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Concentrations of Anthocyanins in Common Foods in the United States and Estimation of Normal Consumption

XIANLI WU,[†] GARY R. BEECHER,[§] JOANNE M. HOLDEN,[#] DAVID B. HAYTOWITZ,[#] SUSAN E. GEBHARDT,[#] AND RONALD L. PRIOR*,[†]

USDA Arkansas Children's Nutrition Center, 1120 Marshall Street, Little Rock, Arkansas 72202, and Food Composition Laboratory and Nutrient Data Laboratory, Beltsville Human Nutrition Center, Agricultural Research Service, U.S. Department of Agriculture, 10300 Baltimore Avenue, Beltsville, Maryland 20705

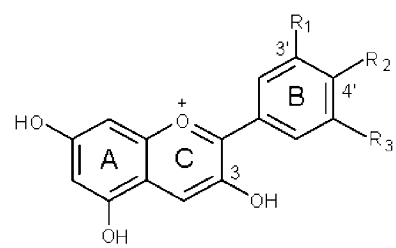
Anthocyanins (ACNs) are water-soluble plant pigments that have important functions in plant physiology as well as possible health effects. Over 100 common foods were screened for ACNs, and 24 of them were found to contain ACNs. Concentrations of total ACNs varied considerably from 0.7 to 1480 mg/100 g of fresh weight in gooseberry ('Careless' variety) and chokeberry, respectively. Not only does the concentration vary, but the specific anthocyanins present in foods are also quite different. Only six common aglycones, delphinidin, cyanidin, petunidin, pelargonidin, peonidin, and malvidin, were found in all of these foods. However, their sugar moieties and acylation patterns varied from food to food. Results from this study will add to the available data for the USDA Nutrient Database of flavonoids. On the basis of the concentration data and updated food intake data from NHANES 2001−2002, the daily intake of ACNs is estimated to be 12.5 mg/day/person in the United States. Of the different aglycones, cyanidin, delphinidin, and malvidin were estimated to contribute 45, 21, and 15%, respectively, of the total ACN intake. Nonacylated contributed 77% compared to 23% from acylated ACNs.

KEYWORDS: Anthocyanin; concentration; daily intake; NHANES

INTRODUCTION

Anthocyanins (ACNs) are water-soluble plant pigments responsible for the blue, purple, and red color of many plant tissues. They occur primarily as glycosides or acylglycosides of their respective aglycone anthocyanidins (*1*). Aglycones are rarely found in fresh plant materials. There are about 17 anthocyanidins found in nature (*1*), whereas only 6 of them, cyanidin (Cy), delphinidin (Dp), petunidin (Pt), peonidin (Pn), pelargonidin (Pg), and malvidin (Mv), are ubiquitously distributed (**Figure 1**). Thus far, over 600 naturally occurring ACNs have been reported (*2*, *3*), and they are known to vary in (1) the number and position of hydroxyl and methoxyl groups on the basic anthocyanidin skeleton; (2) the identity, number, and positions at which sugars are attached; and (3) the extent of sugar acylation and the identity of the acylating agent (*4*).

ACNs have important functions in plant physiology. They are believed to play a major role in both pollination and seed dispersal. Because of their intense color, anthocyanins are also regarded as potential candidates for natural colorants in the food industry. Recently, increased attention has focused on their possible health effects. ACNs have been shown to be strong antioxidants (*5*−*7*) and may exert a wide range of health benefits through antioxidant or other mechanisms (*8*−*16*).



ANTHOCYANIDIN	R_1	R_2	R_3
Pelargonidin (Pg)	H	OH	H
Cyanidin (Cy)	OH	OH	H
Delphinidin (Dp)	OH	OH	OH
Peonidin (Pn)	OMe	OH	H
Petunidin (Pt)	OMe	OH	OH
Malvidin (Mv)	OMe	OH	OMe

Figure 1. Chemical structures of six naturally occurring common anthocyanidins.

From published data, we know that the content of total ACNs in foods varies considerably. Not only does the concentration

* Author to whom correspondence should be addressed (e-mail PriorRonaldL@uams.edu).
[†] USDA Arkansas Children's Nutrition Center.
[§] Food Composition Laboratory, U.S. Department of Agriculture.
[#] Nutrient Data Laboratory, U.S. Department of Agriculture.

