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USE OF CONTENT ANALYSIS OF SELECTED ORGANIC ACIDS FOR THE DETECTION OF BERRY JUICE ADULTERATIONS

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Citric, D-isocitric and L-malic acid contents in juices of three strawberry, raspberry, black currant and red currant varieties, as well as in selected raspberry and black currant juices with 30% addition of strawberry and red currant juices were determined in this study with the use of enzymatic methods. Furthermore, citric acid:D-isocitric acid ratios were estimated in these juices. Examinations were performed in three consecutive years: 1998, 1999 and 2000. The achieved organic acid contents and acid ratios were compared with standard values specified in the Code of Practice.

Deviations from limits of citric acid contents and citric acid:D-isocitric acid ratios accepted in the Code of Practice were found and changes of these limits were proposed. L-malic acid contents can be used for the estimation of raspberry and black currant juice adulterations. The authenticity of juices cannot be estimated on the basis of citric acid, D-isocitric acid contents, as well as citric acid:D-isocitric acid ratios.

INTRODUCTION

Fruit juices are adulterated by the addition of water, sugar, acid, synthetic taste and aromatic substances, dyes, extracts of peels and foots. Furthermore, expensive juices can be adulterated by cheap juices. By adulteration of juices, producers decrease the costs of raw materials and achieve higher economic profit [Czapski & Tyma, 1996; Martinek, 1998; Neuhäuser, 2000; Niewiarowicz, 2000; Obiedziński, 1998; Płocharski, 2000; Stój *et al.*, 2001; Targoński, 2000].

The basic organic acids occurring in fruit juices are: citric, D-isocitric and L-malic. Contents of these organic acids, as well as citric acid:D-isocitric acid ratios are different for different juices, hence they can be the indicators of their authenticity. These parameters are compared with standard values, *e.g.* those contained in the Code of Practice [1996]. The Code of Practice was elaborated by the Association of the Industry of Juices and Nectars from Fruits and Vegetables of the European Union (AIJN) – an organization which is engaged in standardization of the quality rules and authenticity estimation of fruit juices. Deviations from accepted norms are the indicators of non-declared juice addition.

Contents of particular organic acids in berry juices have mainly been determined with the use of liquid chromatography [Boccorh *et al.*, 1998; Durst *et al.*, 1995; Perez *et al.*, 1998; Riaz & Bushway; 1994; Spanos & Wrolstad, 1987], gas chromatography [Kallio *et al.*, 2000], and enzymatic methods [Plowman, 1991]. Organic acid contents are very variable and depend, to a large extent, on the variety of fruit, stage of ripeness, climate and soil conditions and technology of juice production.

Strawberry juices have been reported to contain 7.3 g/L to 15.8 g/L of citric acid and from 2.2 g/L to 6.9 g/L of L-malic acid [Kallio *et al.*, 2000]. According to the Code of Practice, however, strawberry juices should contain 5–11 g/L of citric acid, 0.03-0.09 g/L of D-isocitric acid and 0.6–5.0 g/L of L-malic acid. Raspberry juices have been shown to contain from 8.0 g/L to 29.0 g/L of citric acid, from 0.057 g/L to 0.440 g/L of D-isocitric acid and from 0.033 g/L to 2.168 g/L of L-malic acid [Durst *et al.*, 1995; Spanos & Wrolstad, 1987]. The Code of Practice permits 9-22 g/L of citric acid, 0.06–0.22 g/L of D-isocitric acid and 0.2–1.2 g/L of L-malic acid. Contents of citric acid in black currant juices ranged from 21.15 g/L to 65.44 g/L, those of D-isocitric acid from 0.128 g/L to 0.424 g/L, and those of L-malic acid from 0.90 g/L to 3.07 g/L [Gherardi *et al.*, 1995]. Standard values in black currant juices accounted for: 26–42 g/L of citric acid, 0.16–0.50 g/L of D-isocitric acid, and 1–4 g/L of L-malic acid.

Organic acid contents in native cherry juice and in cherry juices with 10% addition of red beet juice and 10% addition of blackberry juice have also been determined. An increase in citric acid concentration resulted from the addition of red beet juice, and an increase in D-isocitric acid concentration was due to the addition of blackberry juice [Gherardi *et al.*, 1995].

