Rose Petal Tea as an Antioxidant-rich Beverage: Cultivar Effects

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ABSTRACT: Twelve rose cultivars were selected by field tasting as potential sources of edible flowers. Hot water infusions (teas) of air-dried petals of these cultivars were assayed for antioxidant activity, total phenols, and total anthocyanins contents. Their composition was analyzed by high-performance liquid chromatography (HPLC). Green tea was tested in parallel as a reference antioxidant-rich beverage. Rose petal teas from different cultivars exhibited scavenging capacity toward 2,2′-azino-bis-(3-ethylbenzothiazoline)-6-sulfonate cation radical (ABTS+) ranging between 712.7 and 1770.7 μ*M* Trolox equivalents (TE) per gram of dry petals, as compared with 1227.6 μ*M* TE/g dry weight in the green tea. The range of total phenols content in rose teas was 50.7 to 119.5 mg gallic acid equivalents (GAE) per gram of dry matter, as compared with 62.1 mg GAE/g dry weight in the green tea. The rose teas were rich in free gallic acid. The highest values of antioxidant activity, total phenols, and gallic acid contents were found in the cultivars San Francisco, Katharina Zeimet, and Mercedes and in the essential-oil-bearing rose *Rosa damascena*. The correlation coefficients between antioxidant activity, on the 1 hand, and the contents of total phenols and of gallic acid in various rose cultivars, on the other hand, were 0.79 and 0.81, respectively. No clear relationship between anthocyanin level and radical-scavenging activity was revealed. Teas from different rose cultivars significantly differed in their sensory properties. It was concluded that dried rose petals may be used for preparing antioxidant-rich caffeine-free beverages, either separately or in combination with other herbal materials.

Keywords: rose, tea, antioxidant, phenol compounds, sensory analysis

Introduction

Besides the well-recognized wholesome effects of green and black teas prepared from young leaves of *Camellia sinensis*, hot water infusions (teas) of many other plants also may have health benefits. In particular, the tea of rose flowers (*Rosa* sp. var. Rosa de Castillo) was reported to possess 1 of the strongest antioxidant activities among the 30 medicinal plant teas tested (VanderJagt and others 2002). The recent work of Ng and others (2004) linked the antioxidant activity in an aqueous extract of rose flowers (*Rosa rugosa*), primarily with the presence of a phenolic compound identified as a gallic acid derivative. Antioxidants of a polysaccharide structure were also found by VanderJagt and others (2002) but exhibited lower activity. Phenolic compounds were associated with radical-scavenging activity in flower extracts of *R. rugosa* and *R. davurica* (Cho and others 2003).

In addition to antioxidant activity, water extracts of rose flowers were reported to possess anti-inflammatory and analgesic (Choi and Hwang 2003), antibacterial (Anesini and Perez 1993; Perez and Anesini 1994), antiviral (Mahmood and others 1996), and antifungal (Dixit and others 1976; Tripathi and Dixit 1977; Anesini and Perez 1993) effects. In the last 2 studies, the active compound was isolated from the extract and identified as gallic acid.

Roses are known as edible flowers and have been used for centuries as food components, either in the fresh form or in processed products, such as confectionary and beverages (Girard-Lagorce and others 2001). The combination of health benefits with recognized applicability in cuisine raises the possibility of using rose flowers in functional food products. However, the available knowledge about the health-promoting potential of roses is still insufficient. In particular, no comparative study has been undertaken on the effects of the vast natural and breeding-related genetic variability of roses on their health value. Various representatives of the genus *Rosa* were investigated in the studies cited previously; they include *R. hybrida* (Choi and Hwang 2003), *R. borboniana* (Anesini and Perez 1993; Perez and Anesini 1994), *R. chinensis* (Tripathi and Dixit 1977), *R. indica* (Dixit and others 1976), and *R. damascena* (Mahmood and others 1996). The impression is that in many cases, the choice of any particular rose species or cultivar for investigation was determined solely by accessibility of plant material. Similarly, no information is available in the scientific literature regarding the effect of plant genotype on the sensory characteristics of edible products derived from rose flowers.

