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## Sources of variability in the flavonoid content of foods

D.B. Haytowitz<sup>\*</sup>, S. Bhagwat, and J.M. Holden

*USDA-ARS-Nutrient Data Lab, Beltsville, Maryland, 20705 USA*

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### Abstract

In 2012, USDA prepared Release 3.1 of the “USDA Database for the Flavonoid Content of Selected Foods”, which contained data on 508 food items collected from 308 published sources. Flavonoids are secondary metabolites produced by plants in response to various environmental stresses such as climate and ultraviolet radiation. Other sources of variability include cultivar, growing location, agricultural practices, processing techniques and preparation methods, as well as analytical variability. The objective of this manuscript is to examine and report on variability in the flavonoid content of foods. While the required information needed to assess variability is not available for all foods, data for a number of foods was analyzed using analysis of variance for cultivar, location and other factors. For orange juice, data for 247 samples representing 109 mean values, i.e., different cultivars and location, were analyzed. The overall range for hesperetin was 1-39mg/100g; samples from the United States 5-30mg/100g; samples from Brazil 5-25mg/100g; and samples from Corsica (France) 12-26mg/100g. For strawberries, data for 148 samples representing 98 mean values were analyzed. The overall range for pelargonidin was 8-58 mg/100g. The range of quercetin values in raw yellow onions is 0-91mg/100g, and represents 96 mean values for 402 samples from the U.S., Japan, Spain, and other countries. The flavonoid content of foods is extremely variable and is influenced by both location and cultivar, which account for 25 to 33 percent of the variability as well as by numerous other factors, which were not examined in this analysis.

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### 1. Introduction

Flavonoids are secondary plant metabolites and are found mainly in fruits, vegetables, and some grains. The quantity of a specific flavonoid can vary considerably depending on a number of factors, such as diseases, insect/pest attack, climate stress, ultraviolet radiation, and others [1, 2]. Other sources of variability can include cultivar, growing location, agricultural practices, harvesting and storage conditions, and processing and preparation methods [3, 4, 5, 6].

Several properties exhibited by flavonoids include inhibition of lipid peroxidation, anticarcinogenic activity, and antioxidant capacity [7]. The potential to reduce the risks of chronic diseases due to these properties has motivated the scientific community to conduct epidemiological studies to observe the intakes of flavonoids and possible associations with risk reduction for various diseases. To address the needs of the research community, the Nutrient Data Laboratory/USDA (NDL) has developed three separate databases: 1) Flavonoid content of selected foods, Release 3.1, 2012, with values for 26

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<sup>\*</sup> Corresponding author. Tel.: 301-504-0714; fax: 301-504-0713.  
E-mail address: [david.haytowitz@ars.usda.gov](mailto:david.haytowitz@ars.usda.gov)

individual flavonoids (Table 1) for over 500 foods; 2) Isoflavone content of selected foods, Release 2, 2008, with values for 549 foods; and 3) Proanthocyanidin content of selected foods, 2004, with values for over 200 foods. Table 1 shows the list of subclasses of flavonoids as well as the individual aglycone compounds within each subclass. The subclasses are determined by the chemical structure or characteristics of that group of compounds. All of these compounds do not occur together in any one type of food or group of similar foods. Rather, a specific flavonoid subclass or one or more individual flavonoids within that class will predominate in a specific food or type of food.

Table 1. Flavonoid subclasses [8]

Anthocyanidins	Cyanidin, delphinidin, malvidin, pelargonidin, peonidin, petunidin
Flavonols	Quercetin, kaempferol, myricetin, isorhamnetin
Flavones	Apigenin, luteolin
Flavanones	Hesperetin, naringenin, eriodictyol
Flavan-3-ols	Catechin, epicatechin, epicatechin3-gallate, epigallocatechin, epigallocatechin 3-gallate, galocatechin, theaflavin, theaflavin 3-3'-digallate, theaflavin 3'-gallate, theaflavin 3-gallate, thearubigins
Isoflavones	Daidzein, genistein, glycitein

The national and international scientific community recently used these NDL databases to study associations of flavonoid intakes with various disorders such as stroke [9] cardiovascular disease [10] diabetes [11], and colorectal cancer [12]. The objective of this paper is to discuss the mean values and variability for selected flavonoid compounds in three foods from the flavonoid database.

## 2. Methods

As part of the process of developing special interest food composition databases, NDL collects articles from the scientific literature. For Release 3.1 of the flavonoid database, data were obtained from 308 articles [8]. These articles contain varying amounts of information about the food item and related data. The documentation may include the detailed food description and other information about many, but not all of the factors influencing flavonoid contents listed above. However cultivar and sampling location, though not necessarily growing location, were frequently provided.