Citric, D-isocitric and L-malic acid contents in authentic strawberry, raspberry, black currant and red currant

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juices were determined in the present study. Furthermore, citric acid:D-isocitric acid ratios were estimated in these juices. The achieved organic acid contents and acid ratios were compared with standard values specified in the Code of Practice. Deviations from accepted norms of organic acid contents in authentic berry juices were the basis for the proposed changes of selected parameters in the Code of Practice. On the basis of determination results of organic acids, some expensive raspberry and black currant juices adulterated by cheap strawberry and red currant juices were selected for further examinations. The aim of the present study was to indicate these organic acids whose contents can be used for the estimation of adulterations of berry juices produced in Poland. Examinations were performed in three consecutive years: 1998, 1999 and 2000. Use was made of enzymatic methods recommended in the Code of Practice.

MATERIAL AND METHODS

Four berry species were collected in 1998, 1999 and 2000. Strawberries (*Fragaria ananassa*) of Senga, Ducat and Marmolada cv. originated from the plantation in Zemborzyce. Raspberries (*Rubus idaeus*) of Beskid, Canby and Malling Seedling cv. as well as black currants (*Ribes nigrum*) of Ben Lomond, Titania and Ojebyn cv. were from The Experimental Farm in Felin. Red currants (*Ribes rubrum*) of Rondom and Holenderska cv. were collected in a garden in Klementowice and Jonker cv. was collected in Góra Puławska.

Berry fruits were stored in a refrigerator at -28°C. Juices were made from fruits defrosted at ambient temperature in a juice extractor Zelmer 277,8 and then centrifuged in a MPW 365 centrifuge for 15 min at 4°C and 11 000 rpm.

The following adulterated juices were also prepared: raspberry juice of Beskid cv. with 30% addition of strawberry juice of Senga cv., raspberry juice of Beskid cv. with 30% addition of red currant juice of Rondom cv., black currant juice of Ben Lomond cv. with 30% addition of strawberry juice of Senga cv. and black currant juice of Ben Lomond cv. with 30% addition of red currant juice of Rondom cv.

Prior to organic acid determinations, samples of juices were neutralized with NaOH solution and adjusted to pH of *ca.* 8, diluted with redistilled water, decolourized with PVPP (polyvinylpolypyrrolidone) and filtered. Determinations of organic acids were performed using the enzymatic method according to Boehringer Mannheim [1986] in two replications. The following enzymatic kits of the above-mentioned company were used in the study: citric acid – kit No 139 076, D-isocitric acid – kit No 414 433, and L-malic acid – kit No 139 068.

After completing the enzymatic reactions, absorbance of solutions was measured at a wavelength of 340 nm using a UV-VIS Unicam 5625 spectrophotometer. Contents of organic acids were calculated according to the following equation:

$$c = \frac{V \times MW}{\varepsilon \times d \times v \times 1000} \times \Delta A \times r$$

where: c – organic acid content (g/L); V – final volume (mL); v – sample volume (mL); MW – molecular weight of acid (g/mol); d – light path (cm); ε – extinction coefficient of NADH – at 340 nm = 6.3; ΔA – absorbance difference; r – dilution factor.

Determination results were elaborated statistically using the Tuckey's test at a significance level of $\alpha = 0.05$.

RESULTS AND DISCUSSION

Contents of citric acid in strawberry, raspberry, black currant and red currant juices, as well as in black currant juices with 30% addition of strawberry juices were shown in Table 1.

Contents of citric acid in strawberry juices ranged from 4.32 g/L in juice of Marmolada cv. in 1998 to 8.70 g/L in juice of Senga cv. in the same year. Kalio *et al.* [2000] determined organic acid contents in juices of different strawber-

TABLE 1. Contents of citric acid in authentic and adulterated berry juices in 1998, 1999, 2000.