The aim of the present study was to compare antioxidant activity, chemical composition, and sensory characteristics of teas brewed from petals of various rose cultivars to study the genetic variability of these parameters and to choose the genotypes most suitable for utilization in preparation of the antioxidant beverage.

Materials and Methods

Chemicals

The gallic acid standard was purchased from MP Biomedicals, Inc. (Eschwege, Germany). The 2,2′-azino-bis-(3-ethylbenzothiazoline)-6-sulfonic acid (ABTS) was from Sigma-Aldrich (St. Louis, Mo., U.S.A.). Sigma-Aldrich (Milwaukee, Wis., U.S.A.) supplied the following compounds: epicatechin, catechin, the free-radical generator 2,2′-azobis-(2-amidinopropane) dihydrochloride (AAPH) (synonym: 2,2′-azobis(2-methylpropionamidine) dihydrochlo-

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Table 1—Rose cultivars used in the study

Cultivar	Type	Petal color	Fragrance
San Francisco	Hybrid tea	Flame-red	Mild
Brandy	Hybrid tea	Apricot-orange	Mild
Maxim	Hybrid tea	Pink and white	Strong
Sweet Surrender	Hybrid tea	Pink-silver	Strong
English Sachet	Hybrid tea	Light pink-white	Strong
Mount Shasta	Hybrid Tea	Cream-white	Strong
Damascena	*Rosa damascena*	Pink	Strong
Pat Austin	English rose	Copper	Strong
Golden Celebration	English rose	Yellow	Strong
Katharina Zeimet	Polyantha	White	Strong
Trier 2000	Floribunda	Light pink	Mild
Mercedes	Floribunda	Scarlet	Mild

ride), the reference antioxidant Trolox (6-hydroxy-2,5,7,8-tetramethyl-chroman-2-carboxylic acid), and Folin-Ciocalteu phenol reagent. The high-performance liquid chromatography (HPLC)-grade solvents (water and methanol) were from BioLab Ltd (Jerusalem, Israel).

Plant material

Primary selection of cultivars with potentially edible flowers was made by field tasting of fresh petals conducted by 5 members of the research team at the Keren Zur nursery in southwest Galilee, Israel. The evaluation was made by hedonic test according to 9-point hedonic scale of overall liking (from score 1 "dislike extremely" to score 9 "like extremely"). Eleven cultivars were selected because of their pleasant petal taste and texture (scores equal to or greater than 6). In addition, the Mercedes cultivar was taken as a reference because of the high antioxidant activity revealed in preliminary trials, in spite of the low taste score given to this variety because of its petal bitterness. The list of selected cultivars and their characteristics are presented in Table 1.

The flowers of 12 selected cultivars, not treated with pesticides, were picked for tea preparation at the same nursery. Whole flowers were brought to the laboratory within 3 h after picking, and the petals were detached, spread in a thin layer on a plastic net, and dried in the shade at room temperature (25 °C to 27 °C). The air-dry material was stored in hermetically closed plastic containers in a freezer at –20 °C. The green tea sample branded as "Traditional, Unfermented Chinese green tea" (Wissotzky Tea Ltd., Wissotzky House, Tel Aviv, Israel) was purchased from a local store. The residual moisture content in air-dry material was determined by oven drying to constant weight at 80 °C and was found to be 2% to 3%.

Brewing procedure

The air-dry plant material (rose petals or tea leaves) were steeped in boiling water in 15-mL screw-capped plastic centrifuge tubes (10 mL water per 100 mg air-dry material) and infused in a water bath at 85 °C with shaking every 5 min. The infusion time varied from 1 to 30 min. Each sample was prepared at least in triplicate. The tubes were centrifuged for 5 min at 1000 × *g* and the supernatant was separated by filtration through 13-mm Millex-HN syringe filter units of 0.45-μm pore size (Millipore, Billerica, Mass., U.S.A.). The filtered supernatant was used for the analytical procedures as described subsequently.