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Table 1. Foods Screened for the Presence of Anthocyanins[a]

fruits	vegetables	nuts and dried fruits	spices	other foods
apples[b]	artichoke	almonds	basil leaf	bread
apricot	asparagus	Brazil nuts	chili powder	breakfast cereals
avocado	beans, black[b]	cashews	cinnamon	snack
bananas	beans,navy	hazelnuts	cloves, ground	
blackberry[b]	beans, pinto	macadamias	curry powder	apple sauce
blueberry[b]	beans, red kidney	peanuts	garlic powder	baby foods
cantaloupe	beans, small red[b]	pecans	ginger, ground	chips
cherries[b]	beets	pine nuts	mustard seed	chocolate
chokeberry[b]	broccoli	pistachios[b]	onion powder	native American food
cranberry[b]	cabbages[b]	walnuts	oregano leaf	juice, grapefruit
currant, black[b]	carrots		paprika	juice, lemon
currant, red[b]	cauliflower	dates	parsley	juice, lime
elderberry[b]	celery	figs	pepper	juice, tomato
gooseberry[b]	corn	prunes	poppy seed	juice, V8 vegetable
grapefruit	cucumber	raisins	turmeric	ketchup
grapes[b]	eggplant[b]			milk
honeydew	lettuces[b]			peanut butter
kiwifruit	onions[b]			rice
mango	peas			salsa
nectarines[b]	peppers			tomato sauce
orange, navel	potatoes			
peaches[b]	pumpkin			
pears	red radishes[b]			
pineapples	spinach			
plums[b]	sweet potatoes			
raspberry[b]	tomatoes			
strawberry[b]				
tangerines				
watermelons				

[a] Foods listed as only major categories. [b] These foods were found to contain anthocyanins.

vary, but the specific ACNs present in foods are also quite different. The potential dietary intake of ACNs is among the greatest of the various classes of flavonoids. Intake of ACNs was estimated to be as high as 185−215 mg/day/person in an early paper (*17*). These data have been quoted extensively but have limitations due to the lack of comprehensive data on anthocyanin content and daily intake and information on individual anthocyanins. Accurate estimation of ACN content in foods and the daily intake is critical in food science, nutrition, and other related research fields. Recent research has shown that anthocyanins with different aglycones and sugar moieties may have quite different responses in terms of their bioavailability and potential health effects (*16*, *18*−*21*). Acylated ACNs may behave quite differently from nonacylated ACNs in terms of their stability and bioavailability (*22*−*25*); it is not reasonable to consider all anthocyanins as behaving in the same manner. Thus, knowledge regarding the concentrations and daily intake of specific anthocyanins is essential.

The objective of this study was to quantify anthocyanins in common foods on the basis of their specific composition, which has been reported previously (*26*, *27*). All of the foods considered in this study were obtained directly from U.S. markets on multiple occasions as described previously (*28*) and thus are representative of the foods commonly consumed by the U.S. population. One of the major purposes of this study is to provide information for the USDA database of flavonoids. In addition, an estimation of the daily intake of ACNs in the United States was made on the basis of our quantitative data and results from the updated food intake survey, the National Health and Nutrition Examination Survey (NHANES 2001−2002) (*29*).

MATERIALS AND METHODS

Standards and Solvents. Standards of the 3-*O*-β-glucosides of pelargonidin, cyanidin, peonidin, delphinidin, petunidin, and malvidin (six mixed anthocyanin standard, HPLC grade) were obtained from Polyphenols Laboratories (Sandnes, Norway). Formic acid was purchased from Aldrich (St. Louis, MO). All other solvents were purchased from Fisher (Fair Lawn, NJ).

Sampling Method and Sample Preparation. The sources and methods of collection of food samples in this study were described previously (*28*). Fruits and vegetables were sampled from retail outlets in 12 cities around the United States in two different seasons using statistically validated methods (*30*, *31*). Approximately 3 lb of each item was randomly selected from bins in each retail outlet. These samples were composited to form four regional composites: west (Los Angeles, CA; Vancouver, WA; and Longview, WA); central (Wheaton, IL; Conroe, TX; and Beaumont, TX); south (Mena, AR; Springfield, MO; and Franklin, TN); and northeast (Springfield, NJ; Canonsburg, PA; and Franklin, PA). Sweet cherries and red grapes were collected in only one season from the same locations and composited in the same manner. There were normally eight samples for each food item. The samples were collected and processed in the Food and Analysis Laboratory Control Center, Department of Biochemistry of the Virginia Polytechnic Institute and State University (Blacksburg, VA). Concord grape freeze-dried powder was provided by Future Ceuticals (Santa Rosa, CA). One of the strawberry samples was provided as a freeze-dried powder by the Oregon Strawberry Commission (OSC). Black bean, small red bean, red onion, and eggplant were purchased from a local supermarket. Dry matter was determined using AOAC standardized methods [vacuum oven, 934.06 (37.1.10)]. All fresh foods were freeze-dried and kept at −70 °C until analyzed. The freeze-dried powders were extracted using methanol/water/acetic acid (85:15:0.5, v/v, MeOH/H_2O/HAc) as reported previously (*32*). The solutions from the extracted samples were then diluted with acidic methanol as necessary to obtain concentrations in a detectable range and filtered using a 0.22 μm Teflon syringe filter (Cameo, MN) for ACN analysis.

