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Antioxidants, Low Molecular Weight Carbohydrates, and Total Antioxidant Capacity in Strawberries (*Fragaria* × *ananassa*): Effects of Cultivar, Ripening, and Storage

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Four cultivars of strawberries (Senga Sengana, BFr77111, Elsanta, and Honeoye) were studied for their content of antioxidants, total antioxidant capacity, and low molecular weight carbohydrates in relation to harvest year, ripening stage, and cold storage. For ascorbic acid, chlorogenic acid, ellagic acid, and total antioxidative capacity, measured in both water-soluble and water-insoluble extracts, there was a 2−5-fold variation among cultivars. Unripe berries contained lower concentrations of chlorogenic acid and *p*-coumaric acid and also quercetin and kaempferol compared with riper berries. During cold storage for up to 3 days, relatively few changes in the concentration of the different antioxidants occurred. The concentrations of several investigated parameters were interrelated, for example, for ascorbic acid and water-soluble antioxidant capacity and for ellagic acid and water-insoluble antioxidant capacity. The dominating sugars in strawberries were fructose and glucose, but considerable amounts of sucrose were also present, and their contents varied among cultivars, giving a predicted glycemic index of ∼81. Verbascose, raffinose, and stachyose were found in only minor amounts. The study shows that the concentration of a number of bioactive compounds in strawberries varied according to cultivar, ripening stage, and storage. This information should make it possible to select strawberries with an optimal content of bioactive compounds.

KEYWORDS: Flavonol; ascorbate; dehydroascorbate; phenolic acids; hydroxycinnamic acids; ellagic acid; chlorogenic acid; sucrose; fructose; glucose; oligosaccharides; ABTS method; strawberry; *Fragaria* × *ananassa*

INTRODUCTION

Epidemiological evidence shows that a high dietary intake of fruits and vegetables is associated with a lower incidence of cancer and heart disease (*1*−*3*). Both the composition and content of the antioxidants and the carbohydrate pattern in foods may play a role in this respect. Antioxidants can scavenge free radicals and nonradical reactive oxygen species, which have the potential to damage cell components such as DNA, lipids, and proteins. Oxidative damage might be involved in the initiating events of cancer and atherosclerosis. In the past decades much interest has been focused on the antioxidant properties of vitamins C and E and carotenoids. Recently, other substances present in relatively high amounts in plant foods have attracted increasing interest, such as flavonoids and other phenolic compounds. Many of these substances are potent antioxidants and may exert other physiological effects, such as stimulating carcinogen-detoxifying enzymes or counteracting inflammatory processes (*4*−*6*). Also, the carbohydrates in food are of major interest in relation to chronic diseases (*7*−*9*). Different types of carbohydrates give rise to different glycemic responses, and the glycemic potential of glucose is, for example, higher than that of sucrose, which in turn is higher than that of fructose. Of special interest is the probable relationship between the glycemic index (GI) of a food and its ability to stimulate lipogenesis (*10*). A diet inducing high increments of blood glucose usually also leads to a larger stimulation of insulin secretion (*11*) and high levels of blood insulin have been shown to be correlated to high concentrations of very low density lipoproteins (VLDL) and low-density lipoproteins (LDL) and low concentrations of high-density lipoproteins (HDL) (*12*).

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Furthermore, indigestible carbohydrates (e.g., oligosaccharides and dietary fiber) entering the large intestine are fermented to short-chain fatty acids (acetic, propionic, and butyric acid), which have putative beneficial physiological effects. Propionic acid may lower plasma cholesterol concentrations (*13*), whereas butyric acid, the most important energy substrate for the colonic mucosa, has been suggested to play an important role in the prevention of colonic diseases, for example, cancer. Interestingly, different types of indigestible carbohydrates give rise to different profiles of short-chain fatty acids (*14*). Thus, the carbohydrate pattern is of great importance from a nutritional point of view.

Strawberries are an important berry crop in Sweden and several other countries (*15*). They have been reported to contain high amounts of fructose and glucose (*16*, *17*), and some studies have also reported considerable amounts of sucrose (*18*). However, data on the variation in carbohydrate composition between different cultivars and after storage are scarce. Studies on carrots (*19*) and cabbage (*20*) have shown considerable differences in carbohydrate composition between cultivars, and during storage the changes were even more pronounced. Besides the nutritional consequences, differences in carbohydrate pattern may also have considerable effects on the sensory properties of foods.

Strawberries are rich in vitamin C and also contain other compounds that may have physiological effects. Ellagic acid, present in relatively high levels in strawberries (*21*), has been suggested to exert antimutagenic and anticarcinogenic effects (*22*, *23*). Hydroxycinnamic acids exhibit antioxidant activity preventing lipid oxidation in human systems (*24*), whereas certain flavonoids have been shown to inhibit the potentially pro-oxidant enzymes lipoxygenase and xanthine oxidase, which generate free radicals (*4*). For the health effects it is relevant that the content of each antioxidant may vary with cultivar, growth stage, and environmental conditions (*6*, *25*, *26*).

