biology and life sciences forum

MDPI

Proceedings

The Quality Assessment of Oils Obtained From Berry Fruit Seeds Using Pressurized Liquid Extraction †

Iga Piasecka [1,*], Agata Górska [1], Marko Obranović [2], Stanisław Kalisz [3], Ana Dobrincić [2], Erika Dobroslavić [2], Ewa Ostrowska-Ligęza [1], Rita Brzezińska [1] and Verica Dragović-Uzelac [2]

[1] Department of Chemistry, Institute of Food Sciences, Warsaw University of Life Sciences, 159C Nowoursynowska Street, 02-776 Warsaw, Poland; agata_gorska@sggw.edu.pl (A.G.), ewa_ostrowska-ligeza@sggw.edu.pl (E.O.-L.), rita_brzezinska@sggw.edu.pl (R.B.)
[2] Faculty of Food Technology & Biotechnology, University of Zagreb, Pierottijeva 6, 10 000 Zagreb, Croatia; mobran@pbf.hr (M.O.); adobrincic@pbf.hr (A.D.); edobroslavic@pbf.hr (E.D.); vdragov@pbf.hr (V.D.-U.)
[3] Division of Fruit, Vegetable and Cereal Technology, Department of Food Technology and Assessment, Institute of Food Sciences, Warsaw University of Life Sciences, 159C Nowoursynowska Street, 02-776 Warsaw, Poland; stanislaw_kalisz@sggw.edu.pl (S.K.)
* Correspondence: iga_piasecka@sggw.edu.pl (I.P.)
† Presented at The 4th International Electronic Conference on Foods, Focus on Sustainable Food Systems: Current Trends and Advances, online, 15–30 Oct 2023.

Abstract: Berry fruit seeds should be treated as a valuable waste product of fruit industry. In the following study oils from cranberry, black currant, red currant, strawberry and chokeberry seeds were extracted by conventional and pressurized liquid extraction. The quality of oils was assessed by determining oxidative stability (onset and maximum time of induction) with the use of pressure differential scanning calorimetry, fatty acids composition by gas chromatography and health indices, such as atherogenicity, thrombogenicity and hypocholesterolemic indexes. Additionally, health-promoting index was calculated. It was found that fatty acid profile was not affected when pressurized liquid extraction was used. Major fatty acid in the studied oils was linoleic acid ranging from 36% for cranberry seed oil to 69% for chokeberry seed oil, followed by α-linolenoic acid in case of cranberry, strawberry and redcurrant seed oils or by oleic acid for chokeberry and blackcurrant seed oils. The oxidative stability of fats extracted with the use of pressurized solvent was significantly lower, comparing to oils obtained in conventional extraction process e.g., maximum induction time for conventionally extracted chokeberry seed oil was 40.74 ± 0.66 min and 9.24 ± 0.57 min when pressurized liquid extraction was applied. The studied oils had low values of atherogenicity and thrombogenicity indexes, what with combination with high values of hypocholesterolemic index allow to qualify them for high nutritional quality oils. However, further studies, regarding process optimization in order to obtain oils with improved quality, mainly better oxidative stability, is needed.

Keywords: accelerated solvent extraction; pressurized liquid extraction; oil extraction; fruit seeds; oxidative stability; PDSC; GC

Citation: To be added by editorial staff during production.

Academic Editor: Firstname Lastname

Published: date

1. Introduction

The conventional methods of extractions are being gradually replaced by novel, alternative extraction methods, which may be beneficial in terms of energy and solvents consumption and selectivity of the process. One of the techniques, considered as 'green' is pressurized liquid extraction (PLE), which involves using liquid solvent heat up to a temperature above its boiling point and at the same time, applying high pressure to let the solvent remain liquid. The main advantage of the PLE technique is that increased temperature improves the diffusion process and solubility of the extracted molecules with the decrease in amount of solvent used. High pressure also promotes mass transfer during

the process. Both, high temperature and high pressure reduce the surface tension of the solvent which also influences improved extraction efficiency [1].

The main objective of this work was to evaluate the possibility to apply PLE to obtain oil from berry seeds and to study the effect of PLE on the quality of oil, with special regard to oxidative stability and fatty acids profile of fat fraction. For comparison, conventional extraction of oils from berries seeds was performed, followed by studies of their quality characteristics.