HPLC-DAD-ESI/MS/MS Analysis and Quantitation of Anthocyanins. Chromatographic analyses were performed on an HP 1100 series HPLC (Hewlett-Packard, Palo Alto, CA) equipped with an autosampler/injector and diode array detector. A Zorbax Stablebond Analytical SB-C_{18} column (4.6 × 250 mm, 5 μm, Agilent Technologies,

Table 2. Concentration of Anthocyanins Grouped by Aglycones in Common Foods in the United States[a,b]

food	moisture[c] (%)	mg/100 g (of fresh wt or form consumed)							total ACN/ serving[d] (mg)
		Dp-ACN	Cy-ACN	Pt-ACN	Pg-ACN	Pn-ACN	Mv-ACN	total ACN	
fruits	–	–	–	–	–	–	–	–	–
1. apple	–	–	–	–	–	–	–	–	–
Fuji ($n = 4$[e])	84.2	–	1.3 ± 0.7	–	–	–	–	1.3 ± 0.7	1.8
Gala ($n = 3$)	85.8	–	2.3 ± 0.8	–	–	–	–	2.3 ± 0.8	3.2
Red Delicious ($n = 4$)	85.5	–	12.1 ± 1.8	–	–	0.2 ± 0.1	–	12.3 ± 1.9	17.0
2. blackberry	–	–	–	–	–	–	–	–	–
blackberry ($n = 4$)	86.9	–	244 ± 68.0	–	0.7 ± 0.1	T[f]	–	245 ± 68.0	353
Marion blackberry ($n = 1$)	86.9	–	297.7	–	1.7	1.1	–	300.5	433
3. blueberry	–	–	–	–	–	–	–	–	–
cultivated ($n = 7$)	85.0	120.7 ± 27.9	28.6 ± 19.8	71.9 ± 14.0	–	34.2 ± 11.9	131.3 ± 16.5	386.6 ± 77.7	529
wild ($n = 1$)	89.0	141.1	66.3	87.6	–	36.9	154.6	486.5	705
4. cherry, sweet ($n = 4$)	80.2	–	113± 19.6	–	1.4 ± 0.2	7.5 ± 1.9	–	122 ± 21.3	177
5. chokeberry ($n = 1$)	71.8	–	1478	–	2.3	–	–	1480	2147[g]
6. cranberry ($n = 3$)	87.1	0.1 ± 0.1	66.1 ± 16.7	T	0.7 ± 0.1	72.2 ± 13.6	0.8 ± 0.9	140 ± 28.5	133
7. currant	–	–	–	–	–	–	–	–	–
black currant ($n = 6$)	77.5	333 ± 78.1	133 ± 38.6	7.3 ± 5.6	1.9 ± 0.5	1.0 ± 0.5	–	476 ± 115	533
red currant ($n = 1$)	78.1	0.1	12.7	–	–	–	–	12.8	14.3
8. elderberry ($n = 1$)	82.5	–	1373	–	1.8	–	–	1375	1993[g]
9. gooseberry[h]	–	–	–	–	–	–	–	–	–
group 1 ($n = 2$)	88.0	–	10.2 ± 0.1	–	–	0.2 ± 0.1	–	10.4 ± 0.1	15.1[g]
group 2 ($n = 1$)	88.0	–	2.1	–	–	0.1	–	2.2	3.2[g]
group 3 ($n = 1$)	88.0	–	0.7	–	–	–	–	0.7	1.0[g]
10. grape	–	–	–	–	–	–	–	–	–
red grape ($n = 5$)	80.4	1.1 ± 0.8	3.9 ± 1.5	1.1 ± 0.9	–	10.1 ± 4.5	10.5 ± 8.4	26.7 ± 10.9	42.7
Concord grape ($n = 1$)	80.4	70.7	23.8	14.9	T	4.8	5.9	120.1	192
11. nectarine ($n = 7$)	86.8	–	6.8 ± 1.5	–	–	–	–	6.8 ± 1.5	9.2
12. peach ($n = 8$)	88.3	–	4.8 ± 1.2	–	–	–	–	4.8 ± 1.2	4.7
13. plum	–	–	–	–	–	–	–	–	–
plum ($n = 8$)	87.4	–	19.0 ± 4.4	–	–	–	–	19.0 ± 4.4	12.5
black plum ($n = 2$)	87.9	–	124.5 ± 21.6	–	T	T	–	124.5 ± 21.6	82.2
14. raspberry	–	–	–	–	–	–	–	–	–
black raspberry ($n = 1$)	85.8	–	669	–	16.7	1.1	–	687	845
red raspberry ($n = 5$)	85.8	–	90.2 ± 19.2	–	1.9 ± 1.0	–	–	92.1 ± 19.7	116
15. strawberry	–	–	–	–	–	–	–	–	–
strawberry ($n = 8$)	91.1	–	1.2 ± 0.4	–	19.8 ± 3.1	–	–	21.2 ± 3.3	35.0
strawberry OSC[i] ($n = 1$)	91.1	–	9.4	1.0	31.4	–	–	41.7	69.2
vegetables	–	–	–	–	–	–	–	–	–
1. black bean ($n = 1$)	–	18.5	–	15.4	–	–	10.6	44.5	23.1
2. eggplant ($n = 1$)	91.8	85.7	–	–	–	–	–	85.7	35.1
3. red cabbage ($n = 4$)	91.0	–	322 ± 40.8	–	–	–	–	322 ± 40.8	113
4. red leaf lettuce ($n = 8$)	95.6	–	2.2 ± 1.5	–	–	–	–	2.2 ± 1.5	1.5
5. red onion ($n = 1$)	87.7	–	46.4	–	–	2.1	–	48.5	38.8
6. red radish ($n = 9$)	95.6	–	T	–	100.1 ± 30.0	–	–	100.1 ± 30.0	116
7. small red bean ($n = 1$)	–	–	1.9	–	4.8	–	–	6.7	6.2
nuts	–	–	–	–	–	–	–	–	–
1. pistachio ($n = 7$)	–	–	7.5 ± 1.5	–	–	–	–	7.5 ± 1.5	2.1