Evaluation of antioxidant activity

Antioxidant activity was assayed as the scavenging capacity for the stable cation radical ABTS+ according to Araki and others (1999). The radical was prepared by incubating the reaction mixture comprising 75 μ*M* ABTS and 2 μ*M* AAPH (radical initiator) in 50 m*M* acetate buffer (pH 4.3) for 60 min at 45 °C. The resulting solution, containing blue-colored ABTS+ cation radical, was cooled to 21 °C to 23 °C and transferred to plastic cuvettes measuring 10 × 4 × 45 mm (ref. nr 67.742, Sarstedt, Numbrecht, Germany), 1 mL per cuvette. Ten microliters of analyzed samples were added to each cuvette and thoroughly mixed with a pipette tip. The absorbance of the radical solution at 734 nm was measured 15 min after addition of the sample with a GBC 911 UV-visible spectrophotometer (GBC Scientific Equipment Pty Ltd, Dandenong, Victoria, Australia). The solution of Trolox (1 m*M*) in acetate buffer pH 4.3 was used as a standard and acetate buffer pH 4.3 as a blank sample. The calculation of the Trolox equivalent antioxidant capacity (TEAC) of the samples was based on decoloration of the ABTS+·solution according to the formula given below, where A represents the absorption at 734 nm and C the concentration in m*M*:

$$\text{TEAC (m}M\text{)} = C_{standard} \times (A_{sample} - A_{blank}) \,/\, (A_{standard} - A_{blank})$$

Because the radical-scavenging capacity of a tea obviously depends on its strength (the amount of plant material used in brewing), the antioxidant activities of the teas were expressed as TEAC per gram of absolutely dry petals.

Analytical methods

The total content of phenolic compounds in the teas was determined with Folin-Ciocalteu phenol reagent according to Singleton and Rossi (1965) and expressed in gallic acid equivalents (GAE) as milligrams GAE per gram of absolutely dry petals.

The chromatographic analysis of teas was conducted with a Shimadzu HPLC system fitted with an SPD-M10A photodiode array detector (Shimadzu, Kyoto, Japan) and a Hypersil ODS (C-18) column measuring 250 × 4.6 mm, 5 μ*M* (Phenomenex, Torrance, Calif., U.S.A.); the wavelength detection range was 240 to 380 nm; the solvent system was water/methanol/acetic acid (79/20/1, v/v) flowing at 1.5 mL/min. Peak identification was based on matching retention times and ultraviolet spectra against those of standard reference compounds. The quantification of identified compounds (in particular, of gallic acid) was done by external standard method.

The total anthocyanin contents in the teas were determined by the pH differential method of Fuleki and Francis (1968) according to Giusti and Wrolstad (2001), with a slightly modified pH adjustment procedure. The freshly brewed teas had an initial pH about 4.7 to 4.9. Each tea sample was divided into 2 portions and their pH values were adjusted either to 4.5 by adding 1 *N* HCl or to 1.0 with 12 *N* HCl. The method of pH adjustment by buffer dilution originally developed for anthocyanin-rich berry juices (Fuleki and Francis 1968) was not applicable to teas because of their initially high dilution. The absorbance of samples at 2 pH levels was measured at 520 nm and 700 nm with the GBC 911 UV-visible spectrophotometer, and the anthocyanin concentration was calculated as milligrams of cyanidin-3-glucoside per gram of absolutely dry matter of the brewed petals, using molar extinction coefficient of 26900 and a molecular weight of 449.2.

Tea sensory evaluation

The choice of cultivars for sensory evaluation was based on the results of antioxidant activity analysis as explained in the Results and Discussion section. Sensory characteristics of 4 tea samples from different cultivars were evaluated by Quantitative Descriptive Analysis (QDA) using the 15-cm non-anchored paper scale and criteria defined at a preliminary session (Mason and Nottingham 2003). The analysis was performed at a sensory evaluation facility

at the Dept. of Postharvest Science of Fresh Produce, ARO – the Volcani Center by a panel of 6 trained evaluators (2 female, 4 male, age 40 to 53 y) belonging to the department staff. The facility had a controlled environment (temperature, illumination) and a separate booth for each panelist. The tea samples were prepared (brewed) as described previously but without centrifugation and syringe filtration, in amounts sufficient for sensory evaluation (400 mL of each tea sample). Serving size was 50 mL. The taste and aroma of the tea samples were evaluated both when hot (within 2 to 4 min after brewing) and after cooling to room temperature. The tasting was performed under red light illumination to neutralize the effect of tea color on the evaluation results.

