Influence of Domestic Processing and Storage on Flavonol Contents in Berries

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Effects of domestic processing and storage on the flavonols quercetin, myricetin, and kaempferol in five berries were studied using an optimized RP-HPLC method with UV and diode array detection after an acid hydrolysis of the corresponding glycosides. In fresh berries, the total content of flavonols was highest in lingonberry (169 mg/kg) and black currant (157 mg/kg), intermediate in bilberry (41 mg/kg) and strawberry (17 mg/kg), and lowest in red raspberry (9.5 mg/kg). Cooking strawberries with sugar to make jam resulted in minor losses (quercetin 15%, kaempferol 18%). During cooking of bilberries with water and sugar to make soup, 40% of quercetin was lost. Traditional preservation of crushed lingonberries in their own juice caused a considerable (40%) loss of quercetin. Only 15% of quercetin and 30% of myricetin present in unprocessed berries were retained in juices made by common domestic methods (steam-extracted black currant juice, unpasteurized lingonberry juice). Cold-pressing was superior to steam-extraction in extracting flavonols from black currants. During 9 months of storage at −20 °C, quercetin content decreased markedly (40%) in bilberries and lingonberries, but not in black currants or red raspberries. Myricetin and kaempferol were more susceptible than quercetin to losses during storage.

Keywords: *Flavonoid; flavonol; quercetin; myricetin; kaempferol; processing; storage; berry; fruit; HPLC*

INTRODUCTION

Epidemiological evidence suggests that high consumption of flavonol-rich foods may be protective against coronary heart disease (Hertog et al., 1993a, 1995, 1997; Knekt et al., 1996), stroke (Keli et al., 1996), lung cancer (Knekt et al., 1997), and stomach cancer (Garcia-Closas et al., 1999). Long-term health benefits associated with quercetin and other flavonols have raised the interest in the content of these flavonoids in various foods. High levels of quercetin have been found in onion, kale, cherry tomato, certain varieties of lettuce, cranberry, lingonberry, and blueberry (Hertog et al., 1992a,b; Crozier et al., 1997; Justesen et al., 1998). The level of kaempferol is high in kale, broccoli, and endive (Hertog et al., 1992b; Justesen et al., 1998; Price et al., 1998a), and that of myricetin in cranberry (Hertog et al., 1992a; Justesen et al., 1998). Our work on the bioactive phenolic compounds in berries has shown that many of them are rich in flavonols (Törrönen et al., 1997; Häkkinen and Auriola, 1998; Häkkinen et al. 1998, 1999a,b).

We recently reported the contents of quercetin, myricetin, and kaempferol in 25 wild or cultivated berries (Häkkinen et al., 1999b). Quercetin was present in all berries studied, and high contents were found, e.g., in bog whortleberry, lingonberry, cranberry, and crowberry. Myricetin was detected in six berries, with the highest content in cranberry and black currant. Kaempferol was detected in gooseberry and strawberry. Cranberry, bog whortleberry, lingonberry, and crowberry (wild berries) as well as black currant (cultivated) had total contents of these flavonols (100−263 mg/kg) higher than have the commonly consumed fruits or vegetables, with the exception of onion and kale.

In Finland, the season during which fresh berries are available is short, lasting from late June (strawberry) to October (cranberry). Therefore, only a small proportion of berries is consumed fresh, and most of the harvest is preserved by freezing or by processing to juices, jams, jellies, etc. There are no reports on the effects of processing and storage on the flavonol content of berries. Influences of processing (boiling, frying, canning, juicing) and storage on the quality and quantity of flavonol glycosides have been reported in onion (Price et al., 1997; Hirota et al., 1998), broccoli (Price et al., 1998a), green beans (Price et al., 1998b), and apples (Price et al., 1999).