[a] Data expressed milligrams of anthocyanin (anthocyanidin glycosides or acylglycosides) on an "as is" weight basis and presented as mean ± SD for sample number >2. [b] Abbreviations: ACN, anthocyanin; Dp, delphinidin; Cy, cyanidin; Pt, petunidin, Pg, pelargonidin; Pn, peonidin; Mv, malvidin. [c] Expressed as mean of different samples. [d] Serving size from the USDA National Nutrient Database for Standard Reference (www.nal.usda.gov/fnic/foodcomp). [e] Sample number of each food. [f] T, trace. [g] No serving size data available. Estimated as a blueberry serving size, 145 g (1 cup). [h] Group 1, contained the varieties 'Whinham' and 'Lancashine'; group 2, variety was 'Dan's Mistake'; group 3, variety was 'Careless'. [i] OSC, Oregon Strawberry Commission.

Rising Sun, MD) was used for separation. The experimental conditions were described in our former papers (*26*, *27*).

ACN identification in all of the foods has been reported in our previous papers (*26*, *27*). Quantification of anthocyanins followed the procedure reported previously (*32*). The intra- and interassay CVs were 1.5 and 2.5%, respectively. The limit of quantification for ACNs under current experimental conditions was 10 ng. Calculations were made on the basis of the freeze-dried forms, and then the data were converted to a fresh weight (FW) basis for fresh fruits and vegetables. Final concentrations were expressed as milligrams per 100 g of fresh weight (FW) for fresh fruits and vegetables and on an "as is" weight basis for beans and nuts. ACN weights were based upon the total mass [aglycone + glycoside(s)].

Estimation of ACN Intake. The daily intake of ACNs was estimated primarily using dietary intake information from the National Health and Nutrition Examination Survey (NHANES 2001−2002) (*29*). NHANES was conducted by the U.S. Department of Health and Human Service (HHS). It is a multistage, stratified area sample that is representative of the civilian noninstitutionalized population of the United States. Data on average total concentrations of ACNs in grape juices were kindly provided by Dr. JoLynne Wightman from Welch's, in which 23 different brands of commercial grape juices were included.

RESULTS AND DISCUSSION

Anthocyanin Quantitation in Foods. Because of the important roles ACNs play in the food industry and human nutrition, accurate quantification of ACNs is critical. Currently, quantification of anthocyanins is mainly achieved either by spectrophotometry or by HPLC. The pH-differential method is the principal spectrophotometric method, which has been widely used, especially in industry, because it is a rapid and easily performed procedure (*33*). However, this method cannot provide any information regarding the individual aglycones and/or glycoside substitution or acylation pattern of the ACNs.

Table 3. Anthocyanins with Different Sugar Moieties and Acylated or Nonacylated Groups in Common Foods[a,b]

