In this lab we are going to extend the "Caffeine from Tea" experiment we did previously. One way to design a research project is to start with a published procedure and then extend it to explore new aspects of the experimental topic. For example, in past years students have extended the "Caffeine from Tea" experiment by exploring the extraction of caffeine from caffeinated drinks using much the same procedure they used in the "Caffeine from Tea" experiment.

Another way of extending the "Caffeine from Tea" experiment would be to change one of the other variables other than the caffeine source. For example, the student could use another solvent instead of dichloromethane to extract the caffeine from tea and compare his/her results with those obtained with the dichloromethane extraction performed earlier.

Generally, it is advisable to change only one variable at a time in order to fully appreciate the impact of changing that one variable. It would also be advisable to work in groups to more fully explore the experimental topic. For example, three students could extract Red Bull with three different solvents.

In this lab we will attempt to quantitate the caffeine in your caffeine source based on UV and/or HPLC analysis of both the starting beverage and the caffeine obtained after extraction.

Prelab:

____Choose a caffeine source.

____ (1 point) Research the amount of caffeine you would expect to isolate from this source. Websites such as http://wilstar.com/caffeine.htm and http://www.energyfiend.com/the-caffeine-database/ are a good place to start. Record the amount of caffeine and your reference.

____(2 points) Design an experimental protocol, based on your experience with the "Caffeine from Tea" laboratory experiment. An important thing to keep in mind is that the separatory funnel has a maximum capacity of 250 mL. Include the glassware and equipment you will use (such as 200 mL beaker and UV-vis spectrophotometer). Include collecting a 3 mL sample of the caffeine source in your protocol. Hand in the caffeine obtained from the extraction by dissolving it in 10 mL of methanol.

____ (1 point) Consult the OChemOnline 2007 & 2008 student comments on the "Caffeine from Beverages" page <http://ochemonline.pbworks.com/Caffeine-from-Beverages>. Which comment did you find most helpful?

____ (3 points) Read the Journal of Food Chemistry and Toxicology article, "Caffeine Content of Prepackaged National-Brand and Private-Label Carbonated Beverages" and respond to the following questions.

1) What is the significance (importance) of such as study?
2) From Table 3, how many measurements of RC cola were done?
3) What does the term "isocratic" mean when applied to HPLC?

____ (1 point) Sample calculation: An unknown amount of crude caffeine was dissolved in 10 mL of methanol. 50 microliters of this solution were diluted to 2 milliliters in an HPLC sample vial. The HPLC determination was performed by injecting 20 microliters of the sample vial onto the column. The HPLC reported the amount of caffeine to be 3.3 micrograms. What is the amount of caffeine originally dissolved in methanol?

In lab: (4 points)

____Write down a detailed experimental method in your lab notebook of what you are doing in the lab.
____ Include the glassware you used (such as 200 mL beaker and 250 separatory funnel), grams of solid chemicals, and volumes of liquid chemicals.
____ Include the make and model of instruments that are used.
____Obtain the mass of your extracted caffeine before you subject it to chromatography and spectroscopy.
____ Record other observations as you would normally.

Do not sublime your extracted caffeine.

You will perform three experiments to determine the identity and purity of your extracted caffeine.

TLC:

Test your caffeine from this lab side-by-side with your "caffeine from tea" sample and a sample of pure caffeine on a TLC plate (three spots total). Record the results (including R_f values) in your lab notebook. Pay attention to the presence of impurities (non-caffeine spots).

UV spectrum.

Obtain an ultraviolet (UV) spectrum of your caffeine sample.
You will need to dissolve your sample in methanol to do this.
Determine the wavelength of maximum absorption in nanometers (nm).
Compare your UV spectrum to the spectrum of pure caffeine.

`

HPLC (High Pressure Liquid Chromatography):

This method gives much the same information as TLC. The caffeine sample is dissolved in a solvent and introduced into a column that separates the compounds in a mixture in much the same fashion as TLC. The eluant of the column is monitored with UV absorption. Compounds show up as "peaks" on the chromatogram.

Lab Report:

____ (1 point) Compare the mass of your caffeine obtained by weighing it to the mass calculated from the HPLC. Explain why they are different.
____ (1 point) Compare the caffeine content of your caffeine source obtained from a published source to the caffeine content calculated from the HPLC. Explain why they are different.
____ (1 point) Did you isolate caffeine? Explain.
____ (1 point) How pure is your isolated caffeine? Explain.
____ (3 points) Back to the Journal of Food Chemistry and Toxicology article, "Caffeine Content of Prepackaged National-Brand and Private-Label Carbonated Beverages."

a) What wavelength of UV light did they use to detect caffeine? According to your results was that an optimal wavelength?
b) How did they determine that their method was accurate in recording the exact amount of caffeine in 12 ounces of the beverage being tested.
c) What does the term "reversed-phase" mean when applied to HPLC?