The purpose of the present work was to study the effects of domestic processing and storage on flavonols and vitamin C in strawberry, red raspberry, black currant, bilberry, and lingonberry, i.e., the berries most commonly consumed in Finland. Fresh berries were frozen and stored in a domestic freezer up to 9 months. Strawberries were cooked to make jam, and bilberries were cooked to make soup. Lingonberries were partially crushed and stored in their own juice. Lingonberries and black currants were processed to juices. Contents of quercetin, myricetin, kaempferol, and vitamin C in the frozen berries, jam, crushed lingonberries, and juices were analyzed after 3, 6, and 9 months of storage in a

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domestic refrigerator or freezer. In addition to the homemade juices, samples of black currant and crowberry juices prepared by cold-pressing were included in the study.

MATERIALS AND METHODS

Berries. Strawberries (*Fragaria x ananassa* 'Jonsok'), red raspberries (*Rubus idaeus* 'Ottawa'), black currants (*Ribes nigrum* 'Öjebyn'), bilberries (*Vaccinium myrtillus*, wild), and lingonberries (*Vaccinium vitis-idaea*, wild) analyzed in this study were from the same batches as those used in a previous study (Häkkinen et al., 1999b).

Berries were collected from eastern Finland in July-September 1997. The cultivated berries were provided by a local dealer, and wild berries were purchased at a local market place. After harvesting, the berries were stored at +5 °C or, for comparison, at room temperature (+22 °C), and analyzed within 24 h. The berries were frozen in 100-g batches, stored at −20 (±2) °C, and analyzed after 3, 6, and 9 months.

Black currants ("Öjebyn") used for the preparation of cold-pressed juice were harvested from eastern Finland in 1998 and analyzed after 1 month of storage at −20 °C. Crowberries (*Empetrum hermaphroditum*, wild) were collected from Lapland in 1997 and analyzed after 4 months of storage at −20 °C.

Processing. Three sets of strawberry jam, bilberry soup, crushed lingonberries, unpasteurized lingonberry juice, and steam-extracted black currant juice were prepared in the laboratory, and the contents of flavonols and vitamin C were analyzed within 24 h and after 3, 6, and 9 months of storage at +5 (±1) or −20 (±2) °C. Strawberry jam was prepared according to a traditional recipe, by cooking 1500 g of fresh strawberries with 750 g of sugar for 30 min. To make bilberry soup, frozen bilberries (approx 175 g), water (600 mL), and sugar (59 g) were cooked for 10 min and thickened with potato starch (21 g of starch in 50 mL of water). Fresh lingonberries were partly crushed so that the berries were covered with juice when stored in glass containers. To make lingonberry juice, frozen berries (1500 g) were crushed in a food processor and mixed with cool water (2 L) and citric acid (approx. 1 g). After 48 h at +5 °C, the juice was filtered and sugar (750 g) was added. For black currant juice, 3000 g of black currants and 900 g of sugar were cooked in a steamer, and the juice was extracted for 60 min.

Cold-pressing is a widely used commercial scale juicing method for berries in Finland. Two cold-pressed juices were obtained from the manufacturers. Black currant juice was prepared (TUPU 3, Lohjan Mehumetalli, Finland) using pectinase enzyme (Cytolase M102, Gist-Brocades, France; ca. 1.5 mL enzyme/1 kg of berries; yield of juice 75%), and analyzed after 4 days of storage at −20 °C. The cold-pressed crowberry juices were prepared (Joonas-Juicer, Finland) with (ca. 0.7 mL enzyme/1 kg of berries; yield of juice 78%) and without pectinase (yield of juice 74%) and analyzed after 4 months of storage at −20 °C.

Analysis of Flavonols. Frozen samples were analyzed after thawing for 2−3 min in a microwave oven. Three flavonol aglycones (quercetin, myricetin, and kaempferol) were analyzed after extraction and acid hydrolysis of the flavonol glycosides. This method of analysis was developed for the determination of flavonols from frozen berries, and the procedure is described in detail elsewhere (Häkkinen and Auriola, 1998; Häkkinen et al., 1999b). In brief, flavonol glycosides present in the sample (5 g) were extracted and hydrolyzed to their aglycones with 1.2 M HCl in 50% aqueous methanol for 2 h at 85 °C. *tert*-Butylhydroquinone was used as antioxidant. The resulting aglycones were separated by HPLC on a LiChroCART column (Purospher RP-18e) using a gradient of acetonitrile and 1% formic acid as a mobile phase and quantified by UV detection (360 nm). Peak identity and purity were confirmed using a diode array detector (DAD) for on-line recording of the UV−vis spectra of the flavonols present in the samples. Peaks were considered to be pure when there was a correspondence of >900 among the spectra recorded up-slope, apex, and downslope (220−450 nm, 2 nm steps). When the correspondence was <900, unknown coeluting compounds were considered to interfere with the detection/quantification (shown in the tables as "not detectable"). In the berries stored at −20 °C for 3 months, the flavonol peaks were also identified by using electrospray ionization mass spectrometry (Häkkinen and Auriola, 1998).