	mg/100 g (of fresh wt or form consumed)					
	acylation		sugar acylation			
food	nonacylated ACN (%)	acylated ACN (%)	ACN mono-glycoside (%)	ACN di-glycoside (%)	ACN tri-glycoside (%)	total ACN
fruits	–	–	–	–	–	–
1. apple	–	–	–	–	–	–
Fuji (*n* = 4[c])	1.3 ± 0.7 (100%)	–	1.3 ± 0.7 (100%)	–	–	1.3 ± 0.7
Gala (*n* = 3)	2.3 ± 0.8 (100%)	–	2.3 ± 0.8 (100%)	–	–	2.3 ± 0.8
Red Delicious (*n* = 4)	12.3 ± 1.9 (100%)	–	12.3 ± 1.9 (100%)	–	–	12.3 ± 1.9
2. blackberry	–	–	–	–	–	–
blackberry (*n* = 4)	231 ± 56.4 (94%)	14.3 ± 13.9 (6%)	220 ± 89.1 (90%)	25.5 ± 28.8 (10%)	–	245 ± 68.0
Marion blackberry (*n* = 1)	292 (97%)	8.8 (3%)	229 (76%)	71.9 (24%)	–	300
3. blueberry	–	–	–	–	–	–
cultivated (*n* = 8)	348 ± 56.6 (96%)	16.4 ± 8.9 (4%)	365 ± 56.5 (100%)	–	–	365 ± 56.5
wild (*n* = 1)	421 (87%)	65.6 (13%)	487 (100%)	–	–	487
4. cherry, sweet (*n* = 4)	122 ± 21.3 (100%)	–	7.6 ± 3.0 (6%)	114 ± 20.9 (94%)	–	122 ± 21.3
5. chokeberry (*n* = 1)	1480 (100%)	–	1480 (100%)	–	–	1480
6. cranberry (*n* = 7)	140.0 ± 28.5 (100%)	–	140.0 ± 28.5 (100%)	–	–	140 ± 28.5
7. currant	–	–	–	–	–	–
black currant (*n* = 6)	471 ± 115.7 (99%)	5.3 ± 6.7 (1%)	113 ± 33.1 (24%)	363 ± 83.8 (76%)	–	476 ± 115.4
red currant (*n* = 1)	12.8 (100%)	–	0.2 (2%)	5.2 (41%)	7.4 (58%)	12.8
8. elderberry (*n* = 1)	1375 (100%)	–	742 (54%)	550 (40%)	82.6 (6%)	1375
9. gooseberry[d]	–	–	–	–	–	–
group 1 (*n* = 2)	8.2 ± 1.0 (79%)	2.2 ± 1.0 (21%)	7.4 ± 0.7 (71%)	3.0 ± 0.7 (29%)	–	10.4 ± 0.1
group 2 (*n* = 1)	2.0 (91%)	0.2 (9%)	1.7 (77%)	0.5 (23%)	–	2.2
group 3 (*n* = 1)	0.7 (100%)	–	0.7 (100%)	–	–	0.7
10. grape	–	–	–	–	–	–
red grape (*n* = 4)	20.8 ± 7.1 (78%)	5.8 ± 5.7 (22%)	26.7 ± 10.9 (100%)	–	–	26.7 ± 10.9
Concord grape (*n* = 1)	59.9 (50%)	60.2 (50%)	102 (85%)	18.5 (15%)	–	120
11. nectarine (*n* = 8)	6.8 ± 1.5 (100%)	–	6.6 ± 1.4 (97%)	0.2 ± 0.1 (3%)	–	6.8 ± 1.5
12. peach (*n* = 8)	4.8 ± 1.2 (100%)	–	4.8 ± 1.2 (100%)	T[5]	–	4.8 ± 1.2
13. plum	–	–	–	–	–	–
plum (*n* = 8)	18.8 ± 4.4 (99%)	0.2 ± 0.1 (1%)	14.6 ± 3.9 (77%)	4.4 ± 1.1 (23%)	–	19.0 ± 4.4
black plum (*n* = 2)	124 ± 21.4 (99%)	0.5 ± 0.2 (1%)	96.7 ± 17.5 (78%)	27.8 ± 4.1 (22%)	–	124.5 ± 21.6
14. raspberry	–	–	–	–	–	–
black raspberry (*n* = 1)	687 (100%)	–	89.4 (13%)	440 (64%)	158 (23%)	687
red raspberry (*n* = 6)	92.1 ± 19.7 (100%)	–	20.1 ± 9.3 (22%)	47.8 ± 9.4 (52%)	24.2 ± 15.8 (26%)	92.1 ± 19.7
15. strawberry	–	–	–	–	–	–
strawberry (*n* = 8)	21.0 ± 3.4 (99%)	0.2 ± 0.1 (1%)	19.6 ± 3.2 (93%)	1.6 ± 0.4 (8%)	–	21.1 ± 3.3
strawberry OSC[f] (*n* = 1)	37.8 (91%)	4.0 (9%)	34.3 (82%)	7.4 (18%)	–	41.7
vegetables	–	–	–	–	–	–
1. black bean (*n* = 1)	44.5 (100%)	–	36.1 (81%)	8.4 (19%)	–	44.5
2. eggplant (*n* = 1)	85.7 (100%)	–	0.6 (1%)	71.5 (83%)	13.6 (16%)	85.7
3. red cabbage (*n* = 4)	46.7 ± 7.3 (15%)	275 ± 33.8 (85%)	–	T	322 ± 40.8 (100%)	322 ± 40.8
4. red leaf lettuce (*n* = 8)	2.1 ± 1.5 (95%)	0.1 ± 0.1 (5%)	2.2 ± 1.5 (100%)	–	–	2.2 ± 1.5
5. red onion (*n* = 1)	11.0 (23%)	37.5 (77%)	33.6 (69%)	14.9 (31%)	–	48.5
6. red radish (*n* = 7)	T	100.1 ± 30.0 (100%)	–	T	100.1 ± 30.0 (100%)	100.1 ± 30.0
7. small red bean (*n* = 1)	6.7 (100%)	–	2.6 (39%)	4.1 (61%)	–	6.7
nuts	–	–	–	–	–	–
1. pistachio (*n* = 7)	7.5 ± 1.5 (100%)	–	7.5 ± 1.5 (100%)	–	–	7.5 ± 1.5

