Critical Review

Flavanones in oranges, tangerines (mandarins), tangors, and tangelos: a compilation and review of the data from the analytical literature

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Abstract

Flavanones constitute the majority of flavonoids in citrus fruits such as sweet (Citrus sinensis) and sour oranges (C. aurantium) and their near relatives—tangerines/mandarins (C. reticulata), tangors and tangelos. The relevant chemical analytic literature was searched, abstracted, documented, standardized, examined for quality, enumerated, and summarized in a database for these citrus flavanones: hesperidin, naringin, narirutin, eriocitrin, neohesperidin, didymin, neoeriocitrin, and poncirin. Sour oranges had a distinct flavanone profile dominated by naringin and neohesperidin, and were highest in total flavanones (summed means) (48 mg/100 g aglycones). Total flavanones (summed means) in sweet oranges, tangerines, and tangors were similar (~20 mg/100 g), and hesperidin and narirutin dominated the flavanone profiles for these three fruits. Total flavanones (summed means) in tangelos (30 mg/100) were midway between sour and sweet oranges and the tangelo flavanone profile exhibited characteristics of both species. The database provides information on several varieties of citrus and eight flavanone compounds.

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1. Introduction

Currently, there is much biomedical interest in citrus fruits because consumption of them appears to be associated with lower risk of colorectal (Levi et al., 1999), esophageal (Chen et al., 2002; Levi et al., 2000), gastric (Palli et al., 2001), and stomach (McCullough et al., 2001) cancers, and stroke (Feldman 2001; Joshipura et al., 1999). Citrus fruits also appear to be associated with improved blood lipid profiles (Kurowska et al., 2000), and improved survival in the elderly (Fortes et al., 2000). The components responsible for these beneficial effects are unknown, but the citrus flavonoids are one group of compounds that may be involved (Areias et al., 2001; Bae et al., 1999; Bear and Teel 2000a, b; Boraida et al., 1999; de Gregorio Alapont et al., 2000; Leon et al., 2001; Kato et al., 2000; Kim et al., 2000; Kohno et al., 2001; Lee et al., 2001; Manthey et al., 2001; Miyagi et al., 2000; Miyake et al., 2000; Wilcox et al., 2001; Zhang et al., 2000).

The genus Citrus encompasses several orange types—sweet and sour oranges, tangerines (mandarins), tangors, and tangelos. Each species or hybrid cross has one or more varieties (Fig. 1). Sweet oranges are all members of the species, Citrus sinensis, which has four subclasses: common, navel, Valencia, and blood. The general genus and species (i.e. taxon) for tangerines is Citrus reticulate. Other tangerine or mandarin taxons are C. clementina, C. deliciosa, C. nobilis, and C. unshiu. Tangors, such as the popular Temple Orange, are orange-tangerine hybrids (C. reticulate × C. sinensis). Tangelos, such as the Honeybell or Mineola, are tangerine-grapefruit or pummelo hybrids (C. reticulate × C. paradisi or C. reticulate × C. grandis).
In addition, several hybrids have a tangerine for one parent and a tangerine hybrid, usually a tangor, as the other parent. The sour orange is usually eaten in marmalades rather than raw. Its general taxon is *Citrus aurantium* and related taxa are *C. bergamia* or *C. myrtifolia.*

Fig. 2 shows the citrus flavanone glycosides and aglycones. All orange-type citrus fruits contain the flavanone aglycones hesperetin and naringenin, but they rarely occur as free aglycones in the fruit itself. The dominant flavanone glycosides in sweet oranges (*C. sinensis*) are hesperidin and narirutin, whereas in sour oranges (*C. aurantium*) the two predominant flavanone glycosides are neohesperidin and naringin. The major difference between the flavanone glycosides of sweet and sour oranges is in their sugar moieties, which influence taste. The sugar rutinose (6-O-α-L-rhamnosyl-β-D-glucose) causes the flavanones hesperidin and narirutin to have a neutral taste and is relatively high in sweet oranges, tangerines, and tangors. The sugar neohesperidose (2-O-β-L-rhamnosyl-β-D-glucose) is high in tangelos and sour oranges and imparts a tangy or bitter taste to the glycosides neohesperidin and naringin.

The goals of this study were to compile analytic data of acceptable quality from the scientific literature and present values on the flavanones in orange type citrus fruits for a provisional flavonoid food composition table. A similar treatment of grapefruit, lemons and limes is presented in this journal issue (Peterson et al., 2006).