____ (1 point) Environmental question. What effect does the massive consumption of caffeinated beverages have on the environment? Reference(s) please.

Caffeine Content of Prepackaged National-Brand and Private-Label Carbonated Beverages

K.-H. Chou and L.N. Bell

ABSTRACT: Caffeine is a well-known stimulant that is added as an ingredient to various carbonated soft drinks. Due to its stimulatory and other physiological effects, individuals desire to know the exact amount of caffeine consumed from these beverages. This study analyzed the caffeine contents of 56 national-brand and 75 private-label store-brand carbonated beverages using high-performance liquid chromatography. Caffeine contents ranged from 4.9 mg/12 oz (IGA Cola) to 74 mg/12 oz (Vault Zero). Some of the more common national-brand carbonated beverages analyzed in this study with their caffeine contents were Coca-Cola (33.9 mg/12 oz), Diet Coke (46.3 mg/12 oz), Pepsi (38.9 mg/12 oz), Diet Pepsi (36.7 mg/12 oz), Dr Pepper (42.6 mg/12 oz), Diet Dr Pepper (44.1 mg/12 oz), Mountain Dew (54.8 mg/12 oz), and Diet Mountain Dew (55.2 mg/12 oz). The Wal-Mart store-brand beverages with their caffeine contents were Sam's Cola (12.7 mg/12 oz), Sam's Diet Cola (13.3 mg/12 oz), Dr Thunder (30.6 mg/12 oz), Diet Dr Thunder (29.9 mg/12 oz), and Mountain Lightning (46.5 mg/12 oz). Beverages from 14 other stores were also analyzed. Most store-brand carbonated beverages were found to contain less caffeine than their national-brand counterparts. The wide range of caffeine contents in carbonated beverages indicates that consumers would benefit from the placement of caffeine values on food labels.

Keywords: caffeine, carbonated beverages, soft drinks

Introduction

Caffeine, 1,3,7-trimethylxanthine, is an odorless, slightly bitter substance found in numerous plant species (Tarka and Hurst 1998). Extracts derived from these plants, such as coffee and tea beverages, naturally contain caffeine and other methylxanthines. Caffeine is intentionally added as an ingredient to many carbonated soft drinks, including colas, pepper-type beverages, and citrus beverages. Although soda manufacturers may explain that caffeine contributes to the flavor of soft drinks, only 8% of adults were able to differentiate between caffeinated and caffeine-free colas at the concentration of caffeine contained in most cola beverages (Griffiths and Vernotica 2000). These beverages appeal to many consumers because of the stimulatory effect caffeine provides.

Caffeine has drawn more attention in the past decades due to its widespread consumption and physiological effects beyond that of its stimulatory effect (James 1991; Bernstein and others 2002; Mandel 2002). Caffeine is quickly absorbed by the body. The human salivary caffeine level, which indicates the extent of absorption, peaks around 40 min after caffeine consumption (Liguori and others 1997). Various physiological effects on the central nervous, cardiovascular, gastrointestinal, respiratory, and renal systems have been reported (Nehlig and others 1992; Spiller 1998; Hartley and others 2004; Savoca and others 2005). For example, Hartley and others (2004) reported that caffeine causes a mild elevation in blood pressure. In addition, caffeine's diuretic effect is widely known (Spiller 1998).

Various governmental bodies have specified the maximum level of caffeine allowed in carbonated beverages. The U.S. Food and Drug Admin. limits the amount of caffeine in carbonated beverages to a maximum of 0.02% (FDA 2006). Therefore, the highest legal amount of caffeine allowed in a 355 mL (12 oz) can of soft drink is about 72 mg. Likewise, Canada limits caffeine to cola-type beverages at a level of 200 ppm or about 71 mg/12 oz (Dept. of Justice 2007). In Australia, the maximum caffeine level in cola-type beverages must not exceed 145 mg/kg or about 51 mg/12 oz while in New Zealand, the caffeine level is limited to 200 mg/kg or about 71 mg/12 oz (FSANZ 2000).

