Agronomy Research **16**(4), 1781–1795, 2018
https://doi.org/10.15159/AR.18.164

The influence of drying method to the changes of bioactive compounds in lingonberry by-products

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Abstract. The aim of this research was to evaluate the effect of different drying methods on industrially obtained lingonberry pulp juice by-products. For investigations, by-product was dried using hot air dryer (at temperatures 80 °C, 60 °C and 40 °C), microwave-vacuum dryer and freeze-dryer. The freshly defrosted by-product was used as control. All samples were analysed on the basis of their moisture content, colourimetric attributes (CIE L*a*b* colour system), content of vitamin C (iodometric method), content of total carotenes (TC), total phenol content (TPC), total anthocyanin content (TA) and antiradical activity (DPPH, $ABTS^+$).
The obtained data on content of vitamin C showed a 10% decrease between control and sample dried in hot air at 80 °C. Similar changes were noticed with total anthocyanin content, the lowest amount was also foun in this sample (306.72 ± 18.32 mg 100 g^{-1} DW). Total carotenes content was higher in freeze-dried sample (5.61 ± 0.16 mg 100 g^{-1} DW) which was very close to control sample. A significant loss of total phenol content was noticed after drying, up to 80%, similar tendencies were noticed with changes of antiradical activity. After evaluating the obtained data, the use of hot air dryer at 80 °C temperature resulted in the lowest amount of vitamin C and anthocyanins in lingonberry by-products, which makes this method unsuitable for drying by-products of these berries. However, vacuum assisted microwave drying and freeze drying showed less damaging impact on dried material.

Key words: air drying, antioxidant activity, freeze-drying, microwave-vacuum drying, *Vaccinium vitis-idaea.*

INTRODUCTION

Lingonberry (*Vaccinium vitis-idaea L.*), also known in English as partridgeberry, is a wild, perennial, evergreen dwarf shrub that has red edible berries. Berries from this plant are commonly used in Europe, especially the Northern hemisphere (Alam et al., 2016; Alam et al., 2018). These berries have been considered to have health promoting properties, due to their biochemical content. Lingonberries are a great source of different phenolic substances, vitamins and minerals (Kivimäki et al., 2012; Kivimäki et al., 2013). Mainly their health benefits have been connected to the rich content of polyphenols that are secondary metabolites in plants that consist of one or more aromatic rings and hydroxyl groups. Polyphenols in plants and also human body are important, because they protect them from external factors such as pathogens and UV-radiation. Regular intake of dietary flavonoids can decrease the risk factors of cardiovascular

diseases and low-grade inflammation, polyphenols also are known to have positive effects on glucose homeostasis and they inhibit activation of platelets (Kivimäki et al., 2011; Kivimäki et al., 2012; Kivimäki et al., 2013). Also it needs to be taken into consideration that environmental factors like growth place, light, humidity, UV radiation and post-harvest processing (storing, pressing, pasteurization, drying, etc.) are all contributing factors to polyphenolic content of berries (Kivimäki et al., 2012).

In Latvia, lingonberries are consumed in several ways, they can be eaten fresh, frozen and stored for winter, used as decorative element in confectionery, jams, jellies, alcoholic and non-alcoholic drinks. However, for wider use, these berries have a very intense sour and bitter taste, due to the high amounts of organic acids, especially citric acid, which leads to a pH below 4. There is also a high amount of sugars, but the sweetness is masked by acids (Viljanen et al., 2014).

In food manufacturing the use of raw materials, including lingonberries as sufficiently as possible, is a very topical issue. There is a growing interest on creating new products from fruit waste products, which allows to utilize as much of the raw material as possible. One of the ways to process vegetable, fruit and berry by-poducts is to dry them. There are several food drying methods that could be potentially useful, however in this paper we will compare hot air drying (HAD), vacuum freeze-drying (VFD) and microwave-vacuum drying (MWVD).

Food stuff drying is one of the oldest and most commonly known food processing methods that is based on evaporation of free and loosely-bound water from inside the solid material into atmosphere. One of the most known and widely used drying methods is hot-air drying (HAD) or convective drying. In this method, hot air, that has low relative humidity, meets the surface of drying material that transfers t inside the product primarily by conduction. Then the liquid migrates onto the material surface and is transported away by air convection (Karam et al., 2016).

An alternative drying method to HAD is lyophilisation or freeze-drying (FD), this is a gentle dehydration method, which produces high-value dried products. This method is appealing because of its ability to maintain product colour, shape, aroma, nutritional value and overall quality. FD has two steps, in the first step product is frozen (-20 °C) and in the second step a controlled amount of heat (from -2 °C to -10 °C) under vacuum (VFD) or at atmospheric pressure (atmospheric freeze-drying) is applied to promote sublimation i.e. water change from solid to gas without passing through the liquid phase (Karam et al., 2016). Freeze drying can be carried out under vacuum, then heat will be supplied by surface heat exchange if required and evaporated moisture condensed on cold surface, but in case of atmospheric freeze-drying the product will be dried with air.

Microwave-vacuum drying (MWVD) is a fairly new method that has been introduced to replace hot-air drying. The basis of this method is microwave drying (MWD), where the first stage of drying is a heating-up period in which microwave energy is converted into thermal energy inside the moist product and the product temperature increases with time. The second stage is a called a rapid drying period during which thermal energy is used for moisture evaporization and transfer. The last stage is a reduced drying rate period where the local moisture is reduced to a point that the energy needed for moisture vaporization is lower than the thermal energy induced by microwaves. Applying microwave energy under vacuum is suitable for heat-sensitive products like fruits, vegetables and berries. This is also a drying method with efficiency of more energy and improved product quality (Karam et al., 2016).

All three drying methods for this study were chosen for several reasons. HAD was chosen, because this is a very traditional and widely used drying method. VFD is much newer approach and in many other studies, VFD has been proven to be less damaging towards food stuff biochemical content and sensory qualities. MWVD is also a rather new drying method, but mostly gained attention because of its short product drying time.