### 2. Materials and methods

The relevant food composition analytical literature was searched using methods previously described for articles dated from 1968 to 1998 and subsequently abstracted (Peterson and Dwyer, 2000). An additional two articles from 2000 were provided through our collaboration with the United States Department of Agriculture Nutrient Data Laboratory. In all, we obtained 125 articles just on citrus. There were very few citations for flavonoid classes and compounds other than flavanones. There was some data on sweet oranges for flavonols (Berhow et al., 1998; Careri et al., 2000; Drawert et al., 1980a, b; Hertog et al., 1993; Justesen et al., 1998; Nogata et al., 1994), flavones (hydroxyflavones—Berhow et al., 1998; Careri et al., 2000; Hertog et al., 1993; Justesen et al., 1998; Nogata et al., 1994; methoxyflavones—Nogata et al., 1994; Pupin et al., 1998b; Rouseff and Ting 1979; Sendra et al., 1988; Veldhuis et al., 1970) and anthocyanins (Rapisarda et al., 1994), and none for flavan-3-ols or isoflavonoids. Based on our preliminary data collection, total flavonoids for sweet oranges was 19.6 mg aglycones/100 g edible fruit or juice and flavanones were 95% of those measured. For tangerines, total flavonoids were 26.4 mg aglycones/100 g edible fruit or juice and flavanones were also 96% of the total flavonoids measured. Anthocyanins are found in blood oranges and the only quantitative study obtained found they averaged 7 mg aglycones/100 g edible fruit or juice (Rapisarda et al., 1994). Although citrus flavonoid data on flavanones, flavones, flavonols and anthocyanins were compiled, this article will focus on the flavanones as they are the major class of flavonoids present in citrus. Articles on the other compounds are planned for the future.

So that the literature would be thoroughly searched, flavanone compounds in citrus were listed individually. Twenty-three articles, which reported values on these compounds, were used for this study. Studies on 12 flavanone glycosides were found. Eight compounds were composed of only four aglycones with glycosides formed by either the sugar rutinose or neohesperidose. Relatively
Four other minor flavanones were not considered further (naringin—1112, neoeriocitrin—1201, neohesperidin—1208, and poncirin—1124).

abundant data were available for these eight flavanone glycosides (didymin, eriocitrin, hesperidin, narirutin, naringin, neoeriocitrin, neohesperidin, and poncirin) (Fig. 2). Four other minor flavanones were not considered further (naringin-4'-glucoside, naringin-6"-malonate (closed form), narirutin-6"-malonate (open form), narirutin-4'-glucoside) because only one study (Berhow et al., 1998) was available on these compounds.

The quality of the flavanone analytical data was assessed using the criteria for evaluating food composition data that were originally developed and used in compiling United States Department of Agriculture (USDA) food composition tables (Holden et al., 2002; Mangels et al., 1993). The basic ideas of this system were used to make the decisions about the inclusion or exclusion of the data sources in consultation with collaborators familiar with the chemistry and food science of citrus. The five quality assessment criteria (number of samples, sample handling, sampling plan, analytical method, and analytical quality control) were utilized and applied to each citrus datapoint in the analytical literature that could be identified for the flavonoids in citrus.

With respect to the evaluation of the number of samples, a “datapoint” was taken to be each analytical value that was reported, usually a mean of duplicate or triplicate analytical values, but occasionally a single analytical value. Each datapoint was treated as an N of 1 and no weighting factor was applied to adjust for the number of samples included. Datapoints described as “not detected” were assigned a value of zero. Based on statistical analyses (Mangels et al., 1993), “trace” values were assigned to be 71% of the limit of quantification for the method used.

The data were placed in a standardized format with respect to taxons (genus, species). All varieties were verified using Germplasm Resources Information Network (GRIN),2 Evaluer Gérer, Informatiser, Diffuser (EGID),3 and other taxonomic sources (Purdue,4 USDA,5 and Australia6). In addition, lists of commercial cultivars and varieties were obtained using two sources, Fresh Citrus Fruits (Wardowski et al., 1986) and Florida Citrus Varieties (Tucker et al., 1998). Tangerine hybrids where one parent was a tangerine and the other parent was a tangerine hybrid (tangor, tangelo, etc.) were considered tangerines.