Type of juice	Fruit variety	Citric acid (g/L)			
		Year of fruit harvest			Average
		1998	1999	2000	
Strawberry juice	Senga	8.70	5.84	6.16	$6.90^{AB} \pm 1.57$
	Ducat	6.85	7.30	7.29	$7.15^{ABC} \pm 0.26$
	Marmolada	4.32	4.99	5.10	$4.80^{A} \pm 0.42$
Raspberry juice	Beskid	23.56	7.46	19.54	$16.85^{D} \pm 8.38$
	Canby	23.97	8.04	14.36	$15.46^{BCD} \pm 8.02$
	Malling Seedling	25.19	8.02	13.82	$15.68^{CD} \pm 8.73$
Black currant juice	Ben Lomond	41.02	38.85	24.56	$34.81^{G} \pm 8.94$
	Titania	40.46	32.66	27.96	$33.69^{FG} \pm 6.31$
	Ojebyn	38.25	36.11	23.43	$32.60^{FG} \pm 8.01$
Red currant juice	Rondom	19.13	13.00	9.90	$14.01^{BCD} \pm 4.70$
	Jonker	19.91	26.08	21.54	$22.51^{DE} \pm 3.20$
	Holenderska	23.43	17.91	13.51	$18.28^{DE} \pm 4.97$
Adulterated black currant juice	Ben Lomond 70% + Senga 30%	29.84	29.47	18.37	$25.89^{EF} \pm 6.52$

A, B – values in the columns with different letters are significantly different at $\alpha = 0.05$

ry varieties with the use of gas chromatography. According to them, juice of Senga VP cv. in 1997 contained the lowest – 7.3 g/L, and juice of Honeoye cv. also in 1997 – the highest level of citric acid – 15.8 g/L. In our examinations, citric acid content in strawberry juices of Senga cv. was 8.70 g/L in 1998, 5.84 g/L – in 1999, and 6.16 g/L – in 2000. Kalio *et al.* [2000] determined similar concentrations of citric acid in juices of Senga VP cv. in 1997 and in 1998 – 7.3 g/L and 8.4 g/L, respectively. Concentrations of citric acid in the other juices were higher – from 9.1 g/L in juice of Senga PP cv. in 1997 to 11.5 g/L in juice of Senga SW cv. in 1998. According to the Code of Practice, citric acid content, determined with the enzymatic method, in authentic strawberry juice ranged from 5 to 11 g/L. In the present examinations, citric acid concentrations in juices of Marmolada cv. were lower than the standard in 1998 and 1999 – 4.32 g/L and 4.99 g/L, respectively (by 13.6% and 0.2% lower, respectively). Therefore, it is proposed that the lowest content of citric acid in strawberry juice be decreased to *ca.* 4 g/L.

Contents of citric acid in raspberry juices ranged from 7.46 g/L in juice of Beskid cv. in 1999 to 25.19 g/L in juice of Malling Seedling in 1998. Similar citric acid concentrations were met in literature. Durst *et al.* [1995], using liquid chromatography, found from 22.8 g/L to 23.4 g/L of citric acid in raspberry juices of Malling Seedling cv., and in other juices – from 8.0 g/L of citric acid in juice of Meeker cv. to 24.7 g/L of this acid in juice of Veten cv. Spanos & Wrolstad [1987], using the enzymatic method, recorded from 13.8 g/L of citric acid in juice of Meeker cv. (originated from Washington) to 29.0 g/L in juice of Willamette cv. (originated from Canada). The Code of Practice permits the content of citric acid, determined by means of the enzymatic method, to reach from 9 to 22 g/L in raspberry juice. Juices of Beskid, Canby and Malling Seedling cv. in 1998 contained higher levels of citric acid – 23.56 g/L, 23.97 g/L and 25.19 g/L, respectively (by 7.1%, 9.0% and 14.5% higher levels of citric acid, respectively), whereas in 1999 – lower levels of this acid – 7.46 g/L, 8.04 g/L and 8.02 g/L, respectively (by 17.1%, 10.7% and 10.8% lower levels of this acid, respectively). The results obtained in this study indicate that the range of citric acid content in authentic raspberry juice should be increased in the Code of Practice from *ca.* 7 g/L to *ca.* 26 g/L.

In the present examinations, the concentrations of citric acid in black currant juices were from 23.43 g/L in juice of Ojebyn cv. in 2000 to 41.02 g/L in juice of Ben Lomond cv. in 1998. Similarly, Gherardi *et al.* [1995], using ion exchange chromatography, determined from 21.15 g/L to 65.44 g/L of citric acid in blackcurrant juices. In our examinations, deviations from standard (26–42 g/L) were observed in 2000. Juice of Ojebyn cv. contained 23.43 g/L of citric acid (by 9.9% lower), and juice of Ben Lomond cv. – 24.56 g/L of this acid (by 5.6% lower). Therefore, it is proposed that the lowest concentration of citric acid in black currant juice be decreased to a level of *ca.* 23 g/L.