The amount of caffeine contained in various foods and beverages has been analyzed, including coffee (Bell and others 1996), tea (Hicks and others 1996; Friedman and others 2005; Pena and others 2005; Yao and others 2006), carbonated beverages (Bunker and McWilliams 1979; Strohl 1985; Grand and Bell 1997; Pena and others 2005), and chocolate products (Caudle and others 2001; Tokusoglu and Ünal 2002). The last large-scale study involving the caffeine contents of carbonated beverages was conducted 10 y ago where the caffeine contents of 24 fountain, 20 prepackaged national-brand, and 16 prepackaged private-label store-brand carbonated beverages were determined; the store-brand beverages were limited to products from 4 stores (Grand and Bell 1997). The U.S. Dept. of Agriculture Natl. Nutrient Database for Standard Reference, Release 19, provides broad classifications of carbonated beverages and includes average caffeine contents for 8 types of carbonated beverages (USDA 2006). Caffeine data for some national-brand beverages are also reported on manufacturer websites (A&W 2006; Coca-Cola 2006; Pepsi-Cola 2005; Dr Pepper 2006a, 2006b, 2006c; Sundrop 2006). Caffeine data for private-label store-brand beverages are not available.

New flavors, formulas, and brands of carbonated beverages continue to be introduced into the market. Manufacturers may gradually lower caffeine contents due to health concerns of some consumers or increase it to correspond to the demand for greater

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stimulatory effect by other consumers. Without caffeine values placed on the label, consumers are left relatively uninformed regarding the amount of caffeine contained in these beverages. In addition, comprehensive databases on the caffeine contents of specific carbonated beverages are lacking. Therefore, the specific objective of this research project was to measure the caffeine contents of national and private-label store-brand carbonated beverages so that current data will be available to the scientific community and public.

Materials and Methods

Chemicals and reagents

Anhydrous caffeine used for preparing the standard solutions was purchased from Sigma Chemical Co. (St. Louis, Mo., U.S.A.). Sodium phosphate monobasic, phosphoric acid, and high-performance liquid chromatography (HPLC) grade acetonitrile were obtained from Fisher Scientific (Pittsburgh, Pa., U.S.A.). Deionized water was obtained from a water purification system (18 $M\Omega cm^{-1}$ quality).

Samples and sample preparation

Fifty-six varieties of national-brand prepackaged (cans and bottles) carbonated beverages were collected across the southeastern United States; these are listed in Table 1 along with their manufacturers. Seventy-five types of private-label store-brand beverages were acquired from 10 grocery stores, 2 pharmacies, 2 general merchandise stores, and 1 mini-market. Names of these stores, along with the beverage names, are provided in Table 2. Carbonated colas, pepper-type beverages (that is, like Dr Pepper), and citrus beverages (that is, like Mountain Dew), as well as their diet varieties, were analyzed in this study. Average caffeine contents of each carbonated beverage were determined from a minimum of 2 different lots. The beverages analyzed in this study were purchased between June 2005 and July 2006. The unopened beverages were stored at room temperature until analysis.

Each sample was degassed via sonication and diluted 3-fold with deionized water (1 mL sample + 2 mL water). Duplicate dilutions were made for all samples. An aliquot of each diluted sample was injected into the HPLC system to quantify the caffeine concentration.

Apparatus

The caffeine content was determined by isocratic reverse-phase HPLC equipped with a UV/visible detector, adapted from that used by Grand and Bell (1997). The chromatographic separation occurred on a Prodigy (150 × 4.6 mm) C-18 column (Phenomenex, Torrance, Calif., U.S.A.) in series with a Novapak (150 × 3.9 mm) C-18 column (Waters, Eatontown, N.J., U.S.A.). The mobile phase consisted of 20% (v/v) acetonitrile mixed with 80% (v/v) 0.1% aqueous sodium phosphate monobasic, acidified to pH 3 with phosphoric acid. The combination of these 2 analytical columns eliminated interference