However, there is still little information found in literature about the influence of different drying techniques to the biochemical composition and quality of concrete products. Therefore, the aim of this research was to evaluate the effect of different drying methods on industrially obtained lingonberry pulp juice by-products. For this research the obtained by-product pulp was dried using HAD (80 °C, 60 °C, 40 °C), VFD and MWVD. To have more homogeneous samples, the dried samples were made into powders before analysis on moisture content, colour, and content of vitamin C, total carotenes, phenols, anthocyanins and antiradical scavenging activity. A frozen by-product (skin, seeds, etc.) was used as control.

MATERIAL AND METHODS

Sample preparation

Lingonberry (*Vaccinium vitis-idaea*) by-product as pulp was obtained from a local Latvian fruit, berry and vegetable product manufacturer Ltd 'KEEFA' in September 2017, but the by-product drying and analysing was done in November 2017 in Latvia University of Agriculture, Faculty of Food technology, Jelgava. The overall research plan can be seen in Fig. 1.

Lingonberry by-product (seeds, skin etc.) was obtained from industrially prepared lingonberry pulp juice as shown in Fig. 1 and stored frozen at -20 ± 2 °C for 1 month before drying and further testing.

For by-product drying, the obtained material was defrosted and divided into six parts. To each part a different drying method was applied and one was left as a control sample for comparison of data. Depending on drying method, abbreviations were used to identify all of the samples that can be seen in Table 1.

Table 1. The identification of samples

Sample name	Drying method
Control	no drying applied (defrosted lingonberry by-products)
HAD80	hot air drying at 80 °C
HAD60	hot air drying at 60 °C
HAD40	hot air drying at 40 °C
VFD	vacuum freeze-drying
MWVD	microwave-vacuum drying

Hot air drying (HAD) of by-products was done using 'Memmert' Universal Oven UF55 (Memmert, Germany) where by-products were dried at 80 °C for 6 h and 60 °C for 12 h and UF160 (Memmert, Germany) by-products dried at 40 °C for 16 h. The end point for product drying was based on personal experience, touch and visual product observation. By-product drying was stopped when material reached the appropriate degree of dryness for the production of powder.

Vacuum freeze-drying (VFD) was done using vacuum freeze-dryer FT33 (Armfield Ltd, Ringwood England). Sample was frozen in air freezer at -22 ± 2 °C temperature before drying. The temperature in condensation chamber was up to -40 °C and the pressure 20 Pa, drying time was up to 48 h.

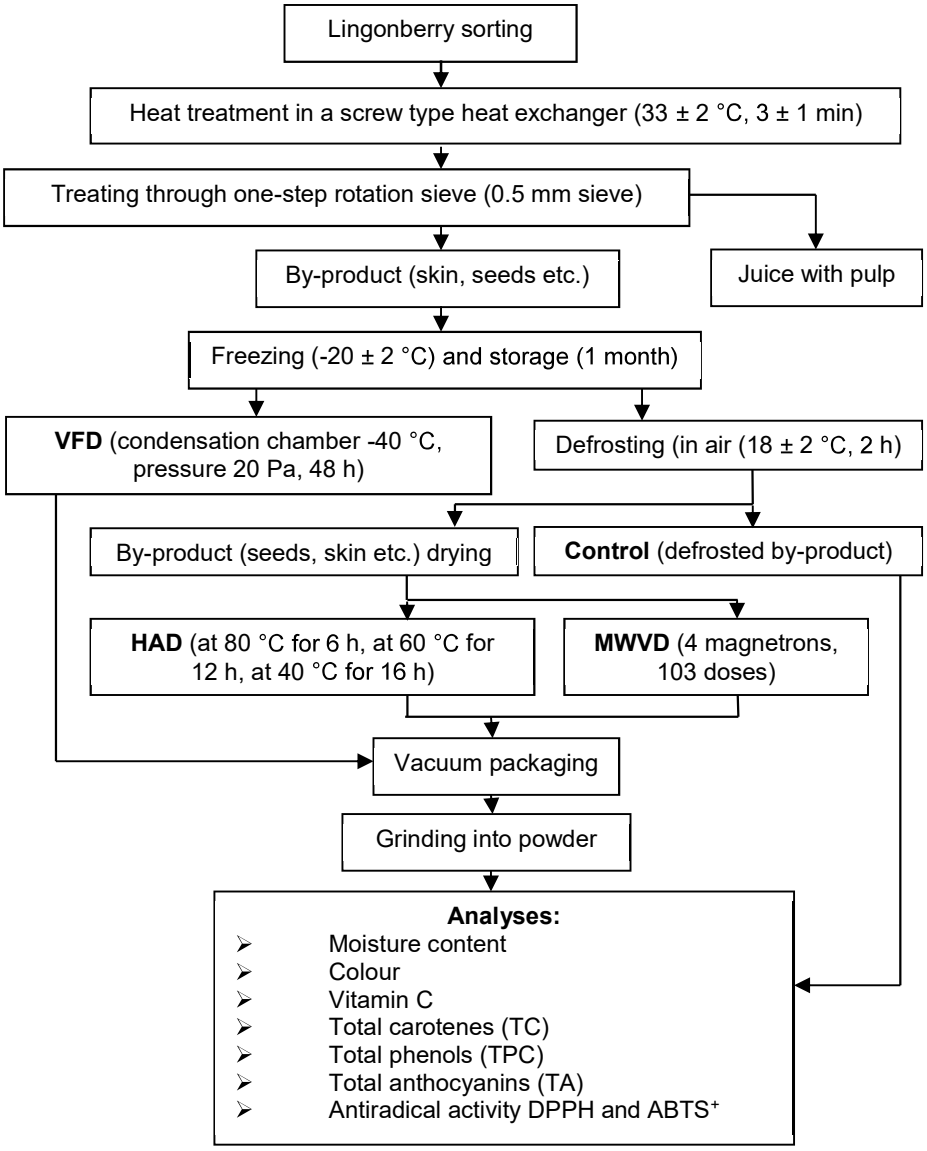

Figure 1. Overall research plan.