[a] Data expressed milligrams of anthocyanin (anthocyanidin glycosides or acylglycosides) on an "as is" weight basis and presented as mean ± SD for sample number >2. [b] Abbreviations: ACN, anthocyanin. [c] Sample number of each food. [d] Group 1, contained varieties 'Whinham' and 'Lancashine'; group 2, variety was 'Dan's Mistake'; group 3, variety was 'Careless'. [e] T, trace. [f] OSC, Oregon Strawberry Commission.

Quantification based upon HPLC can be done in three ways. The first way is to measure ACN aglycones after acid hydrolysis, which reduces the number of peaks on the chromatogram to six or fewer for most samples (*34*, *35*). However, it cannot provide any information regarding sugar acylation and the presence of acylated groups. The second way based upon HPLC is to calculate each ACN peak separately by using one standard, usually cyandin-3-glucoside or malvidin-3-glucoside. The third way is based on HPLC and using relevant anthocyanidin glucosides as standards, which is the method used in this paper. The molecular weight correction factors for the specific calculation of individual anthocyanins were determined by dividing the molecular weight of the anthocyanin to be quantified by that of the standard anthocyanin (*32*, *36*). Given that many anthocyanins are not available commercially, the third way is, to our knowledge, the most accurate method for quantification. The greatest challenge to obtaining good accuracy in the last two HPLC methods is to get good separation of all anthocyanins. This may sometimes require multiple tests and thus be time-consuming. The use of HPLC coupled to a mass spectrometer (MS/MS) can confirm whether adequate separation was achieved during the LC run. Three different gradients were needed for the separation of ACNs contained in the different foods included in this study (*26*, *27*).

Of over 100 different common foods in the U.S. market that were screened, 24 foods were found to contain ACNs (**Table 1**). The concentrations of ACNs in these foods were calculated individually (for detailed information, see Supplemental Tables 1−4, Supporting Information). However, only six common aglycones, delphinidin (Dp), cyanidin (Cy), petunidin (Pt), pelargonidin (Pg), peonidin (Pn), and malvidin (Mv) were found (**Figure 1**). ACNs grouped by these six common aglycones in these foods are presented in **Table 2**. Cy was found to be the most widely distributed aglycone, which was found in almost every food, but the concentrations varied considerably. Black raspberry contained the highest level of Cy-ACNs (669 mg/100 g). However, no Cy-ACN was found in black bean or eggplant. The highest amount of Dp-ACNs was found in black currant (333 mg/100 g), whereas red currant contained very low amounts. The best source of Pt and Mv was blueberries with wild blueberry (87.6 and 154.6 mg/100 g, respectively) containing higher amounts. Pn was found in various berries and grapes, with the highest amounts in cranberry (72.1 mg/100 g). Red radish was found to contain the highest level of Pg-ACNs (100.1 mg/100 g), and all of them were acylated ACNs, confirming previous identification work (*37*). Total ACNs in the foods analyzed in this study ranged from 0.7 mg/100 g of FW in one variety of gooseberry ('Careless') to 1480 mg/100 g of FW in chokeberry.

The concentrations of ACNs in the same foods were also presented according to the different degrees of glycosylation, namely, ACN monoglycoside, ACN diglycoside, and ACN triglycoside, as well as by nonacylated or acylated ACNs (**Table 3**). The reason for presenting the data in this manner is because ongoing research in our laboratory, as well as others, demonstrates that the sugar moieties and acylated groups can significantly affect the bioavailability, stability, and other biological effects of ACNs (*16*, *19*, *21*, *24*). These structural differences may further affect their bioactivities and their usage as natural food colorants. From our results, acylated ACNs are generally minor ACNs in most foods, with the percentages ranging from 0 to 25%. However, in four foods in this study, acylated ACNs comprised a majority of the ACNs with percentages >50%. They were Concord grape (50%), red onion (77%), red cabbage (85%), and red radish (100%) (**Table 3**). In terms of sugar moieties, most foods contained ACN monoglycosides as the major ACNs with percentages >50%. There were six foods in which ACN diglycosides were found to contribute a majority of the ACNs, which included sweet cherry (94%), black currant (76%), black raspberry (64%), red raspberry (52%), eggplant (83%), and small red bean (61%). Almost all ACNs in red cabbage and red radish were found to exist as ACN triglycosides.