Concentrations of citric acid in red currant juices in particular years ranged from 9.90 g/L in juice of Rondom cv. in 2000 to 26.08 g/L in juice of Joker cv. in 1999. The Code of Practice does not define the contents of citric, D-isocitric and L-malic acid, as well as the citric acid:D-isocitric acid ratio in red currant juice. Therefore, it is necessary to complete the Code of Practice with the above-mentioned parameters, which are used for the estimation of red currant juice authenticity.

The results of determinations of citric acid concentrations in berry juices indicated that black currant juices contained significantly higher levels of citric acid in comparison with strawberry juices. Therefore, it was examined whether it is possible to detect the addition of strawberry to black currant juice on the basis of citric acid content analysis. A statistical analysis indicated significant differences between the average citric acid contents over three years of examinations of authentic black currant (34.81 g/L) and black currant juice adulterated with the strawberry one (25.89 g/L). Concentrations of this acid in juices composed of 70% black currant juice of Ben Lomond cv. and 30% strawberry juice of Senga cv. in 1998 and 1999 were

TABLE 2. Contents of D-isocitric acid in authentic and adulterated berry juices in 1998, 1999, 2000.

Type of juice	Fruit variety	D-isocitric acid (g/L)			
		Year of fruit harvest			Average
		1998	1999	2000	
Strawberry juice	Senga	0.06	0.05	0.04	$0.05^{A}\pm0.01$
	Ducat	0.06	0.05	0.06	$0.06^{A}\pm0.01$
	Marmolada	0.05	0.04	0.05	$0.05^{A}\pm0.01$
Raspberry juice	Beskid	0.09	0.09	0.13	$0.10^{B}\pm0.02$
	Canby	0.10	0.09	0.12	$0.10^{B}\pm0.01$
	Malling Seedling	0.10	0.09	0.12	$0.10^{B}\pm0.02$
Black currant juice	Ben Lomond	0.24	0.30	0.23	$0.26^{GH}\pm0.04$
	Titania	0.25	0.32	0.25	$0.27^{H}\pm0.04$
	Ojebyn	0.25	0.30	0.24	$0.26^{GH}\pm0.03$
Red currant juice	Rondom	0.14	0.13	0.13	$0.13^{C}\pm0.01$
	Jonker	0.26	0.24	0.26	$0.25^{F}\pm0.01$
	Holenderska	0.13	0.19	0.17	$0.16^{D}\pm0.03$
Adulterated black currant juice	Ben Lomond 70% + Senga 30%	0.18	0.22	0.18	$0.19^{E}\pm0.02$

A, B – values in the columns with different letters are significantly different at $\alpha=0.05$

29.84 g/L and 29.47 g/L, respectively, therefore, they met the recommended limits for authentic black currant juice. Citric acid content in adulterated black currant juice in 2000 accounted for 18.37 g/L, which was less than recommended in the Code of Practice. Therefore, citric acid can not be the indicator of the addition of strawberry juice or another one with low citric acid content to black currant juice.

Contents of D-isocitric acid in strawberry, raspberry, black currant and red currant juices, as well as in black currant juices with 30% addition of strawberry juices were shown in Table 2.

Strawberry juices in particular years contained from 0.04 g/L of D-isocitric acid in juice of Senga cv. in 2000 and in juice of Marmolada cv. in 1999 to 0.06 g/L in juice of Senga cv. in 1998 and in juices of Ducat cv. in 1998 and 2000.

The contents of D-isocitric acid in raspberry juices were from 0.09 g/L in juices of Beskid cv. in 1998 and 1999, Canby cv. in 1999 and Malling Seedling cv. in 1999 to 0.13 g/L in juice of Beskid cv. in 2000. Other authors found higher concentrations of D-isocitric acid in raspberry juices. According to Durst *et al.* [1995], the content of this acid ranged from 0.057 g/L in juice of Malling Seedling cv. to 0.440 g/L in juice of Willamette cv. Spanos & Wrolstad [1987] determined from 0.083 g/L of D-isocitric acid in juice of Meeker cv. (Washington) to 0.184 g/L in juice of Marcy cv. (New Zealand).