Table 1—National-brand carbonated beverages listed by company

Company (headquarters)	Beverages
Coca-Cola Company (Atlanta, Ga., U.S.A.)	Coca-Cola, Diet Coke, Cherry Coke, Diet Cherry Coke, Coke with Lime, Diet Coke with Lime, Vanilla Coke, Diet Vanilla Coke, Coca-Cola C2, Diet Coke with Splenda, Coke Zero, Coca-Cola Black Cherry Vanilla, Diet Coke Black Cherry Vanilla, Tab, Pibb Xtra, Pibb Zero, Vault Citrus, Vault Zero, Barq's Root Beer, and Mello Yello
Pepsico Inc. (Somers, N.Y., U.S.A.)	Pepsi, Diet Pepsi, Cherry Pepsi, Diet Cherry Pepsi, Pepsi with Lime, Diet Pepsi with Lime, Vanilla Pepsi, Diet Vanilla Pepsi, Pepsi One, Mountain Dew, Diet Mountain Dew, Mountain Dew Code Red, Diet Mountain Dew Code Red
Natl. Beverage Co. (Ft. Lauderdale, Fla., U.S.A.)	Faygo Cola, Faygo Moon Mist, Ritz Cola, Shasta Cola
Carolina Beverage Corp. (Salisbury, N.C., U.S.A.)	Cheerwine, Diet Cheerwine
Dr Pepper/Seven Up Inc. (Plano, Tex., U.S.A.)	Dr Pepper, Diet Dr Pepper, Dr Pepper Berries & Cream, Diet Dr Pepper Berries & Cream, Cherry Vanilla Dr Pepper, Diet Cherry Vanilla Dr Pepper, RC Cola, Diet RC, SunDrop, Diet SunDrop, A & W Cream Soda, Sunkist, and Diet Sunkist
Buffalo Rock Co. (Birmingham, Ala., U.S.A.)	Dr. Wham, Diet Dr. Wham
Big Red Inc. (Waco, Tex., U.S.A.)	Big Red
Red Rock Corp. (Indianalopis, Ind., U.S.A.)	Red Rock Cola

Table 2—Private-label store-brand beverages listed by store

Store (headquarters)	Beverages
Kroger (Cincinnati, Ohio, U.S.A.)	Big K Cola, Big K Diet Cola, Big K Cherry Cola, Big K Diet Cherry Cola, Big K Cola with Lime, Big K Diet Cola with Lime, Dr. K, Diet Dr. K, Big K Citrus Drop, Big K Diet Citrus Drop
Winn-Dixie (Jacksonville, Fla., U.S.A.)	Chek Cola, Chek Diet Cola, Chek Cherry Cola, Chek Vanilla Cola, Chek Diet Vanilla Cola, Chek Diet Cola with Lime, Chek Mate Cola, Dr. Chek, Diet Dr. Chek, Chek Kountry Mist, Chek Diet Kountry Mist, Chek Red Alert
Wal-Mart (Bentonville, Ark., U.S.A.)	Sam's Cola, Sam's Diet Cola, Dr Thunder, Diet Dr Thunder, Sam's Mountain Lightning
Bruno's (Birmingham, Ala., U.S.A.)	Rally Cola, Rally Diet Cola, Dr. Bob, Diet Dr. Bob, Ramp, Ramp Red
Publix Super Markets (Lakeland, Fla., U.S.A.)	Publix Cola, Publix Diet Cola, Publix Cherry Cola, Dr. Publix, Publix Citrus Hit
Dollar General (Goodlettsville, Tenn., U.S.A.)	CloverValley Cola, CloverValley Diet Cola, Dr Topper, CloverValley Citrus Drop
Save-a-Lot Food Stores (Earth City, Mo., U.S.A.)	Bubba Cola, Diet Bubba Cola, Dr Pop, Diet Dr Pop, Mountain Holler
Piggly Wiggly (Memphis, Tenn., U.S.A.)	Piggly Wiggly Cola, Piggly Wiggly Diet Cola, Mr. Pig, Mountain Yeller
7-Eleven (Dallas, Tex., U.S.A.)	Big Gulp Cola, Big Gulp Diet Cola
Supervalu (Eden Prairie, Minn., U.S.A.)	Superchill Cola, Superchill Diet Cola, Dr. Chill, Mountain Chill
Food Lion (Salisbury, N.C., U.S.A.)	Food Lion Cola, Food Lion Diet Cola, Dr. Perky, Mountain Lion
Ingle's Markets (Asheville, N.C., U.S.A.)	Laura Lynn Cola, Laura Lynn Diet Cola, Laura Lynn Cherry Cola, Dr Lynn, Diet Dr Lynn, Mountain Moon Drops
IGA (Chicago, Ill., U.S.A.)	IGA Cola, IGA Diet Cola, IGA Spring Mist, Dr. IGA
Walgreens (Deerfield, Ill., U.S.A.)	Walgreens Cola, Walgreens Diet Cola
Rite Aid (Harrisburg, Pa., U.S.A.)	Big Fizz Cola, Big Fizz Diet Cola

caused by other components in some samples, such as colors, artificial sweeteners, flavors, and preservatives. The wavelength of detection was set at 254 nm, and flow rate was set at 1 mL/min. Separation was performed at room temperature. Caffeine eluted around 4.1 min. Data were recorded by a Hewlett Packard HP3395 integrator (Palo Alto, Calif., U.S.A.). From spiking Caffeine-Free Diet Coke (Coca-Cola, Atlanta, Ga., U.S.A.) with known amounts of caffeine, the percentage recovery for this method was determined to be 96.7% to 100.8% with a coefficient of variation of 0.6%. These values were similar to those reported by Grand and Bell (1997). A sample chromatogram for the analysis of a pepper-type beverage is shown in Figure 1.