Coefficients of variation for within-laboratory repeatability (within-day precision) of the method were 9.3% and 6.1% for quercetin and myricetin in black currant, respectively, and those for within-laboratory reproducibility (day-to-day precision) 9.2% and 9.4% for quercetin and myricetin in bilberry, respectively (Häkkinen et al., 1999b).

Recoveries were measured in triplicate from crushed lingonberries, lingonberry juice, and steam-extracted black currant juice by spiking the extraction solutions with pure flavonols. From crushed lingonberries and lingonberry juice, 67% and 60%, respectively, of the quercetin added was recovered. From black currant juice, the recoveries of quercetin and myricetin were 75% and 42%, respectively.

Recoveries from the berries have been reported earlier (Häkkinen et al., 1999b). For strawberry jam, bilberry soup, and cold-pressed crowberry and black currant juices, the recoveries of strawberry, bilberry, crowberry, and steam-extracted black currant juice, respectively, were used on the basis of the similarities of these berry materials to each other. All data reported in this paper have been corrected for the recoveries.

Analysis of Vitamin C. The fresh berries and strawberry jam (100 g) stored at +5 °C were quickly homogenized in a food processor, and a 20-g sample of the homogenate was weighed. The frozen berries and strawberry jam stored at −20 °C were homogenized after partial thawing at room temperature. Ice-cold 1.5% (w/v) metaphosphoric acid (200 mL), pH 3.5−4.0, was immediately added. Black currant juice was diluted 1:5 with the metaphosphoric acid solution. Each sample was prepared in triplicate. A colorimetric, enzymatic method commercially available for the determination of L-ascorbic acid in foodstuffs and other materials (cat. no. 409 677, Boehringer Mannheim GmbH, Mannheim, Germany) was used as previously described (Häkkinen et al., 1999b).

Statistical Analysis. Statistical analysis was carried out by using nonparametric Friedman's two way analysis of variance (SPSS 8.0 for Windows). Values of $P < 0.05$ were considered statistically significant.

RESULTS AND DISCUSSION

Fresh Berries. The total content of the flavonols detected was highest in lingonberry (169 mg/kg) and black currant (157 mg/kg), intermediate in bilberry (41 mg/kg) and strawberry (17 mg/kg), and lowest in red raspberry (9.5 mg/kg) (Table 1). The content of quercetin was highest in lingonberry, intermediate in black currant and bilberry, and lowest in strawberry and red raspberry. Myricetin and kaempferol were detected in black currant and strawberry, respectively, at levels higher than that of quercetin in these berries. There were some difficulties in the detection/quantification of myricetin from fresh bilberries and bilberry soup (peak purity < 900).

No clear value could be obtained for vitamin C in fresh strawberry. After homogenization of the berries, the concentration of L-ascorbic acid declined rapidly, even in the presence of metaphosphoric acid. A similar problem was observed neither with fresh black currants nor with frozen strawberries analyzed after a partial thawing. Due to its low level in bilberry and lingonberry (Häkkinen et al., 1999), vitamin C was not analyzed.