The contents of D-isocitric acid in black currant juices ranged from 0.23 g/L in juice of Ben Lomond cv. in 2000 to 0.32 g/L in juice of Titania cv. in 1999. Gherardi *et al.* [1995] reported on more differentiated concentrations of D-isocitric acid in black currant juices – from 0.128 g/L to 0.424 g/L.

According to the Code of Practice, the content of D-isocitric acid in authentic strawberry juice should account for 0.03–0.09 g/L, in raspberry juice – for 0.06–0.22 g/L, and in black currant juice – for 0.16–0.50 g/L. In the present examinations, the determined concentrations of D-isocitric acid in strawberry, raspberry and black currant juices met the recommended limits of the Code of Practice.

The contents of D-isocitric acid in red currant juices in particular years ranged from 0.13 g/L in juices of Rondom cv. in 1999 and 2000 and in juice of Holenderska cv. in 1998 to 0.26 g/L in juices of Joker cv. in 1998 and 2000. Differences between the average concentrations of D-isocitric acid in juices of three red currant varieties were confirmed statistically. The average content of this acid in juice of Rondom cv. was 0.13 g/L, in juice of Holenderska cv. – 0.16 g/L, and in juice of Jonker cv. – 0.25 g/L.

Due to significant differences between contents of D-isocitric acid in black currant and strawberry juices, it was examined whether determinations of D-isocitric acid content could be the indicator of adulteration of black currant juice with strawberry juice. A statistical analysis indicated significant differences between the average D-isocitric acid contents of three years in authentic black currant juice (0.26 g/L) and in black currant juice with addition of strawberry juice (0.19 g/L). Juices being the mixtures composed of 70% black currant juice of Ben Lomond cv. and 30% strawberry juice of Senga cv. in 1998, 1999, 2000 contained 0.18 g/L, 0.22 g/L and 0.18 g/L of D-isocitric acid, respectively. Therefore, according to the Code of Practice, adulterated black currant juices would be estimated as authentic.

The calculated citric acid:D-isocitric acid ratios in strawberry, raspberry, black currant and red currant juices, as well as in black currant juices with 30% addition of strawberry juices were shown in Table 3.

The citric acid:D-isocitric acid ratios in strawberry juices were from 82 in juice of Marmolada cv. in 1998 to 150 in juice of Senga cv. in 2000. The average citric acid:D-isocitric acid ratio of three experimental years in juice of Marmolada cv. was significantly lower (106) in comparison with the average ratios of these acids in juices of Ducat (132) and Senga cv. (137). According to the Code of Practice, in authentic strawberry juice the citric acid:D-isocitric acid ratio should range from 100 to 230. That ratio was lower of the lowest value (by 18% lower) in juice of Marmolada cv.

TABLE 3. Citric acid:D-isocitric acid ratios in authentic and adulterated berry juices in 1998, 1999, 2000.

Type of juice	Fruit variety	Citric acid:D-isocitric (g/L)			
		Year of fruit harvest			Average
		1998	1999	2000	
Strawberry juice	Senga	138	123	150	137 ^{F}
	Ducat	125	139	132	132 ^{DEF}
	Marmolada	82	124	114	106 ^{B}
Raspberry juice	Beskid	256	81	153	163 ^{H}
	Canby	246	90	122	152 ^{G}
	Malling Seedling	245	94	119	153 ^{G}
Black currant juice	Ben Lomond	173	129	106	136 ^{EF}
	Titania	166	101	113	127 ^{CDE}
	Ojebyn	153	119	98	123 ^{CD}
Red currant juice	Rondom	138	102	75	105 ^{B}
	Jonker	77	111	82	90 ^{A}
	Holenderska	188	96	79	121 ^{C}
Adulterated black currant juice	Ben Lomond 70% + Senga 30%	168	132	103	134 ^{EF}

A, B – values in the columns with different letters are significantly different at α=0.05

in 1998. Therefore, it is suggested that the lowest value of the citric acid:D-isocitric acid ratio in strawberry juice be decreased to *ca.* 80.