2. Materials and Methods

2.1. Materials

The seeds of cranberry, black currant, red currant, strawberry and chokeberry were used for analysis. Seeds were ground in a laboratory tube mill (IKA Werke, Staufen im Breisgau, Germany) at 25,000 rpm, 30 s.

2.2. Pressurized liquid extraction (PLE) of oil

Ground samples of 5 g of seeds were mixed with diatomaceous earth and were placed in cells made from stainless steel. The process was conducted following the methodology described by Dobroslavić et al. [2] in a Dionex ASE 350 Accelerated Solvent Extractor (Thermo Fisher Scientific Inc., Sunnyvale, CA, USA) in 4 cycles with n-hexane as a solvent. Temperature of the oven was set at 150 °C and the time of static extraction time was 5 min. The pressure was 10.34 MPa, purge with nitrogen 30 s and volume flush was set at 50%. The glass vials with Teflon septa were used to collect the extracts. The solvent was removed using a vacuum evaporator, oils were dried in a nitrogen atmosphere to remove residual hexane. Oil yield values were calculated according to the equation (1):

$$(1)\ Y = \frac{m_o\,[g]}{m_s\,[g]} \times 100\%$$

where Y – oil yield; m_o – mass of oil; m_s – initial mass of powdered seeds.

2.3. Conventional extraction (CE) of oil

The extraction of oil in Soxhlet apparatus was done based on the methodology described by Dobson et al. [3] with modifications. Ground samples of 5 g of seeds were put into the apparatus in a paper thimble with 150 mL of n-hexane. The extraction was done in 5 cycles, then, solvent was evaporated. Obtained oils were dried in a nitrogen atmosphere and oil yield was calculated as for the PLE method.

2.4. Resistance to oxidation

Oxidative stability of oils was determined using pressurized differential scanning calorimetry (PDSC) described by Bryś et al. [4] at 120 °C. A DSC Q20 apparatus (TA Instruments, New Castle, DE, USA) was used. Aluminum pan filled with oil and an empty pan used as a reference were placed in the cell under oxygen atmosphere at initial pressure set at 1400 kPa. The oxidation reaction induction maximum (OIT) and onset times (OOT) of examined oils were determined.

2.5. Fatty acids analysis

The fatty acids profile of extracted oils was determined after converting fatty acids to fatty acid methyl esters (FAME) according to PN-EN ISO:2001 standard method [5] followed by gas chromatography using YL6100 GC apparatus (Young Lin Bldg., Anyang, Hogye-dong, Korea), according to previously described method [6]. The results are expressed as a percentage of each fatty acid.

2.6. Health indices of oils

Health indices based on fatty acids profile: PUFA/SFA ratio, indexes of atherogenicity (IA) and of thrombogenicity (IT), developed by Ulbritcht and Southgate [7], hypocholesterolemic/hypercholesterolemic ratio (HH) developed by Santos Silva et al. [8] and health-promoting index (HPI) proposed by Chen et al. [9] were counted according to equations (2-5):

$$(2)\ IA = \frac{C12{:}0+(4\times C14{:}0)+C16{:}0}{\sum UFA}$$

$$(3)\ IT = \frac{C14{:}0+C16{:}0+C18{:}0}{0.5\times\sum MUFA+(0.5\times\sum n-6\ PUFA)+(3\times\sum n-3\ PUFA)+\frac{n-3}{n-6}}$$

$$(4)\ HH = \frac{cis-C18{:}1+\sum PUFA}{C12{:}0+C14{:}0+C16{:}0}$$

$$(5)\ HPI = \frac{\sum UFA}{C12{:}0+4\times C14{:}0+C16{:}0}$$

2.7. Statystical analysis

Collected results are presented as mean value ± standard deviation. The one-way ANOVA and post-hoc Tukey's test with a p-value of 0.05 chosen to consider significant differences were performed using Statistica 13.3 Software (StatSoft, Kraków, Poland).

3. Results and discussion

3.1. Extraction efficiency

The yields of oil extractions are presented in Table 1. In the case of conventional extraction, all the yield values, except the red currant seed oil, were lower than oil yields of PLE. Usage of high temperatures during PLE process was responsible for an increased efficiency of the extraction as it caused overcoming cohesive interactions between molecules and decreasing activation energy required for molecules release. Therefore, the surface tension of n-hexane was consequently lowered by enhanced temperature, so the oil dissolution in hexane was accelerated. Also, high pressure forced n-hexane to penetrate matrix extensively [1].