Microwave-vacuum drying (MWVD) was done using 'Musson-1' (Ingredient, Russia). The necessary amount of microwave energy (magnetron minutes) was calculated using empirical formulas when the initial moisture of the product is known (78%) and the final is estimated (8%) (Dorofejeva et al., 2011). For sample drying one cycle with four stages was carried out. The number of magnetrons was decreased along the drying process with subsequent doses, starting at 4 magnetrons and 50 doses, following by 3 magnetrons and 30 doses, 2 magnetrons and 13 doses and finally 1 magnetron 10 doses. The pressure in dryer was from 7.466 kPa to 9.332 kPa, cylinder rotation speed was 6 rpm.

All dried samples were vacuum-packaged to protect from air moisture absorption and for analysing purposes milled into powder to create a more homogenous sample. For milling electric Knifetec Mill 'Foss' (FOSS Analytical AB, Sweden) was used, samples were milled for 10 s and kept in LDPE zipped bags. A frozen by-product pulp was used as control and all samples were analysed on their moisture content, colour, and content of vitamin C, total carotenes, total phenol content, total anthocyanins and antiradical activity.

Chemicals and reagents

The chemical analyses of tested samples were carried using the following chemicals and reagents. Oxalic acid dehydrate (126.07 g mol^{-1}), L(+)-Ascorbic acid (176.13 g mol^{-1}), Petroleum ether 80/110, Potassium dichromate, Hydrochloric acid 35-38% (36.46 g mol^{-1}), Sodium carbonate anhydrous (105.99 g mol^{-1}), Potassium persulfate (270.33 g mol^{-1}), Potassium chloride (74.56 g mol^{-1}), Sodium chloride (58.44 g mol^{-1}), di-Sodium hydrogen phosphate anhydrous (141.96 g mol^{-1}), Potassium phosphate monobasic (136.09 g mol^{-1}) and Sodium hydroxide 0.1 N (40.00 g mol^{-1}) were purchased from Chempur (Poland). Iodine concentrate (0.05 mol L^{-1}) FIXANAL was obtained from Fluka Analytical, Sigma-Aldrich (Poland), Starch powder, soluble, ACS (for iodometry) was purchased from Alfa Aesara GmbH & Co KG (Germany). 2,2-Diphenyl-1-picrylhydrazyl was purchased from Sigma-Aldric (Germany), (±)-6-Hydroxil-2,5,7,8-tetra -methylchfomane-2-carboxylic acid was purchased from Sigma-Aldrich (Russian Federation). 2,2'-Azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt ≥ 98% (HPLC) was purchased from Sigma-Aldrich (China). Folin-Ciocalteu, phenol reagent was obtained from Scharlau (Spain), Gallic acid, 98% was purchased from Acros Organics (Belgium). Additional solvents as ethanol (96%) and ultra pure water were provided by Latvia University of Agriculture (Latvia, Jelgava).

Determination of moisture content

Content of moisture in all samples was determined accordingly to standard ISO 6496:1999. For moisture content detection to control (defrosted) sample approximately 5 g was used, for dry samples 2 g of milled sample each in two replications was dried using 'Memmert' Universal Oven UF55, at 105 °C periodically checking the weight changes during drying process by weighing the sample until minimal changes can be observed. Weight loss was used to calculate the moisture content of the sample.

Determination of vitamin C

Content of vitamin C was determined according to the iodine method as described by Kerch et al. (2011) with some modifications. This method is based on determination of L-ascorbic acid, the reduced form of ascorbic acid. A sample of 25 g for defrosted sample and 5 g for dried, milled samples were poured with 100 mL of 6% solution of oxalic acid, homogenized for 1 min, and filtered. Then, 2 mL of 1% solution of starch was added to 10 mL of filtrate and the filtrate was titrated with 0.05 n iodine solution until change of colour, which does not disappear during 30 s. For standard solution of ascorbic acid, 20 mg of ascorbic acid was dissolved in 100 mL of the same 6% oxalic acid solution, 2 mL of the same 1% starch solution was added to 25 mL of the standard

solution and the mixture was titrated. The content of ascorbic acid expressed in milligrams per 100 g DW of product was calculated using Eq. (1), on the basis of four replications (Kerch et al., 2011; Ozola at al., 2017):

$$C = 5{,}000 \cdot \frac{V_{sample}}{m \cdot V_{standard}} \quad (1)$$

where V_{sample} – volume of the iodine solution titrated in a sample, mL; $V_{standard}$ – volume of the iodine solution titrated in a standard solution, mL; m – the amount of sample, g.

Determination of total carotenes

For total carotene determination, a spectrophotometric (UV/VIS spectrophotometer Jenway 6705 (Bibby Scientific Ltd., UK)) method described by Kampuse et al. (2015) with modifications was used. A sample of 2 g for control and 1 g for dried, milled samples was mixed with 20 mL of ethanol and mixed on a magnetic stirrer for 15 min. Further 10 mL of pure water was added to dried samples, to ensure layer formation, and stirred for 10 min after which, 25 mL of petroleum ether was added and stirred for an hour. The absorption of oil layer was measured at 440 nm. The content of TC expressed in milligrams per 100 g^{-1} DW was calculated in three replications (Ozola et al., 2017).

Determination of total anthocyanins (TA)

Total anthocyanin content was determined by spectrophotometric method described by Kerch et al. (2011) with some modifications. A sample of 5 g for control and 1 g for dry samples was mixed with 40 g of ethanol and 1.5 M HCl solution (85 : 15 by volume) and homogenized for 1 min. Then the sample was filtered, and filtrate volume measured. TA where detected on spectrophotometer Jenway 6705 at 540 nm. The sample was diluted until absorption was between 0.6 and 0.8. The content of TA expressed in milligrams per 100 g^{-1} DW was calculated with the Eq. (2).

$$C = \frac{A \cdot v \cdot d \cdot 1{,}000}{980 \cdot m} \quad (2)$$

where A – absorbance, expressed as absorbance units; v – volume of filtrate, mL; d – dilution degree; m – sample weight, g.