Data analysis

Every type of beverage underwent duplicate measurements per lot; these were averaged to give the mean caffeine content for that lot. Data from these duplicate dilutions were typically found to vary by less than 2%. The caffeine contents for the various lots were then averaged to give the mean caffeine contents of the beverages along with the standard deviation. Because these beverages are so commonly distributed and consumed in 12-ounce cans, the caffeine values are reported in terms of milligrams per 12 oz.

Results and Discussion

National-brand colas

The caffeine contents of 31 national-brand colas are listed in Table 3 along with available manufacturer data. The caffeine contents of this group ranged from 10.3 to 57.1 mg/12 oz. The highest value (57.1 mg/12 oz) was found in Pepsi One. Except for the lower caffeine contents of Ritz Cola and Red Rock Cola and the higher caffeine content of Pepsi One, the remaining samples contained 33.3 to 48.1 mg caffeine/12 oz. The caffeine values of some national-brand colas (Coke, Diet Coke, Pepsi, and Diet Pepsi) were 13% to 20% higher than determined 10 y ago (Grand and Bell 1997). Caffeine values for Tab, RC Cola, and Shasta Cola were similar to those reported previously (Grand and Bell 1997). The caffeine values determined in this study were consistent with the available manufacturer data. However, the USDA nutrient database gave an average caffeine content of 29 mg/12 oz beverage for regular cola products (USDA 2006), which was lower than most of the values determined in the present study. For diet cola products, the USDA gave an average caffeine content of 43 mg/12 oz, which also does not adequately represent the range of caffeine values.

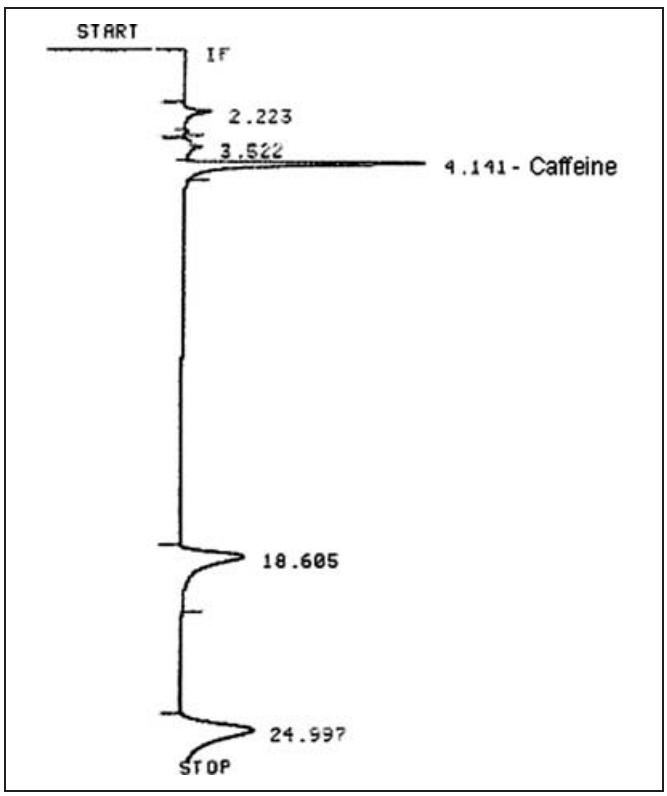

Figure 1—Representative HPLC chromatogram for the analysis of caffeine in Dr Pepper. Separation occurred on 2 C-18 columns in series using a mobile phase of 20% (v/v) acetonitrile and 80% (v/v) 0.1% aqueous sodium phosphate monobasic, acidified to pH 3 flowing at 1 mL/min. Detection occurred at 254 nm.