The flavonoid compounds which are predominant in three selected foods were studied for the effects of cultivar and growing locations. They were hesperetin (a flavanone) in orange juice, pelargonidin (an anthocyanidin) in strawberries, and quercetin (a flavonol) in onions. The flavonoid content for all foods examined was analyzed by HPLC. The variability of quercetin in onions was examined only in different cultivars. The orange juice values for hesperetin were available from Brazil, France, Italy, Spain and the United States and for navel, pera and valencia cultivars. Pelargonidin values were available from the United States, Brazil and Spain and for Allstar, Honeoye, and Camarosa cultivars. Values for quercetin were available for yellow, red, white, and sweet onion cultivars.

The data were analyzed using SAS 9.2 (SAS Institute, Inc. Cary, NC). Descriptive statistics were used to characterize the data overall and across factors. To identify statistical differences in flavonoids content between factors, ANOVA methods were employed.

### 3. Results and Discussion

#### 3.1. Hesperetin in Orange Juice

One of the major flavonoids in citrus fruits is hesperetin and one of the more commonly consumed citrus products is orange juice. The database contains 13 data sources (published articles) with 111 data points. Sufficient data were available for five cultivars: Navel, Pera, Valencia, Hamlin, and Natal. These were used to analyze variability in hesperetin content. If the cultivar was not identified or there was only a single value for a specific cultivar, the values were collapsed into a separate category, "Other".

Table 2. Hesperetin in orange juice by cultivar (mg/100 g, edible portion)

Cultivar	Mean	Median	n	SD	Range
Navel <sup>a</sup>	18	18	20	7.4	5.2 - 29
Pera <sup>a</sup>	12	11	10	3.7	6.3 - 19
Valencia <sup>b</sup>	6.7	5.4	28	3.0	4.2 - 15
Hamlin <sup>a</sup>	15	15	5	7.1	6 - 25
Natal <sup>a</sup>	9	9	5	3.6	5 - 14
Other <sup>a</sup>	16	14	43	9.8	1.3 - 39

F=7.23, p<.0001, R<sup>2</sup> = 0.256

Pairs with the same letter are not statistically different (p<0.05)

There were no significant differences among the mean hesperetin values for orange juice cultivars except for values for the Valencia cultivar. These were significantly lower than the other cultivars (p<.05) (Table 2). Values among all cultivars ranged from 4 to 39 mg/100 g. Median values were very close to the mean values in these cultivars. Differences among cultivars accounted for 26% of the observed variability, indicating that a number of other factors are also present. Most of the orange juice samples were identified as hand-squeezed or fresh-squeezed. Others were obtained from retail outlets and were either often identified as "not from concentrate" or no information was provided. These were included in the "Other" category and may account for the larger variability observed.

Table 3. Hesperetin in orange juice by source location (mg/100 g, edible portion)

Location	Mean	Median	n	SD	Range
Brazil <sup>a</sup>	12	12	28	4.5	4.9 - 25
France <sup>b</sup>	23	21	10	8.3	12 - 36
Italy <sup>a,b</sup>	17	6	8	17.4	2.5 - 39
USA <sup>a,b,d</sup>	16	15	34	6.7	5.2 - 30
Spain <sup>c</sup>	6.2	5.1	23	3.3	4.2 - 18
Other <sup>a,c</sup>	10	9	8	5.3	1.3 - 18

F=10.13, p<.0001, R<sup>2</sup> = 0.325

Pairs with the same letter are not statistically different (p<0.05)

The hesperetin values in orange juice from France, Italy and the United States were not significantly different (Table 3), although the range of values from Italy was wide (2.5 – 39 mg/100g), leading to a larger standard deviation (17.4). The values from Brazil were in the middle of the range, while values from Spain were significantly lower than those from other countries. Median values were close to mean values, except for Italy. Location accounted for 32% of the observed variability. The interaction between cultivar and location was not significant. Most samples were collected in a single location, and are

therefore not nationally representative samples. Consequently, the effects of cultivar and processing are not accounted for in these values.

### 3.2. Pelargonidin in Strawberries

Strawberries contain one of the highest amount of pelargonidin—an anthocyanidin found in any food (25.69 mg/100g). The database contains 11 data sources (published articles), representing 101 individual data points and five locations. Two of the five locations reported data for only one or two samples, and were not included in this particular analysis (Table 4) reducing the total to 98 individual data points across the three locations.