Measurements were carried out in two replications (Ozola et al., 2017).

Determination of total phenol content

The detection of total phenol content was achieved according to the Folin-Ciocalteu method (Yu et al., 2003) with modifications. A sample of 2 g for control and 1 g for dry samples was used for extract preparation. To 0.5 mL of extracted sample 2.5 mL of 0.2 N Folin-Ciocalteu reagent that has been diluted ten times with pure water was added. After 5 min 2.0 mL of 7.5% $NaCO_3$ was added, the resulting solution was mixed and allowed to stand for 30 min at 18 ± 1 °C in a dark place (Priecina & Karklina, 2014). Absorption was read at 765 nm using JENWAY 6300 (Banoworld Scientific Ltf., UK) spectrophotometer. Measurements were carried out in six replications from two separately weighed samples, the obtained data were expressed as gallic acid mg equivalent (GAE mg $100g^{-1}$ dry sample) (Ozola et al., 2017).

Determination of antiradical activity (DPPH) and radical scavenging activity ($ABTS^+$)

The antiradical activity of extracts was measured on the basis of scavenging activities of the stable 2,2-diphenil-1-picrylhydrazyl-(DPPH) free radical (Yu et al., 2003) with modifications. To 0.5 mL of extracted sample, 3.5 mL freshly made DPPH solution was added; the mixture was shaken and kept in the dark place at 18 ± 1 °C for 30 min; absorbance was measured at 517 nm using JENWAY 6300 spectrophotometer, measurements were carried out in six replications from two separately weighed samples. For the quantitative expression of antiradical activity, the Trolox equivalent of 6-Hydroxil-2,5,7,8-tetra-methylchfomane-2-carboxylic acid was used. A Trolox calibration curve was created and, using the red-out absorbance, the antiradical activity was expressed as mg Trolox 100 g^{-1} per dry sample (Priecina & Karklina, 2014; Ozola et al., 2017).

The radical scavenging activity of extracts was also measured by $ABTS^+$ radical cation assay as described by Re et al. (1999). A stock solution of 2,2′-azino-bis (3-ethylbenz-thiazoline-6-sulfonic) acid (ABTS) (2 mM) was prepared by dissolving in 50 mL of phosphate buffered saline (PBS) obtained by dissolving 8.18 g sodium chloride (NaCl), 0.27 g potassium dihydrogen phosphate (KH_2PO_4), 1.42 g sodium phosphate dibasic (Na_2HPO_4) and 0.15 g potassium chloride (KCl) in 1 L of ultra pure water. If the pH was lower than 7.4, it was adjusted with sodium hydroxide (NaOH). Ultra pure water was used to prepare 70 mM solution of potassium persulfate ($K_2S_2O_8$). $ABTS^+$ radical cation was produced by reacting 50 mL of ABTS stock solution with 0.2 mL of $K_2S_2O_8$ solution and allowing the mixture to stand in the dark at room temperature (18 ± 1 °C) for 15–16 h before use. The $ABTS^+$ radical was stable in this form for more than 2 days when stored in these conditions. For the assessment of extracts, the $ABTS^+$ solution was diluted with PBS to obtain an absorbance of 0.800 ± 0.030 at 734 nm. 5 mL of $ABTS^+$ solution were mixed with 0.05 mL of extract. The absorbance was read at ambient temperature (18±1 °C) after 10 min. PBS solution was used as a blank sample. For both types of antiradical scavenging activity determination, the obtained data were expressed as millimolar Trolox equivalents per 100 g^{-1} DW of sample (Tomsone et al., 2012).

Colour analysis

Sample colour was determined using colour analyser *ColorTec-PMC* (Accuracy Microsensors, Inc, New York, USA) that captures the reflected light of the object. In this study the L*a*b* colour system was used, developed by CIE (*International Commission for Lighting and Ilumination*) 1976 version (Mokrzycki & Tatol, 2011).

The CIE L*a*b* system is based on opponent colour model: L* black/white, describes overall colour intensity, where 0 means black, and 100 is the maximum light intensity; +a*/-a* red/green; +b*/-b* -yellow/blue (Mokrzycki & Tatol, 2011).

In case of the L*a*b* space, the ΔE difference between two colours is calculated by Eq. (3):

$$\Delta E = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2} \quad (3)$$

where ΔE – absolute colour difference; ΔL^* – colour intensity difference; Δa^* – red and green colour difference; Δb^* – yellow and blue colour difference (Mokrzycki & Tatol, 2011). The measurements for colour analysis were carried out in five replications.

Statistical analysis

The obtained data was processed using 'Microsoft Office Excel' 2007 version, differences between the results were analysed using one-factor ANOVA followed by Tukey-Kramer method, The linear correlation analysis was performed to determine the relationship between TPC, DPPH and ABTS, and between colour analyses factor a* and total anthocyanin content. The obtained results were presented as their means with standard errors. Differences among results were considered to be significant if $P < 0.05$ (Ozola et al., 2017).

RESULTS AND DISCUSSION

Sample moisture content

The obtained data on dried lingonberry by-product moisture content showed that before drying on average it was 78.8% per 100 g of product (w/w), but after drying the least proportion of moisture was detected in hot air dried samples (HAD80 and HAD60 mainly), where it was approximately 1.8% (w/w). The moisture content of sample HAD40 on average was 3.8% (w/w), in sample that was vacuum freeze-dried (VFD) approximately 4.6% (w/w), but the highest moisture content 8.6% (w/w) was detected in sample MWVD. The detected moisture in samples show that by using HAD at higher temperatures such as 80 °C and 60 °C can create sample with lower moisture than all of the other tested drying methods. HAD is a much more invasive product drying method due to the higher temperatures and prolonged drying time (Karam et al., 2016). These conditions and the fact that the dried material was shredded might have influenced the higher water loss. This type of dried material has a larger surface, the cellular structure was mechanically damaged and therefore could increase the evaporation of water.