National-brand pepper-type beverages

The caffeine contents of 10 national-brand pepper-type beverages are reported in Table 4. All samples in this group contained similar caffeine contents, with values ranging from 39.4 to 44.1 mg/12 oz. These caffeine values were similar to those of national-brand pepper-type beverages determined previously (Grand and Bell 1997), as well as data on the available manufacturer websites. The USDA nutrient database gave an average caffeine content of 43 mg/12 oz for diet pepper-type beverages (USDA 2006), which is consistent with the current data. On the other hand, the database

Table 3—Caffeine contents (mean ± standard deviation) of national-brand colas

	Caffeine content (mg/12 oz)	
Beverage	**Current study**	**Manufacturer data**
Pepsi One ($n = 2$)[c]	57.1 ± 3.3	54[a]
Diet Cheerwine ($n = 2$)	48.1 ± 1.1	n/a[d]
Tab ($n = 2$)	48.1 ± 1.9	46.5[b]
Cheerwine ($n = 2$)	47.5 ± 1.4	n/a
Diet RC ($n = 2$)	47.3 ± 1.6	n/a
Diet Coke ($n = 3$)	46.3 ± 1.7	46.5[b]
Diet Coke with Lime ($n = 2$)	46.3 ± 2.7	46.5[b]
RC Cola ($n = 4$)	45.2 ± 4.1	n/a
Diet Vanilla Coke[e] ($n = 1$)	44.5	46.5[b]
Shasta Cola ($n = 2$)	42.9 ± 2.2	n/a
Faygo Cola ($n = 2$)	41.7 ± 3.0	n/a
Diet Cherry Pepsi ($n = 2$)	40.5 ± 2.7	37.5[a]
Cherry Pepsi ($n = 2$)	39.7 ± 3.2	37.5[a]
Pepsi ($n = 3$)	38.9 ± 1.0	37.5[a]
Pepsi with Lime ($n = 2$)	38.4 ± 2.0	37.5[a]
Diet Vanilla Pepsi[e] ($n = 1$)	38.1	37.5[a]
Vanilla Pepsi[e] ($n = 1$)	37.4	37.5[a]
Diet Coke Black Cherry Vanilla ($n = 2$)	36.8 ± 1.4	34.5[b]
Diet Pepsi ($n = 3$)	36.7 ± 0.6	36[a]
Diet Pepsi with Lime ($n = 2$)	36.4 ± 0.9	37.5[a]
Coke Zero ($n = 2$)	35.8 ± 2.6	34.5[b]
Coca-Cola Black Cherry Vanilla ($n = 2$)	35.1 ± 1.1	34.5[b]
Diet Cherry Coke ($n = 2$)	35.0 ± 2.0	34.5[b]
Cherry Coke ($n = 2$)	34.4 ± 1.8	34.5[b]
Coca-Cola C2 ($n = 2$)	34.4 ± 1.5	34.5[b]
Diet Coke with Splenda (n = 2)	34.4 ± 1.3	34.5[b]
Coca-Cola ($n = 3$)	33.9 ± 0.9	34.5[b]
Coke with Lime ($n = 2$)	33.6 ± 1.1	34.5[b]
Vanilla Coke[e] ($n = 1$)	33.3	34.5[b]
Red Rock Cola ($n = 2$)	26.1 ± 1.0	n/a
Ritz Cola ($n = 2$)	10.3 ± 0.9	n/a

[a] Pepsi-Cola (2005).
[b] Coca-Cola (2006).
[c] *n*: number of lots.
[d] n/a: not available.
[e] These products have been discontinued.

gave an average caffeine content of 37 mg/12 oz for regular pepper products (USDA 2006), which is slightly lower than the values determined in this study.

National-brand citrus beverages

The caffeine contents of 10 national-brand citrus beverages are also reported in Table 4. The caffeine contents of this group ranged from 19.7 to 74.0 mg/12 oz. The greatest caffeine content (74.0 mg/12 oz) was found in Vault Zero. Except for the lowest caffeine content of Faygo Moon Mist (19.7 mg/12 oz), the other beverages contained more than 49 mg caffeine per 12 oz. These data were consistent with the available caffeine data from manufacturer websites. The caffeine contents of regular and diet Mountain Dew and Mello Yello from the present study and those from Grand and Bell (1997) were also similar. The USDA nutrient database gave an average caffeine content of 55 mg/12 oz beverage for regular caffeinated lemon-lime beverages (USDA 2006). For the purpose of this study, it is assumed that the lemon-lime caffeinated beverage classification by USDA refers to regular citrus products because there is no other carbonated citrus beverage category. Another USDA classification (carbonated beverage, low calorie, other than cola or pepper, with aspartame, contains caffeine) could include diet citrus beverages; this beverage category had an average caffeine level of 53 mg/12 oz (USDA 2006). Five out of 10 national-brand citrus products were found to be similar to the data from USDA. The other 5 citrus products were quite different from that in the USDA database. The caffeine contents of regular and diet SunDrop as well as Vault Citrus and Vault Zero were 17% to 34% greater than the values listed by USDA. For the citrus beverages, it was challenging to determine which USDA category was appropriate to use. Clearer descriptions of database categories would reduce this ambiguity.