Table 1. Oil yield in pressurized liquid extraction and conventional extraction.

Oil source	PLE oil yield [%]	CE oil yield [%]
Cranberry	16.00	11.52
Strawberry	13.53	10.94
Chokeberry	5.08	3.59
Black currant	7.60	3.31
Red currant	1.70	5.84

3.2. Oxidative stability of oils

Examples of curves obtained in one run of PDSC study are presented in Figure 1. The results of PDSC study are presented in Table 2. Generally, oils obtained applying pressurized liquid extraction were characterized by low OIT and OOT. OIT of pressurized-liquid extracted black currant and red currant seed oils were undetectable as the peak was evident right on the beginning of the PDSC diagram. Based on the obtained results, it can be stated that conventionally extracted cranberry seed oil with OOT equal to 44.67 ± 2.49 min. and OIT equal to 50.20 ± 2.12 min was the most stable. Also, in case of PLE oils, cranberry seed oil was significantly more resistant to oxidation than others.

Table 2. Onset (OOT) and maximum (OIT) induction time of oils extracted using pressurized-liquid extraction and conventional extraction.

Oil source	PLE oils	CE oils

	OOT [min]	OIT [min]	OOT [min]	OIT [min]
Cranberry	8.44 ± 1.39^{B}	13.94 ± 0.98^{C}	44.67 ± 2.49^{a}	50.20 ± 2.12^{a}
Strawberry	3.06 ± 0.95^{A}	6.71 ± 0.42^{A}	6.34 ± 1.78^{c}	10.80 ± 1.30^{c}
Chokeberry	3.04 ± 1.03^{A}	9.24 ± 0.57^{B}	35.21 ± 0.52^{b}	40.74 ± 0.66^{b}
Black currant	-	-	32.84 ± 2.73^{b}	37.91 ± 2.70^{b}
Red currant	-	-	27.64 ± 0.54^{b}	33.31 ± 0.62^{b}

mean values with different letters (A-C and a-c) in the columns are significantly different at α = 0.05

Comparing oxidative stability of conventionally extracted oils to other plant derived oils, it can be seen that studied oils were characterized by lower OIT, measured in PDSC analysis at 120 °C, than rapeseed oil with OIT at approximately 66-74 min [10,11]. Results obtained for PLE oils in the following study comply with results obtained by De Mello et al. [12], who studied PL-extracted crambe seed oil. Applying higher temperatures of the process resulted in higher oil yield, sterols and γ-tocopherol content, however oxidative stability of oil was lower, comparing to commercial oil.

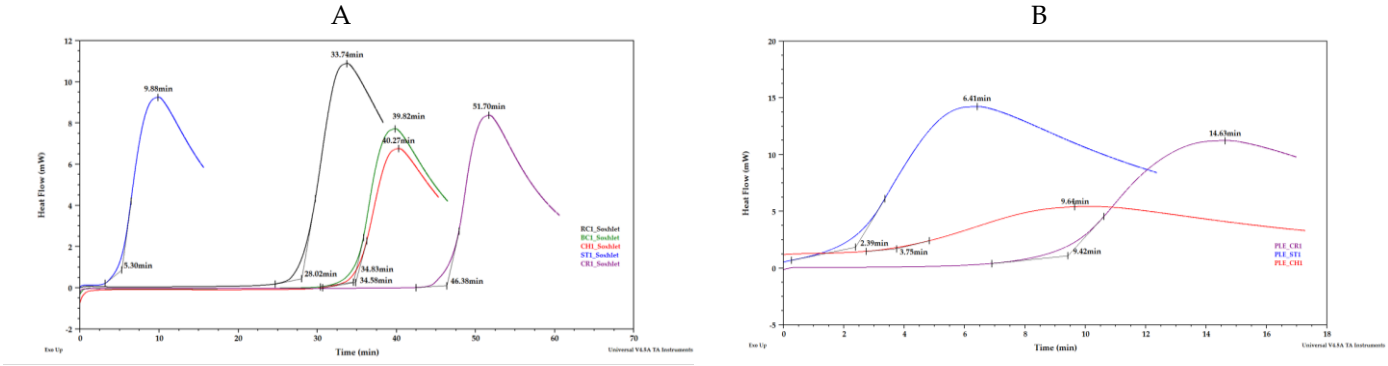

Figure 1. Examples of PDSC diagrams of conventionally extracted oils (A) and PLE oils (B); RC- red currant, BC- black currant, CH- chokeberry, ST- strawberry, CR- cranberry.