The effects of postharvest temperature were studied in strawberry and black currant. The postharvest tem-

Table 1. Quercetin, Kaempferol, Myricetin, and Vitamin C Contents[a] (milligrams per kilogram) in Fresh Berries Kept at +22 °C or +5 °C for 24 h and in Frozen Berries Kept at −20 °C for 3, 6, and 9 Months

	fresh		frozen			
	+22 °C	+5 °C	3 months	6 months	9 months	P^b
strawberry						
quercetin	4.2 ± 0.1	5.2 ± 0.3	6.9 ± 0.6	7.9 ± 0.5	8.0 ± 1.0	0.045
kaempferol	9.7 ± 0.5	11.8 ± 1.1	5.3 ± 0.1	nd	nd	
vitamin C	na	na	420 ± 15	419 ± 6	405 ± 21	0.944
red raspberry						
quercetin	na	9.5 ± 1.2	8.3 ± 1.0	10.9 ± 1.8	9.7 ± 0.9	0.207
vitamin C	na	296 ± 21	252 ± 11	270 ± 12	266 ± 2	0.017
black currant						
quercetin	52.2 ± 1.0	52.9 ± 5.0	43.8 ± 11.2	43.5 ± 7.8	48.7 ± 3.1	0.910
myricetin	85.8 ± 12.3	104.1 ± 10.0	70.8 ± 8.2	72.7 ± 1.5	nd	0.194
vitamin C	715 ± 52	939 ± 61	904 ± 54	854 ± 66	903 ± 78	0.342
bilberry						
quercetin	na	41.2 ± 3.5	29.0 ± 2.7	30.8 ± 2.2	25.3 ± 1.7	0.002
myricetin	na	nd	21.0 ± 2.1	19.0 ± 1.1	nd	
lingonberry						
quercetin	na	169.0 ± 4.7	146.2 ± 56.6	137.3 ± 18.3	101.2 ± 7.5	0.300

[a] Mean ± SD of triplicate assays, data have been corrected for recoveries. [b] Friedman test (values for +22 °C not included). na = not analyzed; nd = not detectable (peak purity < 900).

Table 2. Quercetin, Kaempferol, Myricetin, and Vitamin C Contents[a] (milligrams per kilogram) in Freshly Prepared Berry Products and after Storage at −20 °C or +5 °C for 3, 6, and 9 Months

		3 months		6 months		9 months		P^b	
	freshly prepared	+5 °C	−20 °C	+5 °C	−20 °C	+5 °C	−20°C	+5 °C	−20 °C
strawberry jam									
quercetin	3.2 ± 0.1	3.8 ± 1.4	2.2 ± 0.1	4.0 ± 0.1	3.5 ± 0.1	4.7 ± 0.1	4.7 ± 0.3	0.207	0.002
kaempferol	7.2 ± 1.0	nd	nd	nd	nd	na	na		
vitamin C	236 ± 7	173 ± 13	224 ± 7	122 ± 23	224 ± 5	155 ± 15	226 ± 15	0.002	0.354
bilberry soup									
quercetin	6.0 ± 0.9	na	na	na	na	na	na		
crushed lingonberries									
quercetin	100.2 ± 18.2	na	na	93.4 ± 11.5	na	53.7 ± 4.0	na	0.194	
lingonberry juice									
quercetin	9.3 ± 0.9	na	na	11.3 ± 0.6	na	10.7 ± 4.4	na	0.389	
black currant juice, steam-extracted									
quercetin	7.5 ± 0.3	6.7 ± 0.4	7.6 ± 0.1	7.3 ± 0.7	6.0 ± 0.3	6.8 ± 0.7	6.0 ± 0.4	0.399	0.038
myricetin	28.3 ± 0.2	11.7 ± 1.3	13.3 ± 0.4	8.2 ± 0.4	9.7 ± 0.4	nd	nd	0.028	0.028
vitamin C	421 ± 16	350 ± 28	364 ± 13	356 ± 27	381 ± 30	378 ± 32	406 ± 35	0.002	0.017
black currant juice[c], cold-pressed, with pectinase									
quercetin			25.2 ± 0.3						
myricetin			31.6 ± 1.0						
crowberry juice[d], cold-pressed, with pectinase									
quercetin			39.9 ± 5.9						
myricetin			35.1 ± 7.2						
crowberry juice[d], cold-pressed, without pectinase									
quercetin			37.6 ± 1.5						
myricetin			34.6 ± 2.2						