Because strawberries contain many compounds with antioxidant effect, there is a need to find simple methods that will give a valid summary measure of their total antioxidant potential. Several methods have previously been used to measure the total antioxidant capacity (TAC) in strawberries. By the oxygen radical absorbance capacity (ORAC) procedure, strawberries were found to have the highest TAC among 12 fruits tested (*27*). Addition of vitamin C during jam preparation markedly increased TAC, but there was also a considerable antioxidant capacity left in samples in which ascorbic acid had decayed (*28*). Among 27 fruits in Singapore markets tested with the 2,2′-azinobis(3-ethylbenzothiazoline-6-sulfonate) (ABTS) method, only ciku (*Manilkara zapota*) had higher TAC than strawberries (*29*), and recently strawberries among other dietary plants were screened with the ferric-reducing antioxidant power (FRAP) method (*30*). In the present study, the ABTS method (*31*) was used.

In the work presented here the variation in the content of some antioxidants with cultivar and ripening stage was investigated, along with the effects of cold storage. The content of these antioxidants was also compared with measurements of TAC. The variation in the low molecular weight carbohydrate (LMWC) content and composition in different cultivars was investigated as was the effect of storage during postharvest shelf life. The possibility of enhancement in the level of antioxidants in strawberries by choice of cultivar or degree of ripening was explored.

Table 1. Characterization of the Strawberry Cultivars Honeoye and Senga Sengana at Various Ripening Stages and after Storage at 4 °C

		1999		2000	
day	ripening stage	pH	TSS[a]/°Brix, cv. H[b]	TSS/°Brix, cv. H[b]	TSS/°Brix, cv. SS[c]
1	unripe	3.40	7.6	8.9	6.7
1	ripe (red)	3.75	8.1	9.4	7.1
1	ripe (dark red)	3.43	7.3	9.9	8.0
2	ripe (red)	3.46	7.5	NM[d]	NM
3	ripe (red)	3.51	7.1	NM	NM

[a] Total soluble solids. [b] Cultivar Honeoye. [c] Cultivar Senga Sengana. Both cultivars were grown in Sweden. [d] NM, not measured.

MATERIALS AND METHODS

Fruit Samples. Strawberries (*Fragaria* × *ananassa)* of four different cultivars [Senga Sengana (S = Sweden), BFr77111, Elsanta, and Honeoye] were grown at the same farm in southern Sweden (Grödby, Sweden), using commercial cultivation practice. The berries were harvested in June during two consecutive years, 1999 and 2000. In addition, the cultivar Senga Sengana was grown in Poland (P) to investigate the possible influence of cultivation site. After harvest in the morning, the calyces were removed, and the strawberries, except those to be used for storage experiments, were frozen in an industrial air blast freezer until the center temperature reached −20 °C. The berries were then stored at −80 °C to preserve quality until later analysis.

Various Ripening Stages of Strawberries. Unripe (white-green), ripe (red = r), and fully ripe (dark red = dr) berries of the cultivar Honeoye (years 1999 and 2000) and Senga Sengana (year 2000) were harvested randomly in the same field on the same day in the morning.

Storage Treatments. The ripe (red) berries (cultivar Honeoye) were stored in a cooling room (4 °C) with a relative humidity of 85 ± 5% for up to 3 days. Portions of berries (500 g each) were frozen (year 1999, after 4, 24, 48, and 72 h; year 2000, after 6, 10, 24, 48, and 72 h) as described above. The frozen berries were vacuum packed and stored at −80 °C. In addition, in the year 2000, newly harvested berries were kept cold during transport and extracted on the day of harvest and analyzed for the content of flavonols, hydroxycinnamic acids, ascorbic acid, and TAC.

Analyses of Ascorbate, Dehydroascorbate, Flavonols, Hydroxycinnamic Acids, and Ellagic Acid. The samples for ascorbate analysis, 1.2 g, were homogenized with an ice-cold mortar and pestle in dim green light in a dark room and extracted in 4 mL of 2% (v/v) metaphosphoric acid, followed by additional 3 × 1 mL of the same solution. The samples were centrifuged at 16500*g* for 15 min at 4 °C. The supernatant was filtered through a C_{18} Sep-Pak column (Waters Corp.). The first 3 mL was discarded, and an aliquot of the following 1 mL was used for analysis by HPLC, as described by Wimalasiri and Wills (*32*), with some modifications as described below. The HPLC system consisted of a Waters 600 pump, a Maraton autosampler with a Rheodyne injector, and a Hewlett-Packard UV-1100 detector. A Waters carbohydrate analysis 3.9 × 300 mm column with a Waters C_{18} precolumn was used. The separation was performed with isocratic elution with a flow rate of 1.2 mL/min at room temperature for 5 min. The mobile phase was acetonitrile with 35% (v/v) 15 mM NH_4PO_4, adjusted to pH 4.3 with 1 M H_3PO_4. Detection was carried out at 248 nm. The peak of ascorbate in the samples was identified by comparing the retention time with that of an ascorbate standard. The dehydroascorbate concentration was determined by subtracting the ascorbate concentration from the total ascorbate concentration, obtained after the use of a reduction procedure (*33*). The samples used for the analysis of flavonols, non-cell-wall-bound hydroxycinnamic acids, and free ellagic acid were freeze-dried and analyzed according to the method of Häkkinen et al. (*34*), with some modifications as described below. A Waters XTerra RP18 5 μm, 3.9 × 150 mm column with a C_{18} precolumn was used. The mobile phase was a linear binary gradient with (A) 50 mM acetic acid and (B) acetonitrile/5% (v/v) methanol, with a flow rate of 1 mL/min. Detection was carried out at 254, 320, and 370 nm. The phenolics were identified at HPLC by their retention times and spectral data as compared by standards. The content of the