Citric acid:D-isocitric acid ratios in juices of different raspberry varieties ranged from 81 in juice of Beskid cv. in 1999 to 256 in juice of the same raspberry variety, but in 1998. Of the raspberry juices, the average citric acid:D-isocitric acid ratio in juice of Beskid cv. was significantly higher (163) in comparison with the average ratios of these acids in juice of Malling Seedling cv. (153) and in juice of Canby cv. (152). Durst *et al.* [1995] found significant variations of two acid ratios – from 35 in juice of Meeker cv. to 400 in juice of Malling Seedling cv. Spanos & Wrolstad [1987] published approximate ratio values – from 124 in juice of Meeker cv. (Oregon) to 188 in juice of Skeena cv. (Canada). In our examinations, raspberry juices of Beskid, Canby and Malling Seedling cv. in 1998 had higher citric acid:D-isocitric acid ratios in comparison with the standard (80–200) – 256, 246 and 245, respectively (by 28%, 23% and 22.5% higher). Therefore, it is proposed that the highest value of two acid ratios in raspberry juice be increased to *ca.* 260.

The citric acid:D-isocitric acid ratios in black currant juices were from 98 in juice of Ojebyn cv. in 2000 to 173 in juice of Ben Lomond cv. in 1998. Of the black currant juices, the average ratio of these two acids was significantly higher in juice of Ben Lomond cv. (136) in comparison with the average ratio of acids in juice of Ojebyn cv. (123). The acid ratios in juices of different black currant varieties agreed with those proposed in the Code of Practice (80–200).

The ratios of the two acids in red currant juices in particular years were differentiated, *i.e.* they ranged from 75 in juice of Rondom cv. in 2000 to 188 in juice of Holenderska cv. in 1998. The average citric acid:D-isocitric acid ratio in red currant juice of Jonker cv. was 90, in juice of Rondom cv. – 105, and in juice of Holenderska cv. – 121. These differences were statistically significant.

From the statistical point of view, authentic and adulterated black currant juices did not differ referring to the average ratios of these two acids (136 and 134, respectively). The citric acid:D-isocitric acid ratios in juices composed of 70% black currant juice of Ben Lomond cv. and 30% strawberry juice of Senga cv. in 1998, 1999 and 2000 were 168, 132 and 103, respectively. Therefore, according to the Code of Practice, adulterated black currant juices would be estimated as authentic.

Contents of L-malic acid in strawberry, raspberry, black currant and red currant juices, as well as in raspberry juices with 30% addition of strawberry and red currant juices, as well as in black currant juices with 30% addition of red currant juices were shown in Table 4.

No deviations of L-malic acid concentrations from estimated standards were found in strawberry, raspberry and black currant juices. Standard values in strawberry juice are 0.6–5.0 g/L, in raspberry juice – 0.2–1.2 g/L and in black currant juice – 1–4 g/L.

The contents of L-malic acid in strawberry juice ranged from 1.03 g/L in juice of Marmolada cv. in 2000 to 3.59 g/L in juice of Senga cv. in 1998. The average L-malic acid content of three experimental years accounted for 1.21 g/L in juice of Marmolada cv., for 2.45 g/L in juice of Ducat cv. and for 2.83 g/L in juice of Senga cv. From the statistical point of view, strawberry juices significantly differed referring to the average L-malic acid contents. However, Kallio *et al.* [2000] determined from 2.2 g/L of L-malic acid in juice of Honeoye cv. in 1997 to 6.9 g/L of this acid in juice of Senga SW cv. in 1998. The concentrations of L-malic acid in strawberry juices of Senga cv. in 1998, 1999 and 2000 were 3.59 g/L, 2.95 g/L and 1.95 g/L, respectively. Kallio *et al.* [2000] found 3.1 g/L of L-malic acid in juice of Senga VP cv., which was similar to the concentration of that acid recorded in our examinations in 1999. The other strawberry juices contained higher concentrations of L-malic acid – from 4.3 g/L in juice of Senga E cv. in 1997 to 6.9 g/L in juice

TABLE 4. Contents of L-malic acid in authentic and adulterated berry juices in 1998, 1999, 2000