[a] Mean ± SD of triplicate assays, data have been corrected for recoveries. [b] Friedman test. [c] Berries stored at −20 °C for 3 months, juice stored at −20 °C for 4 days. [d] Berries stored at −20 °C for one week, juice stored at −20 °C for 4 months. na = not analyzed; nd = not detectable (peak purity < 900).

perature had an apparent effect on quercetin and kaempferol contents in strawberry and on myricetin and vitamin C contents in black currant (Table 1). Approximately 10−20% lower levels were detected in the berries stored for 24 h at room temperature (+22 °C) compared to those stored at +5 °C. According to our results, the practice of keeping berries at low temperature during postharvest storage and transportation is advantageous also from the point of view of saving the flavonols and vitamin C, not to mention other obvious advantages.

Cooked Berries. The effects of jam preparation were studied using strawberries, despite their relatively low levels of quercetin and kaempferol. A reason for this was that strawberry jam is the most commonly consumed jam in Finland. In freshly prepared strawberry jam, the contents of quercetin and kaempferol were 3.2 and 7.2 mg/kg, respectively (Table 2). Cooking strawberries with sugar for 30 min caused smaller losses of flavonols than did the other processing methods studied in any of the berries. The losses of quercetin and kaempferol were 18 and 15%, respectively, of the amounts originally present in the berries. This is probably due to the fact that whole berries were cooked without crushing. In addition, polyphenol oxidase is inactivated by the high temperature (Waterman and Mole, 1994). The added sugar might also have protected flavonols during the cooking process. Flavonols were less sensitive than vitamin C (36% loss) during cooking of strawberries.

Bilberries were cooked for 10 min with water and sugar to make a soup, a common dessert in Finland. The skins of the berries were not removed after cooking. Quercetin content in the soup was 6.0 mg/kg (Table 2). Cooking resulted in 40% loss of quercetin from that originally present in the berries. The loss was probably due to the breakdown of quercetin during the cooking

process and/or oxidative reactions (Waterman and Mole, 1994). Cooking of tomatoes and onions by boiling for 15 min resulted in 88 and 75% losses, respectively, of conjugated quercetin (Crozier et al., 1997). Smaller reductions were detected following microwaving and frying. According to Price et al. (1998a), only 14−28% of the individual flavonol glycosides of broccoli florets were retained in the cooked tissue, the remainder being largely leached into the cooking water. Also during cooking of bilberries by boiling, quercetin is probably leached into the water fraction. In the preparation of bilberry soup, however, the cooking water is not discarded. This probably explains the much smaller loss of quercetin than in the case of cooking tomatoes, onions, and broccoli by boiling.

Crushed Berries. Lingonberries were preserved in a traditional way (fresh berries were partially crushed and stored in their own juice in a refrigerator). The level of quercetin was lowered by 40% when crushed lingonberries were kept at +5 °C overnight. This might be due to enzymatic reactions that start when the subcellular compartmentation breaks down and enzymes come into contact with potential substrates to which they are normally not exposed (Waterman and Mole, 1994). Moreover, the loss of membrane integrity increases the potential of oxidation of phenolic compounds (Waterman and Mole, 1994). Despite the loss, the freshly crushed berries had a high level of quercetin, i.e., 100 mg/kg (Table 2).

Juices. When juices were made using common domestic processing methods, considerable reductions in flavonol contents were observed. In lingonberry juice (unpasteurized) and black currant juice (steam-extracted), only 15% of quercetin and 30% of myricetin (in black currant juice only) were extracted into the juices. This is due to the facts that the skins of the berries were removed by filtering, and flavonols are known to be concentrated mainly in the skins of fruits (Hawker et al., 1972; Wildanger and Herrmann, 1973; Price et al., 1999). According to van der Sluis et al. (1997), only 10% of the original quercetin was found in apple raw juice produced by enzymatic pulping. Also, commercial scale pressing from cider apple varieties resulted in juice that contained 9.9% to 12.7% of the flavonols, the rest being retained in the pomace (Price et al., 1999). Peach-based products are completely devoid of flavonol derivatives due to the removal of the skin in the manufacturing process (Bengoechea et al., 1997). Compared to flavonols, vitamin C was somewhat more effectively extracted to black currant juice (44% of that originally present in the berries) in this study.