ACNs were mainly found in fresh berries, fruits, and some vegetables. In processed foods in which foods containing ACNs were added as raw materials, such as canned foods, bread, cereals, and baby foods, ACNs generally could not be detected. ACNs were barely detected in baby foods prepared from fruits high in anthocyanins such as blueberries. In some foods that may contain a mixture of berries and fruits, such as baby foods, the amount of ACN-containing berry added may be so low that the ACNs were unable to be detected. These extremely low ACN concentrations may also result from their poor stability and possible destruction during processing. This is probably due to the unique chemical structure of anthocyanins relative to other flavonoids, which includes a positive charge on the C-ring of the aglycone at low pH, but renders them unstable at neutral pH. In contrast to the high intake of proanthocyanidins (*38*), intake of ACNs in infants is likely to be quite low, even though the infants may be consuming baby foods containing berries that should contain considerable ACNs. There is still much to be learned about the effects of processing and food matrix on anthocyanin stability (*39*−*41*).

Anthocyanin Consumption. On the basis of the calculated concentrations of ACNs in most common foods and in wines from published data (*42*), an estimation of the daily consumption of ACNs was made using NHANES 2001−2002 (*29*) (**Table 4**). The overall intake was estimated as 12.5 mg/day/person. However, in an earlier paper (*17*) published in the 1970s, the average daily intake of total ACNs was estimated to be 215 mg during the summer and 180 mg during the winter. Because the contents of total ACNs of many fruits and berries in the previously published paper (*17*) were not greatly different from our results, this huge difference of total ACN daily intake estimation must result from different food intake data. Valid and accurate food intake data are critical for intake estimation of dietary components. A further complication occurs when the same food is available in varieties or forms that may have distinctly different amounts of ACNs (i.e., red or white wine; dark or light colored juices, etc.), but the food intake information generally is not detailed enough to differentiate the intakes of the different sources. Thus, any estimation of ACN intake may

Table 4. Estimation of Daily Consumption of Anthocyanins from Fruits, Vegetables, and Beverages[a]

food	daily intake (g)	ACN (mg/100 g)	daily consumption (mg)
fruits			
apple, raw	11.77	0.6[b]	0.70
blackberry, raw	0.01	245	0.03
blueberry, raw	0.93	365	3.39
cherry, sweet, raw	0.46	122	0.56
cranberry, raw	0.12	140	0.17
grape, raw	4.83	36.7[c]	1.77
nectarine, raw	0.33	6.8	0.02
peach, raw	2.53	4.8	0.12
plum, raw	0.89	71.8[d]	0.64
raspberry, raw	0.24	390[e]	0.93
strawberry, raw	1.95	21.1[f]	0.41
subtotal			8.75
vegetables			
eggplant, raw	0.15	85.7	0.13
cabbage, red, raw	0.25	322	0.82
lettuce, red leaf, raw	0.27[g]	2.2	0.01
red radish, raw	0.14	100	0.14
onion, raw	7.92	12.1[h]	0.96
bean, black, raw	0.30	44.5[i]	0.13
subtotal			2.19
nuts			
pistachio nut	0.05	7.5	0.004
subtotal			0.004
beverages			
grape juice	6.61	14.0[j]	0.93
wine	12.31	10.7	0.66[k]
subtotal			1.68
total			12.53

[a] Food intake data from the National Health and Nutrition Examination Survey (NHANES 2001–2002) (*29*). [b] Of total apple being consumed, 45% of apples consumed were 'Red Delicious', 10% were 'Fuji', and 13% were 'Gala' based upon market consumption data. [c] Fifty percent of total intake of grapes was estimated as red grapes. [d] Plum and black plum were estimated as 50% each of the total plum consumption. [e] Raspberry and black raspberry were estimated as 50% each of the total raspberry consumption. [f] Because strawberry from OSC is grown exclusively in Oregon, concentration data of other strawberries were used. [g] No intake data for red leaf lettuce are available; however, intake of red leaf lettuce was estimated to be similar to that of green leaf lettuce. [h] Twenty-five percent of total onion was estimated as red onion. [i] Black beans are not normally consumed in a raw state, but usually following domestic cooking or commercial canning. Because ACNs readily leach into cooking water or canning brine, only 50–70% of the ACN would likely be retained in the cooked beans. Hence, this value may be overestimated. [j] Mean of 23 different brands of grape juice (data, based upon determination by pH differential, were provided by Dr. JoLynne Wightman from Welch's). [k] Fifty percent of wine consumed was estimated as red wine on the basis of U.S. supermarket data.