Type of juice	Fruit variety	L-malic acid (g/L)			
		Year of fruit harvest			Average
		1998	1999	2000	
Strawberry juice	Senga	3.59	2.95	1.95	$2.83^{E}\pm0.83$
	Ducat	3.14	2.37	1.83	$2.45^{CD}\pm0.66$
	Marmolada	1.42	1.17	1.03	$1.21^{B}\pm0.20$
Raspberry juice	Beskid	0.56	0.54	0.45	$0.52^{A}\pm0.06$
	Canby	0.59	0.50	0.74	$0.61^{A}\pm0.12$
	Malling Seedling	0.65	0.39	0.92	$0.65^{A}\pm0.27$
Black currant juice	Ben Lomond	2.53	2.57	2.67	$2.59^{D}\pm0.07$
	Titania	2.75	2.36	2.74	$2.62^{D}\pm0.22$
	Ojebyn	2.17	2.38	2.37	$2.31^{C}\pm0.12$
Red currant juice	Rondom	14.56	11.92	10.81	$12.43^{J}\pm1.93$
	Jonker	7.98	2.07	1.39	$3.81^{G}\pm3.62$
	Holenderska	10.92	9.91	10.69	$10.51^{I}\pm0.53$
Adulterated raspberry juice	Beskid 70%+Senga 30%	1.46	1.32	0.90	$1.23^{B}\pm0.29$
Adulterated raspberry juice	Beskid 70%+Rondom 30%	3.75	3.16	3.09	$3.33^{F}\pm0.36$
Adulterated black currant juice	Ben Lomond 70%+Rondom 30%	4.96	3.59	4.27	$4.27^{H}\pm0.69$

A, B – values in the columns with different letters are significantly different at $\alpha=0.05$

of Senga SW cv. in 1998.

In the examined raspberry juices, the lowest content of L-malic acid – 0.39 g/L was found in juice of Malling Seedling cv. in 1999, and the highest one – 0.92 g/L – also in juice of Malling Seedling cv., but in 2000. Durst *et al.* [1995] determined higher concentrations of L-malic acid in raspberry juices. Juices of Malling Seedling cv. contained from 1.125 g/L to 1.737 g/L of this acid. The contents of L-malic acid in other juices were from 0.033 g/L in juice of Willamette cv. to 2.168 g/L in juice of Norna cv. Spanos & Wrolstad [1987] determined similar concentrations of L-malic acid in juices of Meeker (Oregon) – 0.529 g/L and 0.511 g/L, Meeker (Canada) – 0.887 g/L, Meeker (Washington) – 0.411 g/L, Willamette (Oregon) – 0.638 g/L, and 0.614 g/L and Skeena cv. (Canada) – 0.856 g/L, whereas higher concentrations of this acid – in juices of Willamete (Canada) – 1.928 g/L and Marcy cv. (New Zealand) – 1.062 g/L.

The contents of L-malic acid in black currant juices ranged from 2.17 g/L in juice of Ojebyn cv. in 1998 to 2.75 g/L in juice of Titania cv. in the same year. Of the black currant juices, the average contents of L-malic acid of three years examinations in juices of Titania (2.62 g/L) and Ben Lomond cv. (2.59 g/L) were significantly higher in comparison with the average content of this acid in juice of Ojebyn cv. (2.31 g/L). Gherardi *et al.* [1995] determined more differentiated concentrations of this acid in black currant juices, *i.e.* from 0.90 g/L to 3.07 g/L.

Red currant juices, in particular years, contained from 1.39 g/L of L-malic acid in juice of Joker cv. in 2000 to 14.56 g/L in juice of Rondom cv. in 1998. Statistical calculations indicated significant differences between the average contents of L-malic acid in juices of three red currant varieties. The average content of this acid of 1998, 1999, 2000 in juice of Jonker cv. was 3.81 g/L, in juice of Holenderska cv. – 10.51 g/L, and in juice of Rondom cv. – 12.43 g/L.