The level of quercetin was higher in black currant juice prepared by cold-pressing (25 mg/kg) than in that prepared by steam-extraction (7.5 mg/kg) (Table 2). The level of myricetin was almost the same in cold-pressed and steam-extracted juices (31.6 and 28.3 mg/kg, respectively). Of quercetin and myricetin originally present in the berries, 45% and 65% were extracted to the cold-pressed juice, respectively. Thus, the cold-press method was superior to the traditional steam-extraction method in extracting quercetin and myricetin from black currant. This could be due to more effective extraction of flavonols from berry material (mainly the skins) by mechanical cold-pressing compared to chemical steam-extraction. One reason for the differences in the extractability of flavonols might be that black currants were not subjected to freeze−thaw treatment prior to the steam-extraction process as was done with the berries used in the cold-pressing process. According to Sapers et al. (1983), freeze−thaw treatment increased the anthocyanin content of cranberry juice by as much as 15-fold. The treatment facilitated the migration of anthocyanins from the exocarp into the mesocarp and endocarp during thawing of cranberries and thus enhanced pigment extraction during processing. This might also occur for flavonols in black currants.

It was of interest that the myricetin/quercetin ratio in steam-extracted black currant juice was higher (3.8) than in intact berries (2.1) or cold-pressed juice (1.2). One explanation might be that myricetin is more effectively extracted by hot water than is the less-polar quercetin. Differences in these ratios might also reflect the different localization of myricetin and quercetin within the berries. According to Price et al. (1999), the individual flavonol conjugates in apple varieties are not necessarily distributed in the same proportions between the flesh and peel of the fruit. This could also partly explain the observation that the extractability of myricetin to cold-pressed juices varied depending on the berry. Higher percentages of myricetin were extracted to the black currant juice (65%) than to the crowberry juices (approx 45%). Also, the differences in the structures of myricetin glycosides in these berries (Häkkinen and Auriola, 1998) might result in the variation in the extractability.

In the frozen berries used for the preparation of crowberry juices by cold-pressing, the contents of both quercetin and myricetin were 61 mg/kg. The use of pectinase had no influence on the extractability of quercetin in cold-pressed crowberry juices. When pectinase enzyme was used in the process, 48% and 47% of quercetin and myricetin, respectively, were extracted to the juice. Without pectinase treatment, 47% and 43% of quercetin and myricetin, respectively, were extracted to the juice. Rommel and Wrolstad (1993) reported that the red raspberry juice produced by high-speed centrifugation combined with pectinases contained less total quercetin forms (ca. 190 ppm) than the juice prepared without pectinases (280 ppm). This was probably due to deglycosylation of flavonol glycosides to less-stable aglycones in the presence of pectinases. In the crowberry juices such an effect was not observed.

Among all the juices analyzed in the present study, the crowberry juices had the highest levels of quercetin and myricetin (approx 39 and 35 mg/kg, respectively) (Table 2). The black currant and crowberry juices are consumed after dilution with water. Despite the reduction during the juicing, the flavonol content in diluted (1:4 v/v) steam-extracted black currant juice was 9.5 mg/L which compares well with flavonol levels found in fruit juices (2.5−13 mg/L) (Hertog et al., 1993b). In the cold-pressed black currant and crowberry juices, the flavonol concentrations after dilution (1:4 v/v) were 15 and 19 mg/L, respectively. These levels are higher than those reported for fruit juices (Hertog et al., 1993b) and compare well with levels found in red wines (4.6−41.6 mg/L) (McDonald et al., 1998).

Influence of Storage at +5 °C or −20 °C on Flavonols in Berries and Berry Products. To our knowledge, no previous studies on the effects of storage by freezing on flavonol contents in foods are available. The results obtained with the five species of berries studied show that the effects of freezing on flavonols and vitamin C vary among berries and berry products.