Table 4—Caffeine contents (mean ± standard deviation) of national-brand pepper-type, citrus, and miscellaneous beverages

Beverage	Caffeine content (mg/12 oz)	
	Current study	Manufacturer data
Pepper-type beverages		
Diet Dr Pepper ($n = 2$)[h]	44.1 ± 2.3	41[c]
Dr Pepper ($n = 3$)	42.6 ± 2.0	41[c]
Diet Dr Pepper Berries & Cream ($n = 2$)	42.0 ± 1.1	41[e]
Diet Dr. Wham ($n = 2$)	41.9 ± 0.8	n/a[i]
Dr. Wham ($n = 2$)	41.6 ± 0.3	n/a
Pibb Zero ($n = 2$)	41.2 ± 0.2	40.5[b]
Dr Pepper Berries & Cream ($n = 2$)	41.1 ± 0.5	41[d]
Pibb Xtra ($n = 2$)	40.3 ± 2.5	40.5[b]
Diet Cherry Vanilla Dr. Pepper ($n = 2$)	40.1 ± 1.0	37[c]
Cherry Vanilla Dr. Pepper ($n = 2$)	39.4 ± 1.3	37[c]
Citrus beverages		
Vault Zero ($n = 2$)	74.0 ± 1.7	70.5[b]
Diet SunDrop ($n = 2$)	71.5 ± 1.9	69[f]
Vault Citrus ($n = 2$)	70.6 ± 0.7	70.5[b]
SunDrop ($n = 2$)	64.7 ± 2.0	63[f]
Diet Mountain Dew Code Red ($n = 2$)	55.4 ± 1.3	54[a]
Diet Mountain Dew ($n = 2$)	55.2 ± 0.3	54[a]
Mountain Dew ($n = 2$)	54.8 ± 2.5	54[a]
Mountain Dew Code Red ($n = 2$)	54.3 ± 0.3	54[a]
Mello Yello ($n = 2$)	49.5 ± 1.8	52.5[b]
Faygo Moon Mist ($n = 3$)	19.7 ± 3.0	n/a
Miscellaneous beverages		
Diet Sunkist ($n = 2$)	41.5 ± 0.3	n/a
Sunkist ($n = 2$)	40.6 ± 0.2	n/a
Big Red ($n = 2$)	34.0 ± 0.5	n/a
A & W Cream Soda ($n = 2$)	28.6 ± 1.4	28.8[g]
Barq's Root Beer ($n = 2$)	22.4 ± 1.4	22.5[b]

[a]Pepsi-Cola (2005).
[b]Coca-Cola (2006).
[c]Dr Pepper (2006c).
[d]Dr Pepper (2006b).
[e]Dr Pepper (2006a).
[f]Sundrop (2006).
[g]A&W (2006).
[h]*n*: number of lots.
[i]n/a: not available.

Miscellaneous national-brand beverages

The caffeine contents of 5 miscellaneous national-brand beverages are also reported in Table 4. The caffeine content of Big Red (34.0 mg/12 oz) was similar to the majority of national-brand cola beverages. The USDA nutrient database gave no caffeine content for carbonated orange products (USDA 2006), but the regular and diet Sunkist beverages were found to contain 40.6 and 41.5 mg caffeine per 12 oz, respectively. These values were comparable to caffeine values reported previously (Grand and Bell 1997). In addition, the USDA nutrient database gave no caffeine content for root beer or cream soda products (USDA 2006). However, caffeine contents of 22.4 and 28.6 mg/12 oz were found in Barq's Root Beer and A & W Cream Soda, respectively. The USDA caffeine values for these beverage categories are inaccurate based on both current and previous data. Because these products may or may not contain caffeine, careful evaluation of the product's ingredient list is advised.