Content of vitamin C, total carotenes and total anthocyanins

The obtained data on content of vitamin C did not show any large differences overall, however significant differences between some samples were detected $P < 0.05$. In the control sample, there was 79.4 ± 2.5 mg 100 g^{-1} DW of vitamin C, which was very close to content of vitamin C in dried samples (Table 2). However significant differences were detected between HAD80 and all other samples. By-product hot air drying at 80 °C significantly decreased the amount of vitamin C in dried sample (72.1 ± 4.5 mg 100 g^{-1} DW). Although the vitamin C content had only decreased by 10%, these findings are consistent with other researcher data showing from 20% to 60% depletion of vitamin C for several agricultural materials (Karam et al., 2016). When evaluating other samples no significant difference were detected within the changes of vitamin C depending on drying method of lingonberry by-products. These findings are consistent when analysing the obtained data from freeze-drying and microwave drying treatments, but less common with hot air drying. However Ali et al. (2016) in their research have found that not only freeze-drying and microwave 100 W drying, but also oven dried at 80 °C guava fruit slices had retained a relatively high amount of vitamin C. Ali et al. (2016) concludes that drying temperature and drying time are key parameters which directly affect the vitamin C concentration in guava. Despite the fact that with lingonberry by-product hot air drying at 80 °C showed the highest decrease, it could be possible that in our case a lower temperature and prolonged drying time had less damaging effect on vitamin C content during drying at 40 °C and 60 °C.

When analysing the data on TC and TA, some differences were noticed in comparison to content of vitamin C. The highest total carotene (Table 2) content was in control sample (5.83 ± 0.24 mg 100 g^{-1} DW), but after drying a significant decrease ($P < 0.05$) was noticed for all drying methods except for vacuum freeze-drying (5.61 ± 0.16 mg 100 g^{-1} DW). HAD at 80 °C showed a TC decrease of 19%, but at 40 °C approximately 23% and on average 33% decrease in total carotene content was in sample HAD60 and MWVD.

Although by-product drying at 80 °C showed a significantly higher ($P < 0.05$) TC content then drying at 60 °C no supporting literature was found to fully explain these results. Moreover the statistical analyses showed no significant differences between HAD80 and HAD40, and between HAD60 and HAD40 (Table 2.)

Table 2. Content of vitamin C, TC and TA in lingonberry by-product powders

Sample	Vitamin C (mg 100 g^{-1} DW)	Total carotenes (mg 100 g^{-1} DW)	Total anthocyanins (mg 100 g^{-1} DW)
Control	79.4 ± 2.5 ^{abcd}	5.83 ± 0.24 ^{a}	566.7 ± 24.4
HAD80	72.1 ± 4.5	4.68 ± 0.34 ^{b}	306.7 ± 18.3 ^{a}
HAD60	79.3 ± 2.5 ^{aefg}	3.94 ± 0.17 ^{cd}	343.0 ± 14.5 ^{abc}
HAD40	84.9 ± 2.9 ^{behj}	4.41 ± 0.17 ^{bce}	387.5 ± 5.0 ^{bde}
MWVD	85.5 ± 2.9 ^{cfhk}	3.95 ± 0.29 ^{de}	412.8 ± 7.2 ^{df}
VFD	84.0 ± 3.3 ^{dgjk}	5.61 ± 0.16 ^{a}	396.2 ± 3.4 ^{cef}

Mean values in each column followed by different letters do not have a significant difference according to Tukey-Kramer method ($P < 0.05$).

Considering the difference in sample preparation before testing could explain the lack of significant difference in content of vitamin C and TC. Control was more of a heterogeneous mass in comparison to dried materials, which were pulverized and thus making them more of a homogenous sample therefore depicting the content of vitamin C and TC in analysed samples more precisely. In addition, control did not have any pre-treatment before analysing so this could allow for further enzymatic activity continuing decreasing the amount of vitamin C until sample was analysed.

The content of total anthocyanins (Table 2) also showed a significant decrease after drying ($P < 0.05$). The highest content of TA after drying was in sample MWVD (412.8 ± 7.2 mg 100 g^{-1} DW) which was 27% less than in control sample. Also a relatively high content of TA was found in samples HAD40 (387.5 ± 5.0 mg 100 g^{-1} DW) and VFD (396.2 ± 3.4 mg 100 g^{-1} DW) showing no significant difference $P < 0.05$, corresponding to 45% decrease in sample HAD80 (306.7 ± 18.3 mg 100 g^{-1} DW).

When comparing the obtained data with literature it has been observed that depending on the product there have been found occasions where carotenoid compounds (lycopen in particularly) were heat-stable, even after severe heat treatment, but in other author researches around 19% of TC decrease had been noticed after air-dried carrot slices, paprika and sweet potatoes (Karam et al., 2016). Not only vitamin C and TC, but also total anthocyanins are highly unstable compounds and changes of these compounds can differ from the product to which drying has been applied. Drying at low temperature might not be effective to inactivate enzyme or might take longer time to inactivate the enzymes which are responsible for the anti-oxidative properties such as ascorbic acid,

carotenoid and TPC degradation (Sehrawat et al., 2018), and this could potentially explain the TC changes in lingonberry-by product when HAD was applied.

When looking at the obtained data Table 1 it can be clearly seen that there is a tendency of TA decrease when the drying temperature increases (Ruse et al., 2011). Overall Sehrawat et al. (2018) when drying mango cubes detected higher bioactive compound retention. Oxygen deficient environment had provided a better protective effect form oxidative loss of bioactive compounds as compared to HAD (Sehrawat et al., 2018), these findings are also applyable with our research on lingonberry by-product drying.

