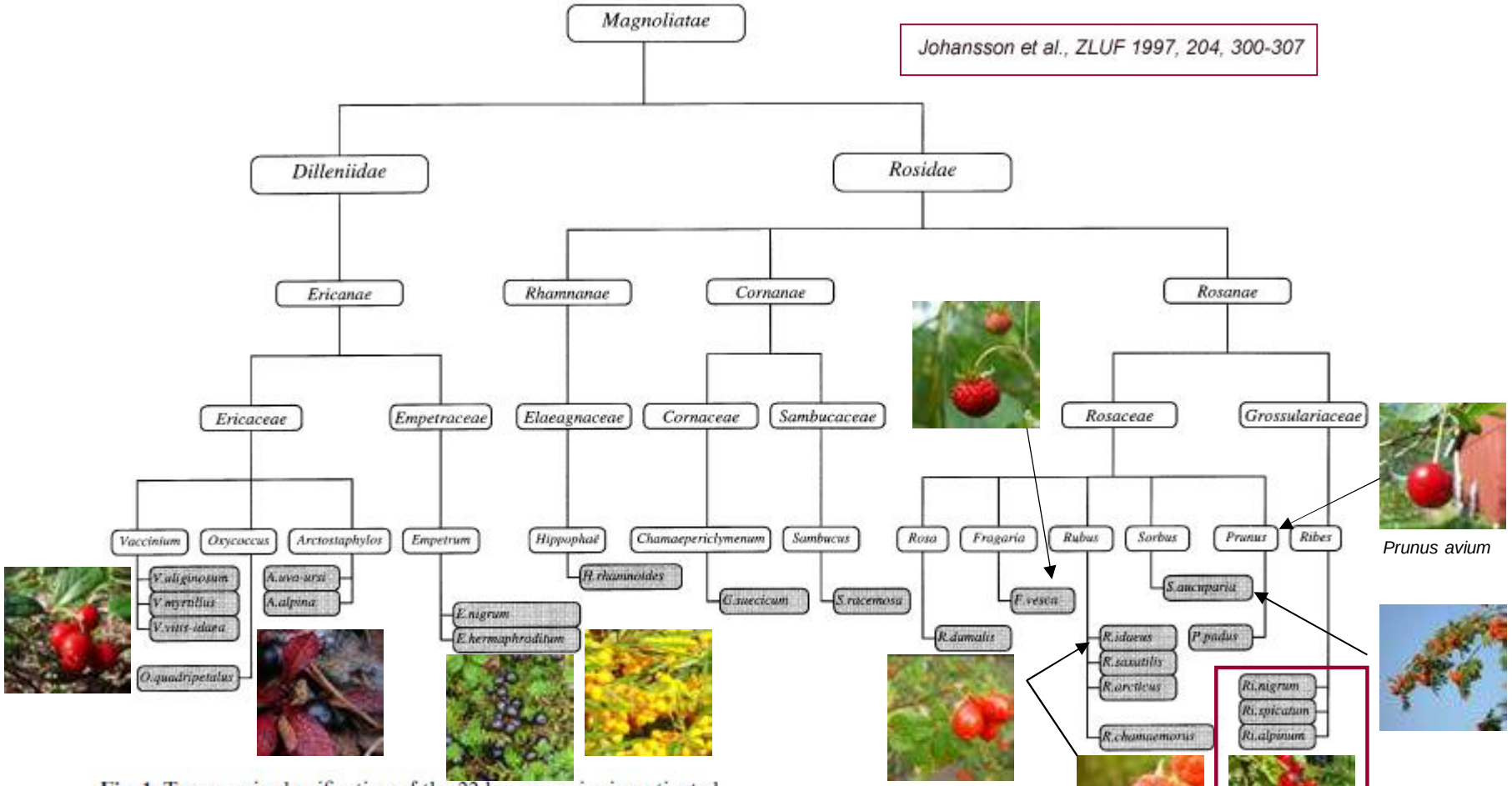

Fig. 1 Taxonomic classification of the 22 berry species investigated

Also other metabolites, e.g. aroma/ flvaor compounds, phenolics, sugars, acids, cutins etc. have been investigated at UTU

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PUBLICATIONS CONCERNING CURRANT BERRIES (*Ribes*) (by FoodChem and FoodDevel, UTU May 05th 2014)

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Turun yliopisto
University of Turku

Thank you!