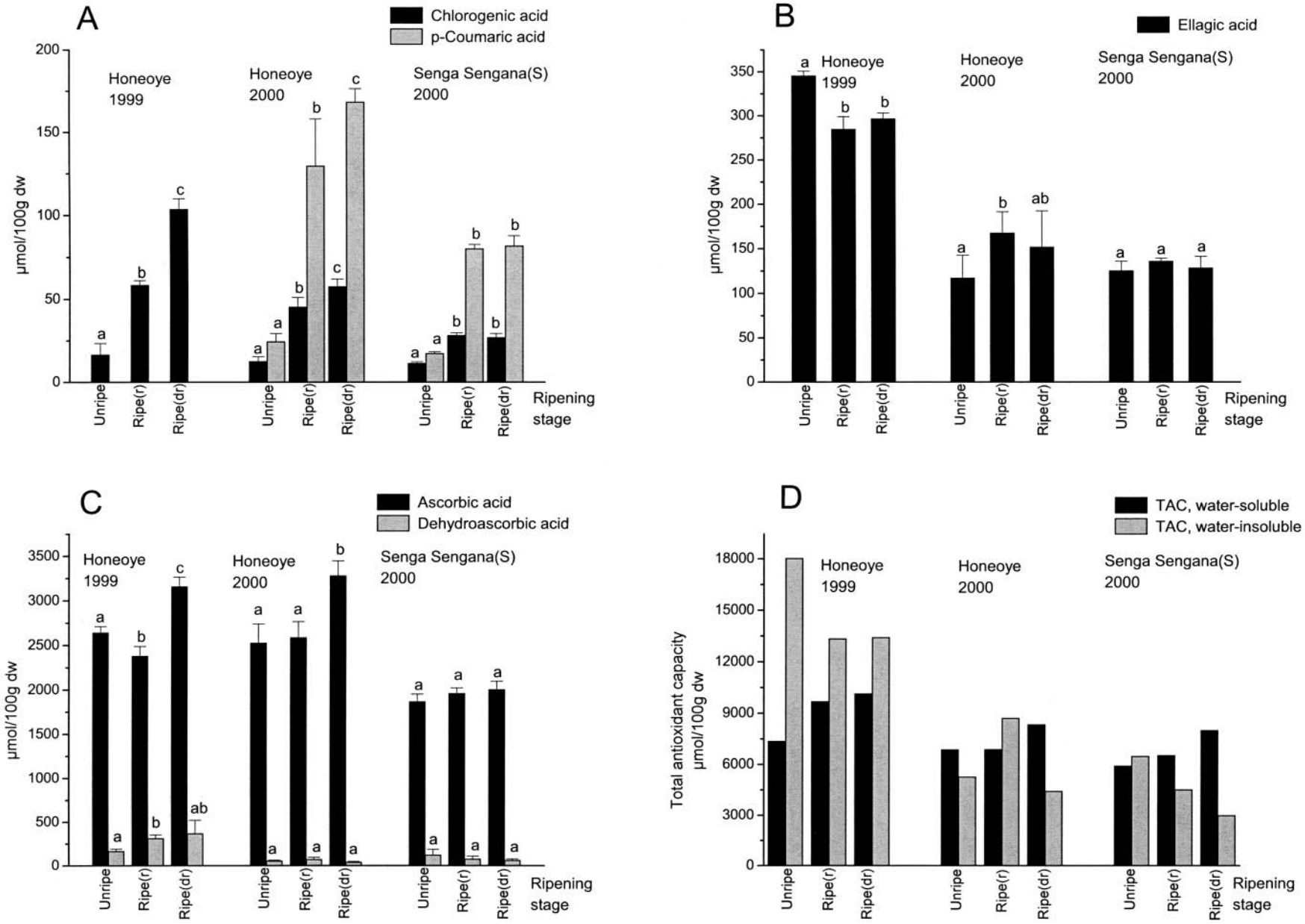

Figure 1. Contents of chlorogenic acid and *p*-coumaric acid (**A**), ellagic acid (**B**), and ascorbic acid and dehydroascorbic acid (**C**) and total antioxidant capacity (TAC; **D**) at different stages of ripening in two cultivars of strawberries. Results are presented as means ± SD based on three to four independent replicate samples. Means marked by different letters are significantly different ($p < 0.05$).