Overall according to the obtained data hot air drying at 80 °C had the most effect on degradation of vitamin C and total anthocyanin content. The most suitable drying method for beather retention of TC proved to be VFD, but lingonberry by-product MWVD showed the highest content of vitamin C and TA.

Total phenol content, DPPH and ABTS$^+$

The lingonberry by-product powder TPC as shown in Fig. 2. in control sample was very high but after drying the obtained data showed an average decrease of 80% with no significant differences between the used drying methods ($P < 0.05$).

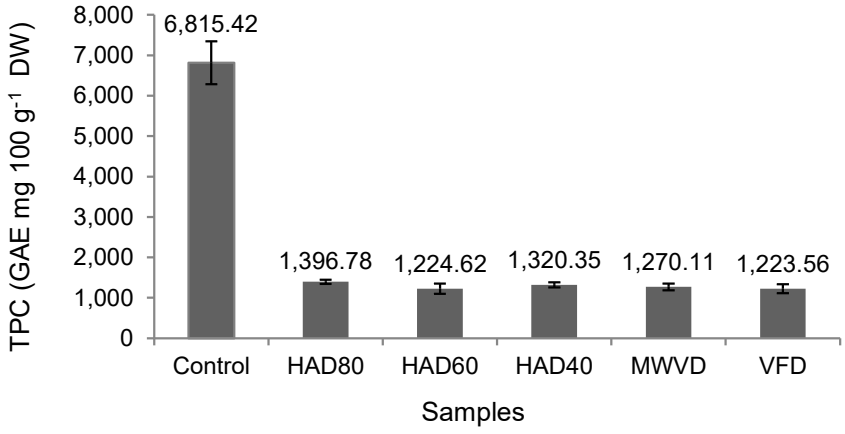

Figure 2. Total phenol content in lingonberry by-product samples (GAE mg 100 g^{-1} DW).

Similar tendencies to TPC were also noticed with antiradical activity, supported by Pearson's coefficient showed a strong correlation $r = 0.9977$ between TPC and DPPH. Also a strong correlation of $r = 0.8585$ was found between TPC and ABTS$^+$, and DPPH and ABTS$^+$ ($r = 0.8628$).

Overall a 60% decrease in antiradical activity was found in all lingonberry-by product powders (Table 2) compared to control ($P < 0.05$), this was also significantly evidenced from ABTS$^+$. Slight difference in DPPH was found between HAD80 (232.67 ± 12.30 mM TE 100g^{-1} DW) and HAD60 (210.14 ± 13.21 mM TE 100g^{-1} DW).

The lowest radical scavenging activity was detected in hot air dried samples (HAD80; HAD60 and HAD40). In sample HAD80 radical scavenging activity had significantly decreased in comparison with control sample, where it was 274.76 ± 12.38 mM TE 100 g^{-1} DW (Table 3). However vacuum freeze-drying showed

to be less invasive (215.99 ± 8.06 mM TE 100 g^{-1} DW). Although strong correlation was found between DPPH and $ABTS^+$, the radical scavenging activity showed more significant differences between dried samples in comparison to DPPH (Table 2). No real differences ($P > 0.05$) between samples were found when comparing HAD80 to HAD60, also between HAD60 and HAD40, HAD40 and MWVD, and MWVD compared to FVD.

Table 3. Content of DPPH and $ABTS^+$ in lingonberry by-product powders

Sample	DPPH (mM TE 100g^{-1} DW)	$ABTS^+$ (mM TE 100g^{-1} DW)
Control	551.96 ±20.67	274.76 ± 12.38
HAD80	232.67 ± 12.30 ^{abc}	153.99 ± 12.17 ^{a}
HAD60	210.14 ± 13.21 ^{adef}	170.37 ± 16.57 ^{ab}
HAD40	205.53 ± 8.56 ^{dgh}	180.38 ± 17.70 ^{bc}
MWVD	227.71 ± 20.39 ^{begi}	195.98 ± 9.38 ^{cd}
VFD	220.04 ± 13.03 ^{cfhi}	215.99 ± 8.06 ^{d}

Mean values in each column followed by different letters do not have a significant difference according to Tukey-Kramer method ($P < 0.05$).

(Ek et al., 2006) in their research found 28 phenolic compounds, that included flavonols, anthocyanidins, catechins and their glycosides, and different caffeoyl and ferulic acid conjugates. Although it was not expected from this study to detect such high depletion of TPC after drying it has been reported before that polyphenolics are heat sensitive and prolonged heat treatment causes irreversible chemical changes to phenol content (Guiné et al., 2015). Other contributing factors may include polyphenol binding with other compounds, alterations in their chemical structure, activity of polyphenol ocidase, organic acid content, sugar content and product pH (Guiné et al., 2015).

When evaluating antioxidant activity with DPPH and $ABTS^+$ in plant materials it is possible to obtain varying results. This happens, because of their different working mechanisms of the two assays (Shalaby & Shanab, 2013). DPPH is a stable free radical with an absorption band at 515 nm that is lost when reduced by an antioxidant or a free radical species. But $ABTS^+$ assay measures the relative ability of antioxidant to scavenge the ABTS generated in aqueous phase, as compared with a Trolox standard (Shalaby & Shanab, 2013).

$ABTS^+$ is also known to be useful to determine antioxidant activity of both lipophilic (α-tocopherol, β-carotene) and hydrophilic antioxidants in various matrices. Also reacting rapidly with antioxidants over a wipe pH. However DPPH interactions between antioxidants are also determied by theyr structural conformation and reaction time with DPPH (Martysiak-Żurowska & Wenta, 2012).

In a different study by Michalska et al. (2017), it was also noticed that prolonged exposure to oxygen can considerably affect the compound radical scavenging activity with assay $ABTS^+$, which was noticed when comparing, HAD samples with MWVD and VFD samples where samples were dried in assistance of vacuum. Also Correia et al. (2017) showed that an increase in drying temperature is deleterious to product TPC and antioxidant activity.