Low oils' resistance to oxidation in case of applying PLE method may occur due to high selectivity of the accelerated solvent extraction. Thus, possibly the only molecules extracted in the PLE were fats, without accompanying antioxidant compounds. In addition, oils by themselves might have been oxidized due to severe conditions of extraction [1,4]. Another reason for lowering OIT could be ability of high temperature-solvent to decrease the activation energy, therefore oxidation reaction may be initiated easier [1].

3.3. Fatty acids profile and health indices

Summarized results of fatty acids profile and counted health indices based on the Chen and Liu review [13] are presented in Table 3. Independently of the applied extraction method, dominant fatty acid in the examined oils was linoleic acid (C18:2 n-6). The contribution of sum of SFA, MUFA and PUFA in the fatty acids profile was similar. The differences between oil sourced from the same berry fruit seeds were only slight; however, even slight changes in fatty acid profile affect the health indices of fat. Fatty acids profiles obtained in the following study were in agreement with previously published [14,15].

4. Conclusions

Pressurized liquid extraction is a novel, green technique of separation which can be successfully used to extract oil from berry fruit seeds. Although the yield of the extraction conducted using pressurized solvent was acceptable and the fatty acids profile was not affected, the oxidative stability of fats was significantly lower, comparing to oils obtained in conventional extraction. However, applying different conditions may be beneficial and

may lead to obtaining oils with better qualitative properties. The studied oils were characterized by low values of atherogenicity and thrombogenicity indexes combined with high values of hypocholesterolemic index allow to consider berry fruit seeds oils as important element of dietoprophylaxis and dietotherapy of cardiovascular diseases. Further studies, regarding process optimization in order to obtain oils with improved quality, mainly better oxidative stability, is needed.

Table 3. Fatty acid profile [%] of extracted oils and nutritional indices of oils based on their fatty acid profile

PUFA/SFA- polyunsaturated fatty acid/saturated fatty acid ratio; IA- index of atherogenicity; IT- index of thrombogenicity; HH- hypocholesterolemic/hypercholesterolemic ratio; HPI- health-promoting index; n.d.- not detected; mean values with different letters (a-f) in the rows are significantly different at $\alpha = 0.05$.