Table 2. Changes in the Content of the Flavonols Quercetin and Kaempferol with Ripening Stage and Storage[a]

ripening stage	quercetin			kaempferol			storage time/h	H-00[c]	
	H-99[b]	H-00[c]	SS-00[d]	H-99[b]	H-00[c]	SS-00[d]		quercetin	kaempferol
unripe	100	100	100	100	100	100	0	100	100
ripe (r)	217	381	178	176	342	132	6	93	105
ripe (dr)	281	663	143	215	437	108	10	75	88
							24	79	96
							48	81	96
							72	75	101

[a] Values are expressed as percent of the content in the unripe berries or as percent of the content before storage. [b] Cultivar Honeoye, year 1999. [c] Cultivar Honeoye, year 2000. [d] Cultivar Senga Sengana, year 2000. Both cultivars were grown in Sweden.

antioxidants was expressed as micromoles per 100 g of dry weight (dw) to make the values comparable to the values of TAC.

Total Antioxidant Capacity. The strawberries were thawed at room temperature for 1 h before homogenization for 2 min in a rotating blade homogenizer. The homogenate was centrifuged at 4 °C and 39000*g* for 30 min, and the supernatant was divided into aliquots and frozen at −80 °C. The pellet was extracted with acetone for 30 min, and the extract was centrifuged at room temperature for 10 min at 1240*g*. The acetone supernatant was frozen at −80 °C until analyzed. The total antioxidant capacity in both the water-soluble and water-insoluble supernatant was measured according to the ABTS method (*31*) at 734 nm using an Ultrospec 3000 UV−vis spectrophotometer. The vitamin E analogue Trolox (Sigma-Aldrich, St. Louis, MO) was used as a standard, and the results were expressed as micromoles of Trolox equivalents per 100 g of dw.

Analysis of Low Molecular Weight Carbohydrates. Each cultivar was prepared and analyzed for LMWC. To inactivate the cell-wall-bound invertase, the frozen strawberries were first put into water and microwave treated (900 W) for 2 min. The samples were then homogenized, and arabinose was added as an internal standard. LMWC were extracted in 80% (v/v) ethanol for 30 min at room temperature. The proteins were allowed to precipitate for 30 min at −20 °C and then separated by centrifugation (3000*g*, 2 °C, 15 min). The supernatant was withdrawn, evaporated, redissolved, and filtered (0.45 μm). The samples were analyzed using high-performance anion exchange chromatography with pulsed amperometric detection (HPAE-PAD). A NaOH gradient (16.5−200 mmol) was used as the eluent with a flow rate of 1.0 mL/min.

Other Measurements. pH was determined by a Radiometer pHC-2005 and total soluble solids by a hand refractometer (Atago, type 500).

Statistics. All values of antioxidants and LMWC were based on three to four independent replicate samples for each harvest. The results were expressed as the mean ± standard deviation (SD) and analyzed with Origin software (Microcal Software Inc.), MS Excel (Microsoft Inc.), and Minitab Statistical software (Minitab Inc.). The data of the antioxidant content were subjected to a one-way analysis of variance (ANOVA), and the level of significance used was $p < 0.05$. For statistical evaluation of possible differences in carbohydrate content between cultivars and after storage, the two-sided Student's *t* test for paired samples and ANOVA followed by Tukey's comparison were used. The standard error means were <5% for these analyses. Calculations of the linear correlation coefficient between TAC and individual antioxidants were made with SPSS 11.5 for Windows.

RESULTS AND DISCUSSION

Effect of Ripening Stage. Strawberries of the cultivars Honeoye and Senga Sengana harvested at different ripening stages had similar pH values and concentrations of total soluble solids (**Table 1**). Regarding individual bioactive compounds, chlorogenic acid has been reported to be present in strawberries (*35*), and it was also detected in the present investigation. With increasing degree of ripening, the content of chlorogenic acid as well as that of quercetin and kaempferol was enhanced in both investigated years in the cultivar Honeoye, whereas less difference could be seen in the cultivar Senga Sengana (**Figure 1**; **Table 2**). Also, the content of ascorbic acid was highest in the dark red berries of cultivar Honeoye, whereas no difference could be seen for Senga Sengana. The increase in vitamin C content with increasing ripening stage is in agreement with other studies (*36*). The content of hydroxycinnamic acids and ellagic acid was similar to data previously reported for strawberries (*21*, *27*, *37*), whereas the content of flavonols was lower. The lower levels could be due to the analytical method used. In the present study a semiquantitative method with simultaneous extraction and analysis of several substances was used, and the method was therefore not optimized for each of the analyzed compounds. No consistent variation could be found between the different stages of ripening in the content of dehydroascorbate or in ellagic acid (**Figure 1**). Maas et al. (*21*) found that the content of ellagic acid was higher in the pulp of green fruit than in the pulp of red fruit in 90% of 36 clones, but the same difference could not be found in the achenes from green or red fruit.