Statistics

All tests described previously were conducted at least in triplicate and repeated 2 to 4 times, with similar results. The results of 1 representative trial are shown. The mean and 95% *t*-confidence interval were used to describe the variability of the analysis results. Pearson's correlation coefficient (*r*) was calculated to determine the linear correlation between antioxidant activity and chemical characteristics of petal teas prepared from the various rose cultivars. The relationship between total phenolics content and antioxidant activity was characterized also by linear regression analysis. Analysis of variance (ANOVA) was used to examine the effects of cultivar on the sensory characteristics of rose petal teas. The calculations were performed by means of the Microsoft Office Excel spreadsheet in combination with the Analyze-it General Statistical Package, version 1.71 (Analyze-it Software Ltd., Leeds, U.K.).

Results and Discussion

Infusion time effects

The extraction procedure was optimized by measuring antioxidant activities of tea samples after various infusion durations (Figure 1). With the green tea, no statistically significant difference was found between the samples extracted (brewed) for the periods from 5 to 30 min. On the other hand, with rose tea samples (cv. San Francisco) the maximal radical-scavenging activity was reached after 20 min of brewing. This duration was used in further experiments for preparing both green tea and rose petal tea samples.

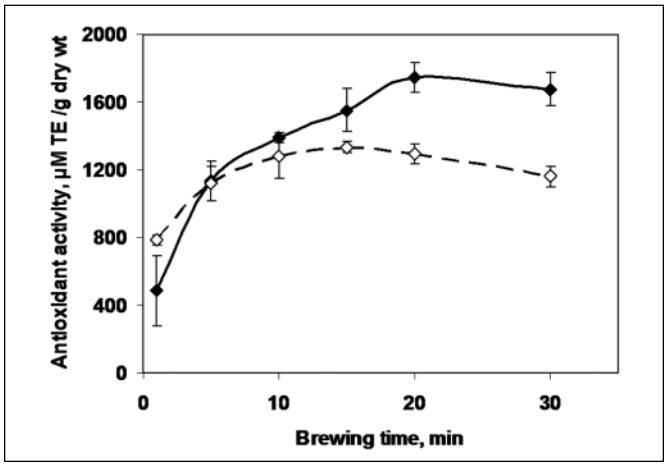

Figure 1—Effect of infusion duration on the antioxidant activity values of the green tea (the dotted line) and of the rose petal tea of San Francisco cv.(the solid line). The antioxidant activity expressed as Trolox equivalents.

Antioxidant activity

Rose petal teas exhibited high antioxidant activity, comparable with that of the green (Figure 2) and the black teas (data not shown). The cultivars varied in their antioxidant activity. For example, the tea prepared from cv. San Francisco roses was twice as effective in ABTS+·radical scavenging as that prepared from cv. Mount Shasta, although the 2 cultivars belong to the same hybrid tea group. Other rose genotypes with outstanding antioxidant activity (equal to or greater than that of green tea) were Katharina Zeimet (polyantha), Mercedes (floribunda), and *R. damascena*. The tea prepared from cv. Golden Celebration, of the English roses group, consistently showed the lowest radical scavenging efficacy among the samples tested. The hot water did not completely extract all antioxidant compounds present in rose petals. Additional activity (30% to 50% of that found in the tea) could be extracted from the petals by organic solvents, for example, methanol, acetonitrile, dimethyl sulfoxide (data not shown).