In strawberries, markedly higher (35%) quercetin levels were measured after 9 months of storage at −20 °C (8.0 mg/kg) than from fresh strawberries (5.2 mg/kg) (Table 1). A gradual increase (32%) was observed also in strawberry jam stored at −20 °C or +5 °C (Table 2). The most probable explanation for these unexpected observations could be that quercetin in frozen strawberries and strawberry jam becomes more easily extractable and hydrolyzable during storage. This might be due to degradation of cell structures during storage. Previously, an increase in flavonol content during storage at 4−5 °C (10 days to 6 months) has been reported for strawberries (Gil et al., 1997), pears (Amiot et al., 1995), and freeze-dried onion bulbs (Price et al., 1997). The contents of quercetin glycosides in apples were not changed during 6 months of cold storage at 0 °C (Burda et al., 1990). Kaempferol was detected in fresh strawberries (11.8 mg/kg) and after 3 months storage in a freezer (5.3 mg/kg), but could not be quantified after 6 months (small peak, peak purity < 900) (Table 1). Similarly, although present in the freshly prepared jam, kaempferol could not be quantified any more after 3 months of storage at −20 °C or at +5 °C (Table 2). Vitamin C level decreased markedly in the strawberry jam stored at +5 °C, but not in that stored at −20 °C (Table 2).

Quercetin was well preserved in frozen red raspberries and black currants, since no changes were observed during 9 months (Table 1). Also in black currant juice, the level of quercetin was reduced maximally by only 20% (at −20 °C but not at +5 °C after 9 months of storage) (Table 2). In contrast to quercetin, myricetin contents were significantly reduced in frozen black currants and black currant juice during 6 months of storage, being nondetectable after 9 months (Tables 1, 2). According to our results, myricetin is more stable in intact berries than in juice (30% vs 66% loss in 6 months at −20 °C).

Vitamin C was well preserved in frozen black currants (Table 1). In red raspberries it appeared to be slightly more sensitive, showing minor, although statistically significant changes during storage at −20 °C. Also in black currant juice, the level of vitamin C slightly decreased during storage at both temperatures (Table 2).

In intact bilberries and lingonberries preserved by freezing, a gradual decline (up to 40%) in the content of quercetin was observed after 9 months at −20 °C (Table 1). In lingonberries, the reduction (from 169 to 101.2 mg/kg) was not statistically significant due to the high variability in the 3-month values. One explanation for the loss of quercetin might the low content of vitamin C in these two berries (not detected in bilberry, 75 mg/kg fresh weight in lingonberry by Häkkinen et al., 1999b). The high content of vitamin C in black currant, strawberry, and red raspberry (904, 420, and 252 mg/kg, fresh weight) might have protected quercetin during the storage in a freezer. After 3 and 6 months of storage at −20 °C, myricetin contents in frozen bilberries were 21 and 19 mg/kg, respectively. After 9 months of storage, myricetin could not be quantified.

In crushed lingonberries, the level of quercetin remained quite stable during 6 months of storage in a refrigerator, most probably due to the slowing down of the enzymatic and oxidative reactions. Even after 9 months, the content of quercetin was rather high (50 mg/kg) (Table 2). However, quercetin was less stable in crushed lingonberries (50% loss) and in intact frozen lingonberries (40% loss) than in unpasteurized lingonberry juice (no losses) stored in a refrigerator for 9 months. Effects of long-term storage on flavonols in bilberry soup were not studied, since homemade soup is consumed within one or 2 days.

CONCLUSIONS

Variable effects of domestic processing and storage were observed in quercetin, myricetin, and kaempferol contents of five berries most commonly consumed in Finland. When juices were made by common domestic processing methods, considerable losses of flavonols were observed. Cold-pressing was superior to steam-extraction in extracting the flavonols. Also, crushing the berries resulted in a considerable loss of quercetin. Our results show that the effects of freezing on quercetin are different in different berries. They also suggest that myricetin and kaempferol are more susceptible than quercetin to losses during processing and storage of berries.

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