The water-soluble TAC increased from the unripe to the fully ripe (dark red) berries in both cultivars, whereas for the water-insoluble TAC there was an opposite tendency (**Figure 1**). For the cultivar Honeoye a similar tendency can be seen for ellagic acid and water-insoluble antioxidant capacity for both years, whereas the correlation was not so consistent for Senga Sengana for the year 2000, which might be due to higher concentrations of other antioxidants in this cultivar.

Cultivar Differences. There were large differences in the content of antioxidants among the four cultivars (**Figure 2**). In the cultivars grown in Sweden [Senga Sengana (S), BFr77111, Elsanta, and Honeoye], the contents of ellagic acid and chlorogenic acid were lower in the year 2000 compared with the year 1999, whereas the content of ascorbic acid was higher. Among cultivars, the content of ascorbic acid varied 2−3-fold, that of chlorogenic acid varied 3-fold, and that of ellagic acid varied 4−5-fold, but for the two latter substances there was a considerable variation between the two years. Maas et al. (*21*) found that the content of ellagic acid varied greatly among 36 cultivars and clones of strawberries examined.

The variation among cultivars in the investigated flavonols was larger in kaempferol than in quercetin, but the variation in the content of kaempferol was not consistent in the two years among the different cultivars (values not shown).

The effect of cultivation site was investigated in the cultivar Senga Sengana, grown both in southern Sweden and in Poland. There were differences between the two cultivation sites in the content of ascorbic acid, chlorogenic acid, kaempferol, quercetin (only year 2000, values not shown), ellagic acid, and water-insoluble TAC (only year 1999) (**Figure 2**).

Both the water-soluble and the water-insoluble TAC varied 2-fold among cultivars, and there was also some variation between the two investigated years within each cultivar (**Figure 2**). The investigated years, 1999 and 2000, were disparate concerning the weather and therefore the environmental condi-