Total phenolics

In most of the rose teas tested, the total content of phenolic compounds was equal to or higher than that in the green tea (Figure 3), and conformed to a pattern similar to that of the antioxidant activity. The relationship between the phenolics content and the ABTS+ radical scavenging activity tended to be linear (Figure 4), with a correlation coefficient *r* between the 2 parameters of 0.79 (signif-

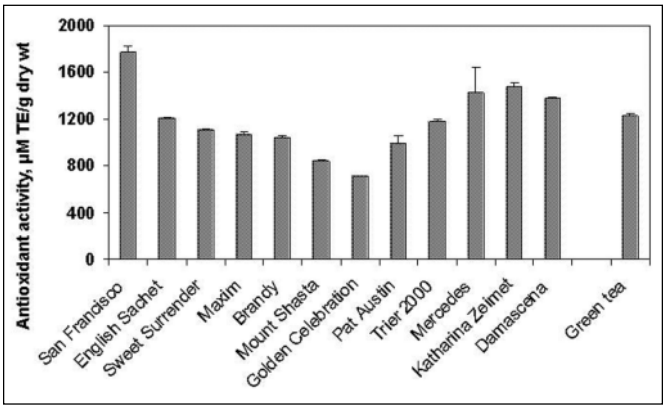

Figure 2—Antioxidant activity (as Trolox equivalents) in the teas prepared from dried petals of various rose cultivars

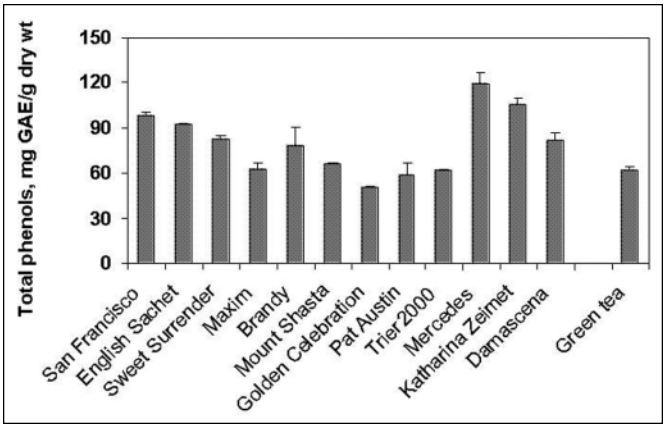

Figure 3—Total contents of phenolic compounds (expressed as gallic acid equivalents) in the teas prepared from dried petals of various rose cultivars

icant at the 1% level). The cultivars Mercedes, Katharina Zeimet, and San Francisco produced teas with the highest total phenolics contents, and that from cv. Golden Celebration the 1 with the lowest (Figure 3).

Gallic acid

Free gallic acid was found in the teas prepared from various rose cultivars (Figure 5). The comparison of gallic acid content (Figure 6) with total phenolics content (Figure 3) showed that in some cultivars (San Francisco, Maxim, Brandy, Katarina Zeimet, *R. damascena*) the free gallic acid accounted for 35% to 55% of the total phenolics. At the same time, in other cultivars (Sweet Surrender, Golden Celebration) and in the green tea both absolute and relative content of free gallic acid was rather low, accounting for less than 10% of total phenolics (Figure 6). The correlation coefficient, *r*, between antioxidant activity and gallic acid content was 0.81 (significant at the 1% level). No peaks corresponding to catechin or epicatechin were revealed by HPLC analysis of rose teas, but they were present in the green tea (data not shown).

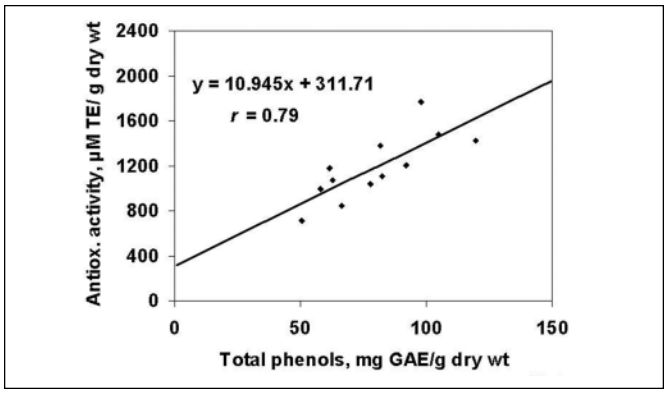

Figure 4—The relationship between total content of phenolic compounds and antioxidant activity in the teas prepared from dried petals of various rose cultivars