Information was insufficient to classify certain varieties.
(including sambokan, shunko-kan, tengu, ujuikitsu and a Korean tangerine) into classes or subclasses of orange type citrus; therefore, data concerning them were not included.

For sample handling, most samples were either commercial and refrigerated or frozen until analysis or fresh fruit were processed in the laboratory prior to analysis. With the exception of one article that used gas chromatography, the analytic methodology used in generating these data was all high-performance liquid chromatography (HPLC). Values determined by the Davis method, a spectrophotometric method that measures total flavanones, were not used. Data for each of the eight flavanone glycosides in each orange type group were reviewed and descriptive statistics were calculated. Analytical quality control was also reviewed. Older articles tended to be less explicit in the mention of these details in part perhaps because at the time such details were understood as standard operating procedure. Newer articles tended to be more detailed.

Data on juice and fruit were separated from values provided for the peel or other citrus products. All citrus flavonoid data were converted from their original units to mg/100 g by using Brix data to obtain the density or by employing the USDA standard density of 1.045 for freshly squeezed and 1.052 for commercially processed orange juice. All glycosides were converted to mg/100 g aglycones.

Data were aggregated taxonomically (e.g., by species and hybrid) into five orange types (sour orange, sweet orange, tangerine, tangelo, tangor) for each of the eight flavanone glycosides studied. Descriptive statistics were calculated as aglycones, means of all flavanone aglycones were summed for each orange type, and box plots on each type were produced (Table 1). Outlying values were examined and all were judged to be within the normal limits of environmental variability and included in the analysis. After examination of the degree to which sweet oranges, tangerines and tangors had similar flavanone profiles and levels, the datapoints for these citrus fruits were aggregated; the mean of each flavanone and the total flavanones are given in Table 2.

Although not considered here, included in our database are extraction process, fruit portion, geographic location, date of harvest, and additional method data.

3.Results

Table 1 provides total flavanone content of the individual flavanone compounds for oranges and near relatives (tangerines, tangors, and tangelos). At least two articles for each flavanone compound in sweet oranges were found (except poncirin that had two tangerine articles). Hesperidin had 18 articles for sweet oranges. There were no significant differences between the four subclasses of sweet oranges for hesperidin and narirutin. Comparison of juice (hand and commercial) and fruit (pulp and vesicle) revealed no statistically significant difference among any of these for narirutin. There was a slightly significant difference for hesperidin but only between juice squeezed by hand and the whole fruit (both pulp and vesicle). There was no statistical difference between commercial juices and juices squeezed by hand and between commercial juices and whole fruit (both pulp and vesicle) for hesperidin.

Oranges and their near relatives differed in two characteristics of possible health significance; their individual flavanone profiles and the amount of total flavanones provided. In sweet oranges, tangerines and tangors, the flavanone glycoside pattern consisted predominantly of hesperidin and narirutin. In sour oranges, naringin, neoeriocitrin, and neohesperidin predominated. Tangelos contained some of all of these glycosides but neohesperidin predominated.

Table 2 presents a comparison of the flavanone content of these orange type citrus fruits. Sweet oranges were the lowest in total flavanones (summed means) at 17 mg total flavanone aglycones/100 g edible fruit or juice whereas tangerines and tangors averaged slightly, but not significantly, higher amounts (23 and 25 mg aglycones/100 g edible fruit or juice respectively). All of these varieties provided similar types of flavanones. Tangerines averaged the most hesperidin at 19 mg (+11.6 SD (standard deviation)) aglycone/g edible fruit or juice while sweet oranges and tangors each averaged 15 mg (+7 or 8 SD respectively) aglycone/100 g edible fruit or juice hesperidin, a difference that was not statistically significant. Tangors were significantly higher in narirutin (7 mg aglycone/100 g edible fruit or juice, ±2.7 SD) than oranges (2 mg aglycone/100 g edible fruit or juice, ±1.3 SD, p<0.0001) or tangelos (3 mg aglycone/100 g edible fruit or juice, ±2.8 SD, p=0.0013).