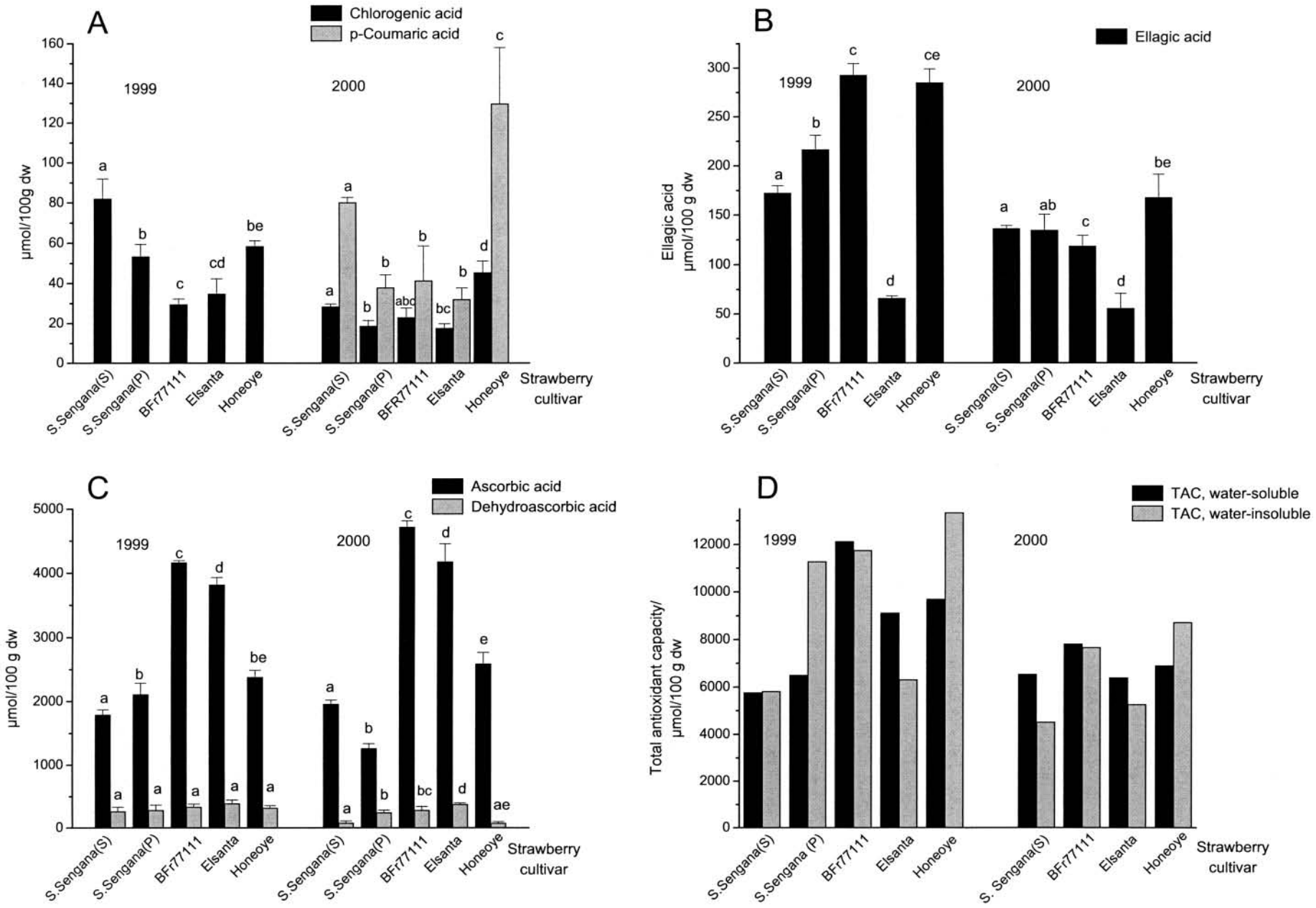

Figure 2. Contents of chlorogenic acid and *p*-coumaric acid (**A**), ellagic acid (**B**), and ascorbic acid and dehydroascorbic acid (**C**) and total antioxidant capacity (TAC; **D**) in four cultivars of strawberries. Cv. Senga Sengana was cultivated in both Sweden and Poland; the three other cultivars were cultivated only in Sweden. Results are presented as means ± SD based on three to four independent replicate samples. Means marked by different letters are significantly different ($p < 0.05$).

Table 3. Content of Low Molecular Weight Carbohydrates in Four Different Cultivars of Strawberries[a]

	SS (S)[b]	BFr77111	Elsanta	SS (P)[c]	Honeoye
glucose	313 a	258 b	317 a	261 b	321 a
fructose	363 a	324 bc	346 ab	308 c	368 a
sucrose	130 a	105 b	128 a	115 ab	20 c
raffinose	0.52 a	0.72 a	0.59 a	0.60 a	0.19 b
stachyose	*d*	*d*	0.19	*d*	*d*
verbascose	1.7 ab	1.6 bc	2.0 a	1.5 c	1.8 ab
∑ LMWC	808	689	794	686	711

[a] Values are expressed in mg/g of dw. Means followed by different letters are significantly different ($p < 0.05$). [b] Cultivar Senga Sengana, cultivated in Sweden. [c] Cultivar Senga Sengana, cultivated in Poland. [d] The result was below the detection limit of 0.1 mg/g of dw.

tions for the strawberries. The temperature was on average higher during spring 2000 than in spring 1999, with some periods of summer weather in the spring months of April and May 2000 (*38*, *39*). This might explain some of the differences in TAC, as well as in the content of some of the antioxidants, between the two years. There was some resemblance between the variation in ascorbate and water-soluble TAC among cultivars and also for ellagic and water-insoluble TAC, respectively, but because several other antioxidants occurred in the two fractions, a more detailed interpretation is not possible.

For comparison with literature data the TAC values were recalculated from micromoles per 100 g of dry weight to micromoles per 100 g of fresh weight (fw). The values then varied between 430 and 900 μmol/100 g of fw (water-soluble TAC) and between 390 and 1040 μmol/100 g of fw (water-insoluble TAC). Leong et al. (*29*) found the antioxidant capacity in strawberries to be 2680 ± 530 μmol/100 g of fw when measured with the ABTS method, with ascorbic acid as standard. This was 3 times more than the water-soluble TAC found in the present study. Also, Viberg (*28*) used the ABTS method and found the water-soluble TAC in strawberries to be 1300 μmol/100 g of fw. Using the FRAP method Halvorsen et al. (*30*) found the TAC to be 1850 μmol (Fe^{2+})/100 g of fw in the strawberry cultivar Senga Sengana and 2330 μmol (Fe^{2+})/100 g of fw in the cultivar Honeoy (Fe^{2+} was used as standard). Because Trolox had double the activity compared to Fe^{2+} in the FRAP method (*40*), these results could be recalculated to 925 and 1165 μmol/100 g of fw in the unit used by us and thus were comparable with the results obtained in the present study. When strawberries were analyzed with the FRAP method (J. Nilsson, unpublished data), the water-soluble TAC varied between 370 and 810 μmol/100 g of fw and the water-insoluble TAC between 240 and 820 μmol/100 g of fw, and there were high correlations between the results from ABTS and FRAP methods, although the proportionality factor deviated from one for the water-insoluble TAC, the ABTS method giving the higher values. A third method, ORAC, has been used to measure the TAC in strawberries, and with this procedure the TAC in strawberries was 1536 ± 238 μmol/100 g of fruit, results from water-soluble and water-insoluble extracts being added (*27*). Some of the variations among data from these studies are probably explained by methodological differences, for example, concerning the extraction technique and absorbance reading time. In general, all methods seem to give data in the same range. The present study shows that a large part of the variations occurs among varieties.