Table 4. Pelargonidin in strawberries by cultivar (mg/100 g, edible portion)

Cultivar	Mean	Median	n	SD	Range
Allstar <sup>a,c</sup>	25	25	14	3.3	19 - 31
Honeoye <sup>b</sup>	48	49	12	6.0	39 - 57
Camarosa <sup>a,b,c</sup>	36	35	6	15.9	19 - 54
Other <sup>a,c</sup>	26	23	66	12.9	8 - 58

F=12.82, p<.0001, R<sup>2</sup> = 0.290

Pairs with the same letter are not statistically different (P<0.05)

Pelargonidin values among the three cultivars from the same countries listed above ranged from 19 to 57 mg/100 g (Table 4). The Honeoye cultivar was significantly different from Allstar but was similar to the Camarosa cultivar (p<.05). Median values were very similar to the mean values, indicating a normal distribution. Cultivar accounted for 29% of the variability.

Table 5. Pelargonidin in strawberries by source location (mg/100 g, edible portion)

Location	Mean	Median	n	SD	Range
USA <sup>a</sup>	34	32	64	12.1	8.0 - 58
Spain <sup>b,c</sup>	19	14	27	11.4	8.5 - 54
Brazil <sup>c</sup>	25	23	7	11.0	12 - 43

F=16.41, p<.0001, R<sup>2</sup>=0.259

Pairs with the same letter are not statistically different (p<0.05)

Pelargonidin values in strawberries from Brazil and Spain were not significantly different from each other, while those from the United States were higher than the other two locations (Table 5). Although the ranges and standard deviation for each source location were similar, the mean values were different, indicating that the distribution within each range was different. Median values were close to the mean values for all the countries. Location accounted for 26% of the variability. The interaction between cultivar and location was not significant.

### 3.3. Quercetin in Onions

Onions are one of the major dietary sources of quercetin. The database contains 27 data sources (published articles), representing 201 individual data points.

There is a great deal of variability in quercetin content among the different colored onions: yellow, red, white and sweet (Table 6). All varieties showed large ranges and high standard deviations, sometimes exceeding the mean value as seen in the white variety, where the mean value was 13 mg/100 g and the standard deviation is 18.2. For the white cultivars, the mean and medians are very different, further illustrating the large variability. The minimum values for all onion colors were zero or close to zero. On the other hand, the maximum values varied among the different types. As a result of this high variability,

yellow, white, and sweet types were not significantly different, while red onions were significantly different from the other types. The type of onion accounted for 33% of the observed variability.

Table 6. Quercetin in various onion types (mg/100 g, edible portion).

Cultivar	Mean	Median	n	SD	Range
Yellow <sup>a</sup>	20	14	96	17.0	0 - 91
Red <sup>b</sup>	62	44	50	47.2	0 - 191
White <sup>a</sup>	13	0.3	35	18.2	0 - 63
Sweet <sup>a</sup>	14	8	20	12.7	1 - 46

F=32.58, p<0.0001 R<sup>2</sup>=0.33

Pairs with the same letter are not statistically different (p<0.005)

#### 4. Conclusion

These observations illustrate wide variations in selected flavonoid compounds within and among cultivars and locations. In general, cultivar and location accounted for between 25 and 33% of the observed variability for the selected foods and selected flavonoids examined, suggesting that other factors such as the effects of climate stress, insect infestation, and agricultural practices may account for the remaining variability. Analyses of flavonoid values for foods and their variability by factors such as cultivar and location could be used to indicate the optimum cultivars or conditions for a particular food application. Garcia-Viguera [13] observed that the Camarosa variety of strawberries had the highest total anthocyanidins and would be better for jam preparations. Wang and Murphy [14] studied the effects of variety, crop year and location on the isoflavone contents of the soybean variety Vinton 81 and observed that the isoflavone contents were influenced by all these factors, but the crop year had a major impact, perhaps due to climate conditions. More research is needed to control and assess the effect of these factors on the variability of specific compounds.

Systematic food descriptions are frequently absent or inadequate in published studies. Scientific names (including cultivars), growing locations, year, and part of the food analyzed, storage length and condition, and information on processing (raw, cooked, canned etc.) are necessary elements for proper classification and analyses of food composition data.

The individual data used to calculate the mean values and other statistics presented in this manuscript are available on NDL's web site ([www.ars.usda.gov/nutrientdata](http://www.ars.usda.gov/nutrientdata)) in Release 3.1 of the USDA Database for the Flavonoid Content of Selected Foods [8].

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