Colour analysis

Dried lingonbery by-products were mild into powder and their colourimetric properties were measured (Fig 3.) The obtained data on dried powder analysis showed that there is a slight significant difference in L* factor value and therefore sample MWVD was darker in comparison to HAD80 and VFD. When calculating hue angle values and placing them on CIE L*a*b* 1976 colour wheel the dominant colour for all samples was light red.

The distinct red colour in lingonberries is created by anthocyanins, however compairing lingonberry-by product powder colour analysis factor a* value and total anthocyanin content showed a low correlation $r = 0.382$.

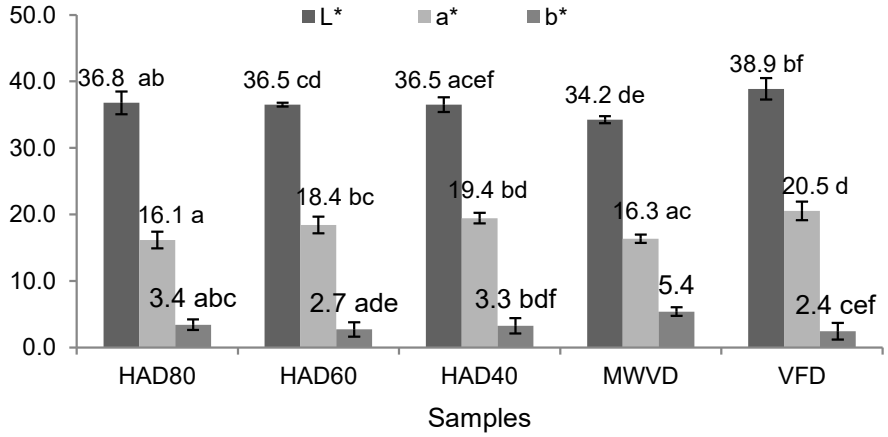

Figure 3. Dried lingonbery by-product colour analysis (CIE L*a*b). Mean values in each row followed by different letters do not have a significant difference according to Tukey-Kramer method ($P < 0.05$).

Mokrzycki & Tatol (2011) described how mathematically it would be possible to determine colour perception and analyse if two samples are different in colour and whether human eyes are able to distinguish them. In CIE L*a*b* system the perceptual colour difference can be determined by ΔE value (Table 4.) where:

$0 < \Delta E < 1$ the observer does not notice the difference;

$1 < \Delta E < 2$ only experienced observer can notice the difference;

$2 < \Delta E < 3.5$ inexperienced observer also notices difference;

$3.5 < \Delta E < 5$ there is a clear difference in colour and it is noticeable;

$5 < \Delta E$ observer notices two different colours (Mokrzycki & Tatol, 2011).

Table 4. Dried lingonberry by-product powder absolute colour difference

Sample	ΔE	ΔE	ΔE	ΔE
HAD80	ΔE with HAD80			
HAD60	2.4	ΔE with HAD60		
HAD40	3.3	1.2	ΔE with HAD40	
MWVD	3.2	4.1	4.4	ΔE with MWVD
VFD	5.0	3.2	2.7	4.7

After calculating ΔE and evaluating colour differences between each sample, the results can be seen in Table 4. The results show that there is no noticale difference between sample HAD60 and HAD40 colour, because absolute colour ddifference between both ΔE values is 1.2. Nevertheless, there is a clear and noticeable difference between HAD80 and VFD also between samples HAD60 and MWVD, HAD40 and MWVD, MWVD and VFD, because $3.5 < \Delta E < 5$. The data coincide with information given in Fig. 3, where microwave-vacuum dried sample is noticeably darker and VFD lighter than other samples.

CONCLUSIONS

In conclusion, of this research it is very difficult to unambiguously advice the most suitable drying method for drying lingonberry pulp juice by-products due to the inconsistencies of the results and lack of significant differences between the used drying methods.

By-product drying at 80 °C coused the highest degradation of vitamin C an total anthocyanin content. More suitable drying methods where vacuum assisted microwave drying and freeze-drying that resulted in better retention of total carotenes, total anthocyanins and vitamin C content.

After by-product drying an 80% degradation of total phenol content was detected which strongly correlated with antiradical activity (DPPH) $r = 0.9977$ and radical scavenging activity ($ABTS^+$) $r = 0.8585$. Because phenols are heat sensitive, drying might have caused changes in their chemical structure, organic acid, sugar content, pH etc. therefore reducing TPC in dried samples, however no significant differences were detected between dried samples ($P > 0.05$).

Vacuum assisted drying methods also showed slightly better retention of DPPH and $ABTS^+$. Similarly, to TPC sample, antiradical activity showed no significant differences between used drying methods, but the highest radical scavenging activity was determined in VFD in contrary the lowest was found in HAD80, however no significant difference was found when compared to HAD60.

In reference to the findings it is possible to say that hot air drying at 80 °C is not the most suitable method for lingonberry by-product drying, however vacuum assisted freeze-drying could be suggested due to the highest retention of vitamin C, total carotenes and by DPPH and $ABTS^+$.

ACKNOWLEDGEMENTS. The research was financed by program 'Scientific Capacity Building LUA' project A05-06 'Development of special dietary foods with high bioavailability'.

REFERENCES

Alam, Z., Morales, H.R. & Roncal, J. 2016. Environmental conditions affect phenolic content and antioxidant capacity of leaves and fruits in wild partridgeberry (*Vaccinium vitis-idaea*). *Botany* **13**, 509–521.

Alam, Z., Roncal, J. & Peña-Castillo, L. 2018. Genetic variation associated with healthy traits and environmental conditions in *Vaccinium vitis-idaea*. *BMC Genomics* **19**(1), 1–13.

Ali, M.A., Yusof, Y.A., Chin, N.L. & Ibrahim, M.N. 2016. Effect of different drying treatments on colour quality and ascorbic acid concentration of guava fruit. *International Food Research Journal* **23**, S155–S161.