The total amount of LMWC was between 686 and 808 mg/g of dw (**Table 3**), which is at a similar level as that reported by others (781 mg/g of dw) on 10 cultivars from different parts of Sweden (*16*). The dominating sugars in strawberries were fructose (308−371 mg/g dw) and glucose (258−321 mg/g dw), but considerable amounts of sucrose (20−130 mg/g dw) were also present (**Table 3**). Altogether LMWC amounted to ∼70−80% of the total dry weight. Senga Sengana (S) and Elsanta had the highest contents of sugars (**Table 3**), which may lead to a higher sweetness of these cultivars. A high fructose content of strawberries may also be of importance for people with functional bowel diseases. The amount of fructose from half a liter of strawberries may be 15−20 g as calculated from the analyses of carbohydrates, which is an intake that has been reported to cause discomfort in sensitive patients (*41*). The load may vary depending on the cultivar selected. Except for Honeoye, which had a very low content of sucrose (20 mg/g dw), the distribution of the sugars was similar for the different cultivars, 37−40% for glucose, 44−47% for fructose, and 15−17% for sucrose, which also agrees quite well with the results of Fuchs and Wretling (*16*), suggesting a similar glycemic index for the cultivars investigated. When using earlier reported GI values for glucose, fructose, and sucrose (*42*), a predicted GI value of 80−83 was found for the cultivars from the carbohydrate analyses in the present study.

The content of α-galactosides was low. Among these, verbascose was the dominating oligosaccharide. Raffinose and stachyose were found in only minor amounts, of which the latter was below the limit of detection in some cultivars.

Impact of Cold Storage. Ripe (red) strawberries of the cultivar Honeoye were stored at 4 °C for up to 3 days. The content of ascorbic acid was highest in the fresh berries, whereas in the stored and then frozen berries an increase was found during the first day (1999, 4−24 h; 2000, 10−24 h), but no statistically significant changes could be found during storage

Table 4. Content of Some Antioxidants and Total Antioxidant Capacity during Storage of the Cultivar Honeoye at 4 °C[a]

	ascorbic acid	dehydroascorbic acid	*p*-coumaric acid	chlorogenic acid	ellagic acid	TACws[b]	TACwis[c]
1999							
4 h	2380 ± 111	314 ± 40	NM[d]	NM	NM	9680	13330
24 h	2740 ± 57	176 ± 70	NM	NM	NM	8620	10660
48 h	2866 ± 43	310 ± 17	NM	NM	NM	7420	9740
2000							
0 h	3170 ± 181	241 ± 38	73.8 ± 5.3	43.7 ± 4.6	145 ± 14	5900	11140
6 h	2430 ± 146	228 ± 14	74.4 ± 8.2	42.6 ± 1.5	136 ± 16	4760	5450
10 h	2100 ± 70	166 ± 26	71.7 ± 3.8	38.9 ± 1.4	145 ± 2	5030	3730
24 h	2770 ± 260	229 ± 9	64.4 ± 1.3	38.6 ± 2.6	134 ± 80	5820	6150
48 h	2600 ± 83	212 ± 48	61.2 ± 13.1	33.1 ± 8.6	144 ± 43	6970	6200
72 h	2810 ± 214	184 ± 42	58.4 ± 10.7	31.8 ± 7.5	198 ± 41	6440	4950

[a] Values are the means of three to four replicates, ± SD, expressed in μmol/100 g of dw. [b] Water-soluble total antioxidant capacity (μmol/100 g of dw). [c] Water-insoluble total antioxidant capacity (μmol/100 g of dw). [d] NM, not measured.

Table 5. Content of Low Molecular Weight Carbohydrates in the Cultivar Honeoye during Storage at 4 °C[a]

	0 h	24 h	48 h
glucose	321 b	321 b	326 a
fructose	368 b	369 b	401 a
sucrose	20	20	20
raffinose	0.19 a	0.19 a	0.17 a
stachyose	*b*	*b*	*b*
verbascose	1.8 a	1.8 a	1.8 a
∑ LMWC	711	712	749

[a] Values are expressed in mg/g of dw. Means followed by different letters are significantly different ($p < 0.05$). [b] The result was below the detection limit of 0.1 mg/g of dw.

thereafter (**Table 4**). This good retention of vitamin C during cold storage of strawberries is in accordance with the results of Hägg et al. (*37*). There was a decrease in the content of chlorogenic acid and quercetin, but no statistically significant change could be found for kaempferol or ellagic acid (**Tables 2** and **4**). Gil et al. (*36*) reported that the content of ellagic acid, although calculated per gram of fresh weight, increased slightly during storage for 5 days in 5 °C.

Both water-soluble and water-insoluble TAC tended to decrease during 2 days of storage in 1999, but this change was not consistent in the year 2000. A similar time course in ascorbate and water-soluble TAC values upon storage was found in the 2000 samples, but otherwise we could not find any explanation for the variations.

The contents of glucose and fructose tended to be higher after the second day of cold storage (**Table 5**). The higher content of fructose and glucose could be due to a breakdown of sucrose and other higher forms of carbohydrates. Hägg et al. (*36*) reported that the sugar content (glucose, fructose, and sucrose) in strawberries stored in 5 °C increased somewhat during 4 days of storage of unripe (degree of ripeness 3/4) berries, whereas the content decreased slightly in ripe berries. The differences in the results between the study of Hägg et al. and the present investigation might be due to how the stage of ripeness was defined.