The results of determinations of L-malic acid concentrations in berry juices indicated that raspberry juices contained significantly lower levels of L-malic acid in comparison with strawberry and red currant juices, as well as that black currant juices contained significantly lower levels of this acid than red currant juices. Therefore, the contents of L-malic acid in adulterated raspberry and black currant juices were examined. A statistical analysis indicated significant differences between the average L-malic acid contents of three experimental years in authentic raspberry juice (0.52 g/L) and in raspberry juice adulterated with strawberry juice (1.23 g/L). The average L-malic acid content in raspberry juice with the addition of red currant juice was significantly higher (3.33 g/L) in comparison with the average content of this acid in authentic raspberry juice (0.52 g/L). The average content of L-malic acid in black currant juice adulterated with strawberry juice was significantly higher (4.27 g/L) than the average content of this acid in authentic black currant juice (2.59 g/L). Deviations from the range of L-malic acid concentration published in the Code of Practice were found at 30% addition of strawberry juice to raspberry juice in 1998 and 1999 (1.46 g/L and 1.32 g/L of L-malic acid, respectively). Exceeding the permissible values was also observed at 30% addition of red currant juice to raspberry juice in three years of examinations (3.75 g/L, 3.16 g/L and 3.09 g/L of L-malic acid), as well as at 30% addition of red currant juice to black currant juice in 1998 and 2000 (4.96 g/L and 4.27 g/L of L-malic acid, respectively). Therefore, L-malic acid content indicated 30% addition of strawberry and red currant juice to raspberry juice, as well as 30% addition of red currant juice to black currant juice.

The contents of citric acid, D-isocitric acid and L-malic acid in juices made of the same berry species were variable. It was due to differences in variety, stage of ripeness and climatic conditions. The high variability of determination results limits the usefulness of standard values specified in the Code of Practice for the examination of juice authenticity. Therefore values recommended in the Code of Practice should be updated.

CONCLUSIONS

1. It is necessary to complete the Code of Practice with the permissible contents of citric, D-isocitric and L-malic acid, as well as the citric acid:D-isocitric acid ratio in red currant juice.

2. Deviations from ranges of citric acid contents and citric acid:D-isocitric acid ratios accepted in the Code of Practice were found. Therefore, it is suggested that the lowest content of citric acid in strawberry juice be decreased to 4 g/L and in black currant juice – to 23 g/L, as well as that the limits of citric acid in raspberry juice be increased from 7 g/L to 26 g/L. Furthermore, it is proposed that the lowest citric acid:D-isocitric acid ratio in strawberry juice be decreased to 80 and the highest ratio of these two acids in raspberry juice increased to 260.

3. The content of L-malic acid can be used for the estimation of some berry juice adulterations.

4. Raspberry and black currant juice authenticity cannot be estimated on the basis of citric acid, D-isocitric acid contents and citric acid:D-isocitric acid ratio.

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WYKORZYSTANIE ANALIZY ZAWARTOŚCI WYBRANYCH KWASÓW ORGANICZNYCH DO WYKRYWANIA ZAFAŁSZOWAŃ SOKÓW Z OWOCÓW JAGODOWYCH

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W niniejszej pracy oznaczono metodami enzymatycznymi zawartości kwasu cytrynowego, D-izocytrynowego i L-jabłkowego w sokach otrzymanych z trzech odmian truskawek, malin, czarnych porzeczek i czerwonych porzeczek, a także w wybranych sokach z malin i czarnych porzeczek z 30% dodatkiem soków z truskawek i czerwonych porzeczek. Ponadto ustalono stosunki zawartości kwasu cytrynowego do zawartości kwasu D-izocytrynowego w tych sokach. Badania wykonano w trzech kolejnych latach: 1998, 1999 i 2000. Porównano otrzymane zawartości kwasów organicznych oraz stosunki kwasów z wartościami standardowymi zawartymi w Kodeksie Praktyki.

Stwierdzono odstępstwa od granicznych zawartości kwasu cytrynowego i stosunków zawartości kwasu cytrynowego do zawartości kwasu D-izocytrynowego przyjętych w Kodeksie Praktyki i zaproponowano zmiany tych wartości. Zawartości kwasu L-jabłkowego mogą być wykorzystane do oceny zafałszowań soków z malin i czarnych porzeczek. Na podstawie zawartości kwasu cytrynowego, kwasu D-izocytrynowego oraz stosunków zawartości kwasu cytrynowego do zawartości kwasu D-izocytrynowego nie można ocenić autentyczności tych soków.