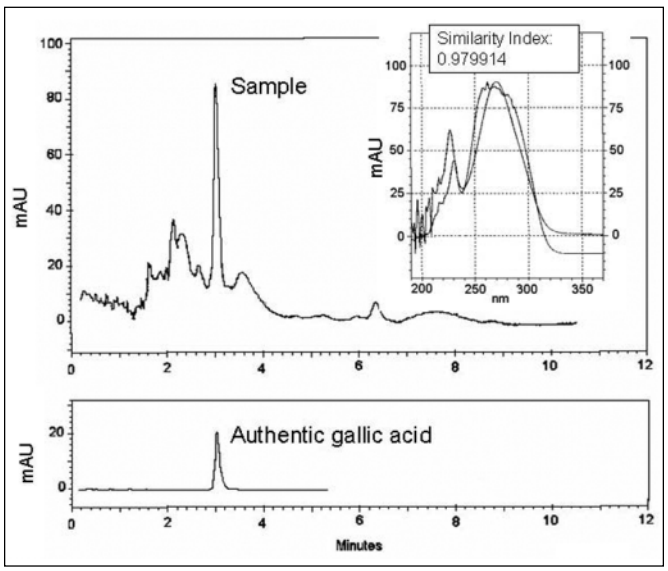

Figure 5—High-performance liquid chromatography (HPLC) chromatograms of the 'San Francisco' rose tea sample (above) and of gallic acid standard (below). Detection wavelength 280 nm. In the insert: the comparison between the UV absorption spectra of the gallic acid standard and of the putative gallic acid peak in the rose tea sample.

Anthocyanins

Obviously, the total anthocyanin content in rose teas was correlated with the petal color, with relatively high values in those prepared from the red-flowered cultivars, San Francisco and Mercedes (Figure 7). The relationship between petal color and antioxidant activity was not clear-cut: on the 1 hand, within the group of hybrid tea roses the red cultivar San Francisco exhibited the highest activity, the light-colored (pink, apricot) cultivars were intermediate, and the white cultivar Mount Shasta exhibited the lowest radical-scavenging activity; on the other hand, another white-flowered cultivar, Katharina Zeimet (polyantha group), exhibited 1 of the highest activity levels among the samples tested. One can calculate by comparing the data of Figure 2 and Figure 6 that anthocyanins accounted for about 10% of total phenolics in the tea from cv. San Francisco roses. In the other cultivars, the contribution of anthocyanins to total phenols and, presumably, to the antioxidant activity was minor.

Sensory evaluation

Four cultivars, representing various rose groups and differing in antioxidant activity, total phenolics and gallic acid content, were chosen for sensory evaluation: San Francisco, Katharina Zeimet, Golden Celebration, and *Rosa damascena*. Only the San Francisco tea exhibited slight pink coloration after brewing, at its natural pH of about 4.8. Slight acidification to pH 3 to 4 (for example, with citric acid) markedly enhanced the red/pink color in all tea samples.

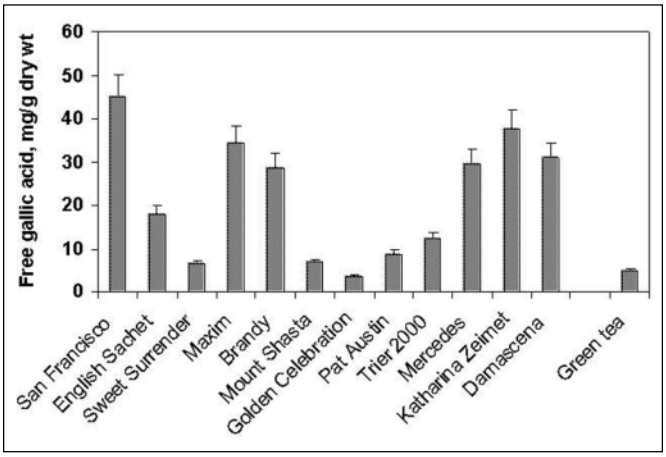

Figure 6—Contents of free gallic acid in the teas prepared from dried petals of various rose cultivars