Color Models CIELAB. *http://dba.med.sc.edu/price/irf/Adobe_tg/models/cielab.html.* Accessed 14.1.2018

Correia, P.M.R., Guiné, R.P.F., Correia, A.C., Brito, M.F.S., Ribeiro, R.P. 2017. Physical , chemical and sensory properties of kiwi as influenced by drying conditions. *Agricultural Engineering International: CIGR Journal* **19**(3), 203–212.

Dorofejeva, K., Rakcejeva, T., Galoburda, R., Dukalska, L. & Kviesis, J. 2011. Vitamin C content in Latvian cranberries dried in convective and microwave vacuum driers. *Procedia Food Science* **1**, 433–440.

Ek, S., Kartimo, H., Mattila, S. & Tolonen, A. 2006. Characterization of Phenolic Compounds from Lingonberry (*Vaccinium vitis-idaea*). *Journal of Agricultural and Food Chemistry* **54**(26), 9834–9842.

Guiné, R.P.F., Barroca, M.J., Gonçalves, F.J., Alves, M., Oliveira, S. & Correia, P.M.R. 2015. Effect of Drying on Total Phenolic Compounds, Antioxidant Activity, and Kinetics Decay in Pears. *International Journal of Fruit Science* **15**(2), 173–186.

ISO 6496:1999. Animal feeding stuffs – determination of moisture and other volatile matter content. *https://law.resource.org/pub/in/bis/S06/is.iso.6496.1999.pdf*. Accessed 14.1.2018

Kampuse, S., Ozola, L., Straumite, E. & Galoburda, R. 2015. Quality parameters of wheat bread enriched with pumpkin (*Cucurbita Moschata*) by-products. *Acta Universitatis Cibiniensis. Series E: Food Technology* **19**, 3–14.

Karam, M.C., Petit, J., Zimmer, D., Baudelaire Djantou, E. & Scher, J. 2016. Effects of drying and grinding in production of fruit and vegetable powders: A review. *Journal of Food Engineering* **188**, 32–49.

Kerch, G., Sabovics, M., Kruma, Z., Kampuse, S. & Straumite, E. 2011. Effect of chitosan and chitooligosaccharide on vitamin C and polyphenols contents in cherries and strawberries during refrigerated storage. *European Food Research and Technology* **233**, 351–358.

Kivimäki, A.S., Ehlers, P.I., Siltari, A., Turpeinen, A.M., Vapaatalo, H. & Korpela, R. 2012. Lingonberry, cranberry and blackcurrant juices affect mRNA expressions of inflammatory and atherothrombotic markers of SHR in a long-term treatment. *Journal of Functional Foods* **4**, 496–503.

Kivimäki, A.S., Ehlers, P.I., Turpeinen, A.M., Vapaatalo, H. & Korpela, R. 2011. Lingonberry juice improves endothelium-dependent vasodilatation of mesenteric arteries in spontaneously hypertensive rats in a long-term intervention. *Journal of Functional Foods* **3**, 267–274.

Kivimäki, A.S., Siltari, A., Ehlers, P.I., Korpela, R. & Vapaatalo, H. 2013. Lingonberry juice lowers blood pressure of spontaneously hypertensive rats (SHR). *Journal of Functional Foods* **5**, 1432–1440.

Martysiak-Żurowska, D. & Wenta, W. 2012. A comparison of abts and dpph methods for assessing the total antioxidant capacity of human milk. *Acta Sci. Pol. Technol. Aliment* **11**(1), 83–89.

Michalska, A., Wojdyło, A., Lech, K., Łysiak, G.P. & Figiel, A. 2017. Effect of different drying techniques on physical properties, total polyphenols and antioxidant capacity of blackcurrant pomace powders. *LWT - Food Science and Technology* **78**, 114–121.

Mokrzycki, W.S. & Tatol, M. 2011. Colour difference dE- A survey. *Machine Graphics and Vision* **20**, 383–411.

Ozola, L., Kampuse, S. & Galoburda, R. 2017. The effect of high pressure processing on enteral food made from fresh or semi-finished ingredients. In: *11th Baltic Conference on Food Science and Technology.* Foodbalts2017, Latvia, pp. 80–85.

Priecina, L. & Karklina, D. 2014. Natural antioxidant changes in fresh and dried spices and vegetables. *International Journal of Biological, Biomolecular, Agricultural, Food and Biotechnological Engineering* **8**, 492–496.

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M. & Rice-Evans, C. 1999 Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine* **26**, 1231–1237.

Ruse, K., Rakscejeva, T., Galoburda, R. & Dukalska, L. 2011. Anthocyanin content in Latvian cranberries dried in convective and microwave vacuum driers. *Food Sciences*, 100–106.

Sehrawat, R., Nema, P.K. & Kaur, B.P. 2018. Quality evaluation and drying characteristics of mango cubes dried using low-pressure superheated steam, vacuum and hot air drying methods. *LWT – Food Science and Technology* **92**, 548–555.

Shalaby, E.A. & Shanab, S.M.M. 2013. Comparison of DPPH and ABTS assays for determining antioxidant potential of water and methanol extracts of *Spirulina platensis*. *Indian Journal of Marine Sciences* **42**(5), 556–564.

Tomsone, L., Kruma, Z. & Galoburda, R. 2012. Comparison of different solvents and extraction methods for isolation of phenolic compounds from horseradish roots *Research for Rural Development* **4**(1), 236–241.

Viljanen, K., Heiniö, R.L., Juvonen, R., Kössö, T. & Puupponen-Pimiä, R. 2014. Relation of sensory perception with chemical composition of bioprocessed lingonberry. *Food Chemistry* **157**, 148–156.

Yu, L., Perret, J., Harris, M., Wilson, J. & Haley, S. 2003. Antioxidant Properties of Bran Extracts from 'Akron' Wheat Grown at Different Locations. *Journal of Agricultural and Food Chemistry* **51**, 1566–1570.