Sour oranges were distinct from sweet oranges, tangerines, and tangors in a number of respects. They were high in naringin (19 mg aglycone/100 g edible fruit or juice, ±8.7 SD), neoeriocitrin (14 mg aglycone/100 g edible fruit or juice, ±4.9 SD), and neohesperidin (11 mg aglycone/100 g edible fruit or juice, ±5.4 SD) but devoid of hesperidin. They also had the highest overall mean total flavanone content (summed means) 48 mg aglycone/100 g edible fruit or juice. Tangors were closer to sour oranges in the amounts of flavanones they contained, ranking second in total flavanones (summed means) (30 mg aglycones/100 g edible fruit or juice) of all the oranges and their near relatives. In their flavanone profiles, tangelos were highest in neohesperidin (14 mg aglycone/100 g edible fruit or juice, ±23.8 SD), followed by naringin (6 mg aglycone/100 g edible fruit and juice, ±11.7 SD), hesperidin (4 mg aglycone/100 g edible fruit and juice, ±2.9 SD) and narirutin (2 mg aglycone/100 g edible fruit or juice, ±1.9 SD) and were significantly different from sweet and/or sour oranges for these four compounds.

4. Discussion

This database provides information on more varieties of citrus and more flavanone compounds than have
previously been available, and may facilitate research on the associations between intakes and health.

Sweet oranges, tangerines (mandarins), and tangors (such as Temple oranges) are similar in both the amounts of flavanones they contain and in their distinctive or "signature" flavanones—hesperidin and narirutin, with virtually no neohesperidin or naringin. Because their flavanone content was similar they were aggregated and presented together in this flavanone food composition table.

In contrast to the sweet orange (*Citrus sinensis*) and its relatives, the sour orange (*C. aurantium*) was qualitatively and quantitatively separate and distinct with respect to both its total flavanone content and flavanone profile. Therefore, it was listed separately in the food table.

The flavanone content of tangelos was different from that of sour and sweet oranges, tangerines, and tangors; it provided as much neohesperidin but only a third as much naringin as sour oranges. Tangelos contained the same amount of narirutin but only a fourth of the hesperidin.

Table 1
Flavanones in sweet oranges, tangerines, tangors, tangelos, and sour oranges

<table>
<thead>
<tr>
<th>Citrus type</th>
<th>Glycosides</th>
<th>Mean</th>
<th>SD</th>
<th>Median</th>
<th>Range</th>
<th>N</th>
<th>Refs</th>
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<td>Sweet orange</td>
<td>Didymin</td>
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<td>0.47</td>
<td>0.45</td>
<td>0.00–1.24</td>
<td>25</td>
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<td>Eriocitrin</td>
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<td>0.07</td>
<td>0.00–0.05</td>
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<td>Hesperidin</td>
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<td>8.21</td>
<td>14.19</td>
<td>2.56–39.26</td>
<td>128</td>
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<td>49</td>
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<td>Narirutin</td>
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<td>114</td>
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<td>Poncirin</td>
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<td>22</td>
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</tr>
<tr>
<td></td>
<td>Poncirin</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>2</td>
<td>1</td>
</tr>
</tbody>
</table>

Lines indicate segregated species. All amounts are in mg aglycone/100 g juice or edible fruit (without rind, pith and seeds).

found in sweet oranges. The same amount of the aglycones hesperetin and naringenin was present in their parent, the tangerine. However, like its grapefruit or pummelo parent, in the tangelo, more glycosylation of hesperetin and naringenin with neohesperidose was evident (and consequently more neohesperidin and naringin was present), whereas there was less glycosylation with rutinose (and thus less hesperidin and narirutin). More data are needed to determine if tangelos are a distinct and separate entity with respect to their flavanone content.

Table 2 describes mean flavanone values and number of samples per datapoint that were included in the estimates. Since the purpose of our study was to describe the different patterns of flavonoid glycosides in various citrus fruits we reported glycosides; in contrast, the USDA food flavonoid database (USDA, 2003) reported only aglycones. There were also other differences between the two databases. We included HPLC data from 1978–1990 and some German articles that were omitted from the USDA database. Nevertheless, in spite of the larger number of values in the Tufts database, the mean flavonone values reported are similar to the USDA values and differed by no more than 1–3 mg for most of the fruits (data not shown). The exception was tangelos for which only one article was included in the USDA database (versus two articles in our study) and the difference was large—89 mg.

In conclusion, data on sweet oranges, tangerines (mandarins) and tangors, which are similar in their flavanone profiles and the quantity of flavanones they contain, were aggregated and presented together in this provisional table of food flavanone composition. Sour oranges and tangelos, which are distinct in both their flavanone content and the flavanone compounds they contain, constitute distinctive categories and were presented separately.

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References