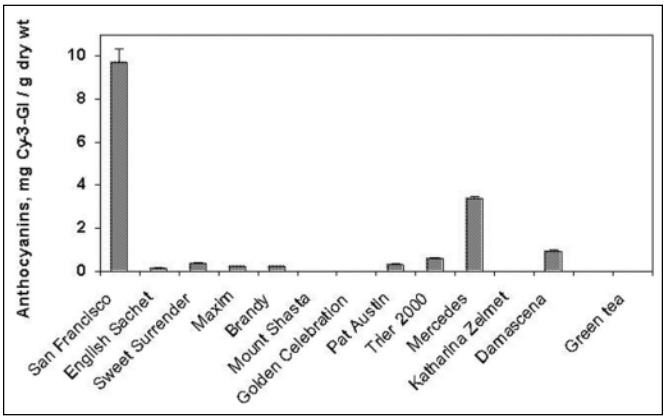

Figure 7—Total anthocyanin contents in the teas prepared from dried petals of various rose cultivars

However, the teas assigned for sensory evaluation were not acidified because it was required to perceive the original taste.

In contrast to the other cultivars, the dry petals of *Rosa damascena* retained a distinct rose scent but, surprisingly, the aroma of brewed *R. damascena* tea was relatively slight as compared with those prepared from cvs Katharina Zeimet and Golden Celebration. It could be that highly volatile essential oil compounds of *R. damascena* were partly lost during brewing. The aromas of the rose teas included both flowery and atypical (for example, fruity, grassy) components, but in general they were positively evaluated by the panelists.

The taste of Katharina Zeimet tea was distinguished by an apparent bitterness that was disliked by the panelists; it was less noticeable in hot teas but became more obtrusive when the samples cooled down. The tea of *R. damascena* was found to be significantly sweeter than those of the other rose cultivars tested (Figure 8). No direct relationship was found between the sensory characteristics of rose teas (for example, bitterness) and their antioxidant activities or phenol contents. In spite of its high contents of phenolic compounds in general and of gallic acid in particular, the tea from San Francisco roses showed the mildest sensory characteristics, with a somewhat bland taste and a faint aroma.

Discussion

The mean values of antioxidant activity of rose teas found in the present study were comparable with the findings of VanderYagt and others (2002), who used the same evaluation method. However, the most active rose cultivars tested (for example, San Francisco, Katharina Zeimet) exhibited the tea activity approximately twice that of 804 μM TE/g reported for rose tea in the aforementioned publication. The activity of rose petal teas of these cultivars even exceeded the value reported by VanderYagt and others (2002) for the most active item in their study: the mate tea from *Ilex paraguariensis*. This illustrates the possibility of enhancing the nutritional quality of foods of plant origin by choosing an appropriate genetic material.

The significant correlation between the radical-scavenging potential, on the 1 hand, and the total phenols and gallic acid contents, on the other hand, may indicate that these compounds make an important contribution to the total antioxidant activity in rose petals, in agreement with the results of Cho and others (2003) and Ng and others (2004). On the other hand, in the cultivars with relatively low level of gallic acid and anthocyanins (for example, hybrid tea roses Sweet Surrender and Mount Shasta, English-type roses Pat Austin and Golden Celebration) the antioxidant activity was apparently contributed by other phenolic compounds. In particular, it might be related to flavonols kaempferol and quercetin present in rose petals mainly in glycoside-bound form. Composition and amount of these compounds vary in different rose cultivars (Biolley and others 1994).