Fatty acids		Cranberry seed oil		Strawberry seed oil		Chokeberry seed oil		Black currant seed oil		Red currant seed oil	
		PLE	CE	PLE	CE	PLE	CE	PLE	CE	PLE	CE
SFA	16:0	5.84 ± 0.06	5.74 ± 0.05	5.54 ± 0.01	5.07 ± 0.04	6.38 ± 0.04	5.22 ± 0.31	9.14 ± 0.11	6.98 ± 0.01	7.50 ± 0.04	5.88 ± 0.06
	18:0	1.23 ± 0.08	1.31 ± 0.08	2.00 ± 0.12	1.98 ± 0.13	1.63 ± 0.03	1.48 ± 0.01	2.62 ± 0.04	1.97 ± 0.03	1.65 ± 0.00	1.56 ± 0.01
	∑SFA	7.06 ± 0.14^{a}	7.04 ± 0.03^{a}	7.53 ± 0.11^{b}	7.05 ± 0.09	8.01 ± 0.01^{b}	6.70 ± 0.30^{a}	11.75 ± 0.07^{b}	8.95 ± 0.04^{a}	9.15 ± 0.04^{b}	7.43 ± 0.06^{a}
MUFA	16:1	0.11 ± 0.01	0.11 ± 0.01	0.23 ± 0.03	0.22 ± 0.02	0.18 ± 0.00	0.18 ± 0.01	0.21 ± 0.01	0.14 ± 0.01	0.36 ± 0.01	0.16 ± 0.00
	18:1 n-9	23.52 ± 0.03	24.01 ± 0.08	18.79 ± 0.08	18.31 ± 0.10	17.38 ± 0.06	17.99 ± 0.05	20.51 ± 0.04	15.86 ± 0.02	13.73 ± 0.01	11.98 ± 0.00
	∑MUFA	23.63 ± 0.01^{a}	24.11 ± 0.08^{b}	19.02 ± 0.11^{b}	18.53 ± 0.12^{a}	17.56 ± 0.06^{a}	18.16 ± 0.04^{a}	20.71 ± 0.03^{b}	16.00 ± 0.01^{a}	14.09 ± 0.03^{b}	12.14 ± 0.00^{a}
PUFA	18:2 n-6	36.35 ± 0.04	36.40 ± 0.06	44.88 ± 0.09	44.84 ± 0.11	67.23 ± 0.07	69.38 ± 0.06	45.03 ± 0.11	45.82 ± 0.12	38.54 ± 0.01	40.30 ± 0.02
	18:3 n-6	0.12 ± 0.02	0.12 ± 0.01	0.12 ± 0.01	0.10 ± 0.00	n.d.	n.d.	8.72 ± 0.02	12.08 ± 0.06	4.00 ± 0.01	4.27 ± 0.01
	18:3 n-3	31.80 ± 0.11	31.07 ± 0.04	28.46 ± 0.14	29.49 ± 0.08	1.99 ± 0.06	0.87 ± 0.01	10.15 ± 0.06	13.19 ± 0.01	31.48 ± 0.09	32.77 ± 0.04
	∑PUFA	68.27 ± 0.05^{b}	67.59 ± 0.11^{a}	73.46 ± 0.22^{a}	74.43 ± 0.20^{b}	69.22 ± 0.01^{a}	70.24 ± 0.07^{b}	63.89 ± 0.03	71.08 ± 0.06^{b}	74.01 ± 0.07^{a}	77.33 ± 0.07^{b}
Other		1.04 ± 0.06	1.28 ± 0.01	n.d.	n.d.	5.23 ± 0.06	4.90 ± 0.18	3.67 ± 0.08	3.98 ± 0.03	2.76 ± 0.06	3.10 ± 0.01
Health indices of oils based on fatty acid profile											
PUFA/SFA		9.67	9.60	9.75	10.56	8.65	10.44	5.44	7.95	8.09	10.41
IA		0.06	0.06	0.06	0.05	0.07	0.06	0.11	0.08	0.09	0.07
IT		0.06	0.06	0.06	0.06	0.16	0.11	0.17	0.11	0.07	0.06
HH		35.22	35.79	16.66	18.10	13.58	16.90	9.24	12.46	11.71	15,19
HPI		15.75	15.99	16.71	18.35	13.61	16.93	9.26	12.48	11.75	15.23

Author Contributions: Conceptualization, I.P., A.G. and S.K.; methodology, I.P., M.O., A.D. and E.D.; software, I.P..; validation; formal analysis; data curation; writing—original draft preparation; investigation, I.P., M.O. and R.B.; resources, A.G. and S.K.; writing—review and editing, A.G..; visualization, I.P. and E.O.-L.; supervision, A.G. and V.D.-U.; project administration, I.P. and A.G.; funding acquisition, I.P., A.G. and V.D.-U. All authors have read and agreed to the published version of the manuscript.

Funding: Not applicable.