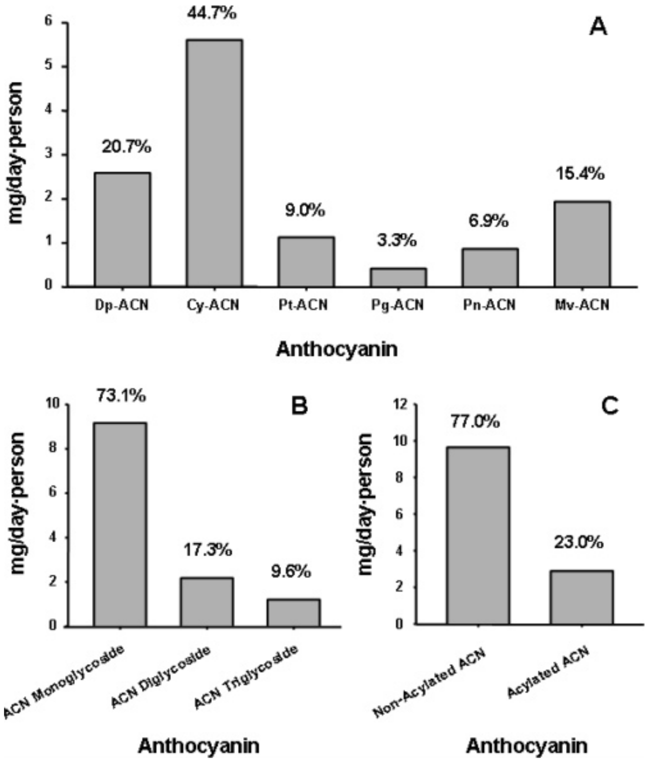

Figure 2. Daily intake of (**A**) ACNs with different anthocyanidins, (**B**) ACNs with different sugar moieties, and (**C**) nonacylated versus acylated ACNs.

be subject to considerable variability. On the basis of the present results and the NHANES food intake data, the earlier estimations of ACN intake (*17*) were very likely overestimated. In contrast, the daily intake of ACNs in Germany was estimated to average 2.7 mg/person in 2002 and varied between 0 and 76 mg/person (*43*). Our data provide a revised intake estimate and also provide intake estimates for specific ACNs (**Figure 2**).

On the basis of our data, the contribution of the different anthocyanidins to total ACN daily intake was quite different. Cy-ACNs contributed to 44.7% of total ACN daily intake, followed by Dp-ACNs (20.7%), Mv-ACNs (15.4%), Pt-ACNs (9.0%), Pn-ACNs (6.9%), and Pg-ACNs (3.3%) (**Figure 2A**). Furthermore, when comparing ACNs with different sugar moieties, we found that ACN monoglycosides contributed 73.1% of total ACN daily intake, whereas ACN diglycosides and triglycosides contributed only 17.3 and 9.6%, respectively (**Figure 2B**). For ACNs with nonacylated versus acylated groups, nonacylated ACNs were predominant, contributing 77.0% of the total daily ACN intake. Acylated ACNs contributed only 23.0% of total daily ACN intake (**Figure 2C**).

The daily intake of ACNs may vary widely among populations in different regions and seasons and among individuals with different education, financial status, and culture and as a result of other factors. Availability of certain berries and fruits may also be significant. In the United States and other countries, there has been increased emphasis on the need to consume more fruits and vegetables. Therefore, we may find more ACNs than what we estimated (**Table 4)** being consumed in the future. From **Table 4**, it is clear that the majority of the overall daily ACN intake is actually from fruits, berries, and two beverages (grape juice and wine). However, from the data in **Table 2**, it is also clear that intakes of >100 mg/day could be achieved easily with regular consumption of selected fruits or berries, such as blackberries, raspberries, blueberries, or Concord grapes. Unfortunately, many of these fruits or berries may be readily available only in certain seasons, so consistent consumption of high levels may be difficult. However, frozen storage is probably the best conventional commercial process to preserve ACNs in whole fruits, which allows consumers access to fresh fruits throughout the year. Consumption of food supplements or natural food colorants rich in ACNs is probably another way to increase intake.

Conclusions. ACNs were quantified in 24 foods, primarily fresh fruits and vegetables, from over 100 common foods sampled in the U.S. market. ACN composition and concentration varied significantly among these foods. On the basis of the quantification and intake data from NHANES 2001−2002, daily consumption of total ACNs in the United States was estimated to be 12.5 mg/day. Cy-ACNs contributed 44.7% of total ACN daily intake, followed by Dp-ACNs, Mv-ACNs, Pt-ACNs, Pn-ACNs, and Pg-ACNs. When ACNs with different sugar moieties were compared, ACN monoglycosides contributed 73.1% of total ACN daily intake, whereas ACN diglycosides and triglycosides contributed only 17.3 and 9.6%, respectively. For ACNs with nonacylated versus acylated groups, nonacylated ACNs were predominant, contributing 77.0%, and acylated ACNs contributed only 23.0% of total daily ACN intake.

ABBREVIATIONS USED

ACN, anthocyanin; Cy, cyanidin; Dp, delphinidin; FW, fresh weight; MS, mass spectrometer; Mv, malvidin; *m*/*z*, mass-to-charge ratio; Pg, pelargonidin; Pn, peonidin; Pt, petunidin, SPE, solid-phase extraction; TFA, trifluoroacetic acid.

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Supporting Information Available: Concentrations of anthocyanins in wild blueberry, Concord grape, berries, fruits, vegetables, nuts, red cabbage, and red radish. This material is available free of charge via the Internet at http://pubs.acs.org.

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