The radical-scavenging activity of rose teas was comparable with or even exceeded that of the green tea, which is known to be 1 of the healthiest beverages. The results obtained for the green tea in the present study (antioxidant activity, total phenols content) were close to those reported in the literature (Prior and Cao 1999). The low content of free gallic acid in the green tea found in the present study was in agreement with the findings of Shahrzad and others (2001) that in the green tea, in contrast to the black one, gallic acid is present predominantly in a conjugated form. The presence of caffeine in green and black teas causes a transient increase in blood pressure after their consumption (Hodgson and others 1999). Because rose petals do not contain caffeine or similar alkaloids (Ashihara and Suzuki 2004), the rose tea may serve as a safe, caffeine-free, antioxidant beverage, especially for individuals to whom blood pressure fluctuations are undesirable.

We suppose that gallic acid and other phenolic compounds play a significant role in the antioxidant activity of rose petal teas. Documented biological activities of gallic acid include antioxidant, antimutagenic, anticarcinogenic, antimicrobial, anti-inflammatory, and analgesic effects (Krogh and others 2000; Kawada and others 2001; Shahrzad and others 2001; Friedman and others 2003). It is notable that this list has much in common with the list of health effects of rose extracts presented in the introduction to the present article. Gallic acid is rapidly absorbed by humans after oral administration, for example, through consumption of black tea (Shahrzad and others 2001).

In the present study, the teas prepared from different rose cultivars varied significantly in their sensory characteristics. Most probably, the sensory characteristics of the teas were determined by the presence of specific taste- and aroma-forming constituents rather than by the total contents of phenolic compounds. At the same time, none of the teas studied possessed tastes and/or aromas that made them unacceptable for human consumption. The tea of the essential oil–containing species *Rosa damascena* was distinguished by a perceptible sweet taste, which might be due to the transport of sucrose into the petals of the opening flower that is associated with essential oil synthesis (Pogorel'skaya and others 1980). Rose cultivars with a high antioxidant activity but a bland taste, such as cv. San Francisco, could be used as additives to tea mixes of other herb species to improve their antioxidant functionality without changing the taste. On the other hand, a cultivar that exhibits a certain degree of bitterness, such as cv. Katharina Zeimet, may also be acceptable in combination with other herbal tea components.

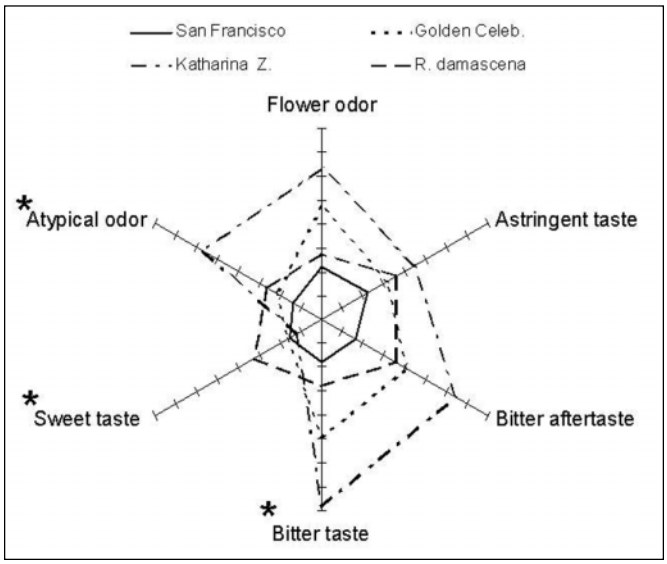

Figure 8—The results of sensory evaluation of teas prepared from dried petals of 4 rose cultivars: San Francisco, Golden Celebration, Katharina Zeimet, and *Rosa damascena*. In parameters marked with an asterisk the difference between cultivars was statistically significant at the 5% level.

Conclusions

Rose-petal tea may serve as a caffeine-free beverage with high antioxidant capacity. The radical-scavenging activity in rose tea is mostly due to the high content of phenolic compounds, in particular free gallic acid. Among the rose teas tested, those prepared

from cvs San Francisco (of the hybrid tea roses group), Katharina Zeimet (polyantha group), Mercedes (floribunda group), and from the essential oil rose *R. damascena* exhibited the highest antioxidant activities (higher than that in the green tea). Rose-petal teas have acceptable sensory characteristics and may be consumed either separately or in combination with other herbal materials.

Acknowledgments

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