Acknowledgments: This research was partially performed during research stay supported by Warsaw University of Life Sciences' Own Scholarship Fund for academic staff and PhD students.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Mustafa: A.: Turner, C. Pressurized liquid extraction as a green approach in food and herbal plants extraction: A review. *Anal Chim Acta* **2011**, 703(1), 8-18. doi:10.1016/j.aca.2011.07.018
2. Dobroslavić, E.; Elez Garofulić, I.; Šeparović, J.; Zorić, Z.; Pedisić, S.; Dragović-Uzelac, V. Pressurized Liquid Extraction as a Novel Technique for the Isolation of Laurus nobilis L. Leaf Polyphenols. *Molecules* **2022**, 27, 5099. https://doi.org/10.3390/molecules27165099
3. Dobson, G.; Shrestha, M.; Hilz, H.; Karjalainen, R.; McDougall, G.; Stewart, D. Lipophilic components in black currant seed and pomace extracts. *Eur. J. Lipid Sci. Technol.* **2011**, 114, 575–582. https://doi.org/10.1002/ejlt.201100313
4. Bryś, J.; Wirkowska, M.; Górska, A.; Ostrowska-Ligęza, E; Bryś, A. Application of the calorimetric and spectroscopic methods in analytical evaluation of the human milk fat substitutes. *J Therm Anal Calorim* **2014**, 118, 841-848. https://doi.org/10.1007/s10973-014-3893-1
5. Polish Norm: PN-EN ISO: 5509:2001: Oil and Vegetable and Animal Fats. Preparation of Methyl Ester of Fatty Acids; Polish Committee for Standardization: Warsaw, Poland, 2001
6. Piasecka, I.; Górska, A.; Ostrowska-Ligęza, E.; Kalisz, S. The Study of Thermal Properties of Blackberry, Chokeberry and Raspberry Seeds and Oils. *Appl. Sci.* **2021**, *11*, 7704. https://doi.org/10.3390/app11167704
7. Ulbricht, T.L.V.; Southgate, D.A.T. Coronary heart disease: seven dietary factors. *The Lancet* **1991**, 338(8773), 985-992. https://doi.org/10.1016/0140-6736(91)91846-M.
8. Santos-Silva, J.; Bessa, R.J.B.; Santos-Silva, F. Effect of genotype, feeding system and slaughter weight on the quality of light lambs: II. Fatty acid composition of meat. *Livest. Prod. Sci.* **2022**, 77(2–3), 187-194. https://doi.org/10.1016/S0301-6226(02)00059-3.
9. Chen, S.; Bobe, G.; Zimmerman, S.; Hammond, E.G.; Luhman, C.M.; Boylston, T.D.; Freeman, A.E.; Beitz, D.C. Physical and sensory properties of dairy products from cows with various milk fatty acid compositions. *J Agric Food Chem*. **2004**, 52(11), 3422-3428. doi: 10.1021/jf035193z.
10. Kowalska, D.; Kostecka, M.; Tarnowska, K.; Kowalski, B. Oxidative stabilities of enzymatically interesterified goose fat and rapeseed oil blend by rancimat and PDSC. *J Therm Anal Calorim* **2014**, 115, 2063–2070. https://doi.org/10.1007/s10973-013-3125-0
11. Symoniuk, E.; Ratusz, K.; Krygier, K. Comparison of the oxidative stability of cold-pressed rapeseed oil using Pressure Differential Scanning Calorimetry and Rancimat methods. *Eur. J. Lipid Sci. Technol.* **2017**, 119, 1600182. https://doi.org/10.1002/ejlt.201600182
12. de Mello, B.T.F.; Iwassa, I.J.; Cuco, R.P.; dos Santos Garcia, V.A; da Silva, C. Methyl acetate as solvent in pressurized liquid extraction of crambe seed oil. *J Supercrit Fluids* **2019**, 145, 66-73. https://doi.org/10.1016/j.supflu.2018.11.024
13. Chen, J.; Liu, H. Nutritional Indices for Assessing Fatty Acids: A Mini-Review. *Int. J. Mol. Sci.* **2020**, *21*, 5695. https://doi.org/10.3390/ijms21165695
14. Bederska-Łojewska, D.; Pieszka, M.; Marzec, A.; Rudzińska, M.; Grygier, A.; Siger, A.; Cieślik-Boczula, K.; Orczewska-Dudek, S.; Migdał, W. Physicochemical Properties, Fatty Acid Composition, Volatile Compounds of Blueberries, Cranberries, Raspberries, and Cuckooflower Seeds Obtained Using Sonication Method. *Molecules* **2021**, 26, 7446. https://doi.org/10.3390/molecules26247446
15. Pieszka, M.; Migdał, W.; Gąsior, R.; Rudzińska, M.; Bederska-Łojewska, D.; Pieszka, M.; Szczurek, P. Native Oils from Apple, Blackcurrant, Raspberry, and Strawberry Seeds as a Source of Polyenoic Fatty Acids, Tocochromanols, and Phytosterols: A Health Implication. *J. Chem.* **2015**, 2015, 1-8. https://doi.org/10.1155/2015/659541