Interrelationships among Bioactive Compounds. To study whether the contents of different compounds co-varied, linear correlations were computed for the whole material with a focus on total antioxidant capacity. The concentration of ascorbic acid was significantly associated with the water-soluble antioxidant capacity ($r = 0.47$, $P = 0.022$) and that of ellagic acid with the water-insoluble antioxidant capacity ($r = 0.82$, $P = 0.000$) (**Figure 3**), suggesting that these antioxidants are important components when total antioxidant capacity is measured. For ascorbic acid this was expected from its concentration accounting for a large part of the water-soluble antioxidant capacity. Additional antioxidants must have occurred in both fractions, which may explain why the correlation coefficients were not even higher.

Consistent Changes for the Two Investigated Years. Remarkable variations in the contents of antioxidants were shown for different years, cultivars, ripening stages, and conditions of storage. Some of these differences were consistent for the two years, for instance, the levels of chlorogenic acid and *p*-coumaric acid, which were lowest in the unripe stage in both of the investigated cultivars (**Figure 1**). Contrary to the declines in the content of ascorbate that has been reported in some green vegetables after harvest (*43*−*45*), the changes in the content of this antioxidant during storage of strawberries were only modest in the present study (**Table 4**). The study

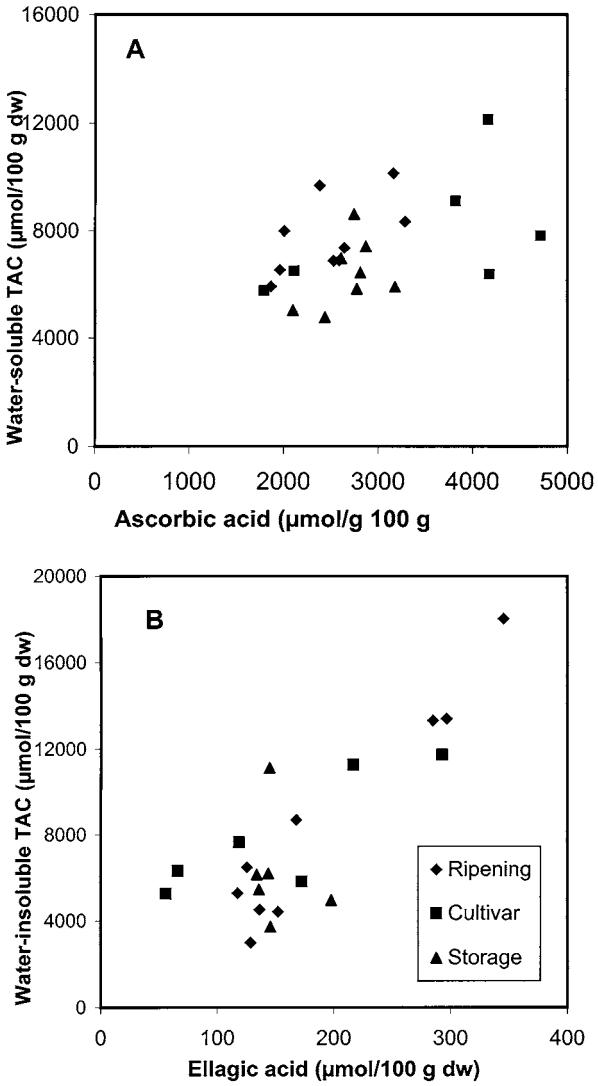

Figure 3. Correlations between antioxidants and TAC in the three different parts of the study (ripening, cultivar, and storage): (**A**) amount of ascorbic acid in relation to water-soluble TAC; (**B**) amount of ellagic acid in relation to water-insoluble TAC.

also shows that measurement of water-soluble and water-insoluble total antioxidant capacity are good markers of ascorbic acid and ellagic acid content, respectively.

The cultivar Honeoye had the highest or second highest content of chlorogenic acid, *p*-coumaric acid, and ellagic acid for the two investigated years, whereas the cultivar Elsanta had the lowest or second lowest content of these antioxidants but had the second highest content of ascorbic acid (**Figure 2**). It thus seems possible to pick high-content or low-content cultivars, for certain antioxidants or groups of antioxidants, even if there is a variation between years, probably due to different environmental conditions. This might be of interest in the future, when treatment with targeted antioxidants for certain health conditions, or prevention of these conditions, might be a possibility, consisting of a diet containing high or low amounts of specific antioxidants in plant foods.

ABBREVIATIONS USED

ABTS, 2,2′-azinobis(3-ethylbenzothiazoline-6-sulfonate); ANOVA, one-way analysis of variance; dw, dry weight; FRAP, ferric-reducing antioxidant power; GI, glycemic index; HDL, high-density lipoproteins; HPAE-PAD, high-performance anion

exchange chromatography with pulsed amperometric detection; LDL, low-density lipoproteins; LMWC, low molecular weight carbohydrates; ORAC, oxygen radical absorbance capacity; SD, standard deviation; TAC, total antioxidant capacity; VLDL, very low density lipoproteins.

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