Private-label store-brand colas

The caffeine contents of 41 private-label store-brand regular and diet colas are reported in Table 5. The caffeine contents of regular colas ranged from 4.9 mg (IGA Cola) to 46.4 mg (Rite Aid's Big Fizz Cola) caffeine per 12 oz. The caffeine contents of diet colas ranged from 10.3 mg (IGA Diet Cola) to 61.9 mg (Rite Aid's Big Fizz Diet Cola) caffeine per 12 oz. The range of caffeine contents of this group

Table 5—Caffeine contents (mean ± standard deviation) of private-label store-brand regular and diet colas

Beverage	Caffeine content (mg/12 oz)	
	Regular cola	Diet cola
Big Fizz Cola[a]	46.4 ± 15.8 ($n = 3$)[p]	61.9 ± 2.4 ($n = 3$)
Big K Cherry Cola[b]	43.0 ± 2.9 ($n = 2$)	39.9 ± 1.8 ($n = 2$)
Walgreens Cola[c]	39.2 ± 8.1($n = 3$)	45.0 ± 6.7 ($n = 3$)
Big K Cola[b]	38.8 ± 2.2 ($n = 3$)	30.0 ± 1.6 ($n = 3$)
Big Gulp Cola[d]	38.6 ± 0.6 ($n = 3$)	30.0 ± 1.6 ($n = 2$)
Chek Vanilla Cola[e]	36.3 ± 2.3 ($n = 2$)	28.9 ± 2.0 ($n = 2$)
Bubba Cola[f]	35.4 ± 1.6 ($n = 3$)	42.0 ± 2.2 ($n = 4$)
Chek Cola[e]	34.7 ± 1.8 ($n = 3$)	27.5 ± 1.7 ($n = 3$)
Big K Cola with Lime[b]	30.3 ± 0.5 ($n = 2$)	18.6 ± 0.3 ($n = 2$)
CloverValley Cola[g]	28.8 ± 5.7 ($n = 6$)	22.9 ± 6.4 ($n = 7$)
Chek Cherry Cola[e]	26.3 ± 1.2 ($n = 2$)	n/a[q]
Food Lion Cola[h]	25.3 ± 0.8 ($n = 3$)	11.9 ± 0.8 ($n = 3$)
Laura Lynn Cola[i]	24.4 ± 1.8 ($n = 4$)	11.3 ± 0.6 ($n = 3$)
Superchill Cola[j]	24.2 ± 0.9 ($n = 3$)	34.5 ± 0.6 ($n = 3$)
Publix Cola[k]	23.1 ± 2.2 ($n = 3$)	35.2 ± 2.9 ($n = 3$)
Rally Cola[l]	13.3 ± 1.5 ($n = 3$)	13.0 ± 2.1 ($n = 3$)
Piggly Wiggly Cola[m]	12.7 ± 1.5 ($n = 3$)	11.9 ± 2.0 ($n = 3$)
Sam's Cola[n]	12.7 ± 1.0 ($n = 3$)	13.1 ± 1.3 ($n = 3$)
Publix Cherry Cola[k]	12.4 ± 2.1 ($n = 2$)	n/a
Laura Lynn Cherry Cola[i]	8.4 ± 1.8 ($n = 2$)	n/a
IGA Cola[◦]	4.9 ± 1.1 ($n = 3$)	10.3 ± 1.2 ($n = 3$)
Chek Mate Cola[e]	n/a	26.2 ± 1.4 ($n = 2$)
Chek Diet Cola with Lime[e]	n/a	45.8 ± 4.2 ($n = 2$)

[a]Rite Aid; [b]Kroger; [c]Walgreens; [d]7-Eleven; [e]Winn-Dixie; [f]Save-a-Lot; [g]Dollar General; [h]Food Lion; [i]Ingle's; [j]Supervalu; [k]Publix; [l]Bruno's; [m]Piggly Wiggly; [n]Wal-Mart; [◦]IGA; [p]n: number of lots; and [q]n/a: not available.

was unlike the spread of national-brand colas, being much wider. Big Fizz Diet Cola contained more caffeine than any cola product, national or store brand; many other store brands contained less than 20 mg caffeine per 12 oz. Because of the large caffeine content range of these products, it is difficult to generalize the amount of caffeine being consumed from such products.

The caffeine values of Winn-Dixie's Chek Diet Cola, Kroger's Big K Diet Cola, Wal-Mart's Sam's Cola, and Sam's Diet Cola were comparable to those determined previously (Grand and Bell 1997). Big K Cola was found to contain 38.8 mg caffeine/12 oz, which is over 600% higher than the value of 5.2 mg caffeine/12 oz, reported 10 y ago by Grand and Bell (1997). Similarly, Chek Cola contained 29% more caffeine in this study (34.7 mg/12 oz) than that reported previously (27.0 mg/12 oz) by Grand and Bell (1997). These products have clearly been reformulated over the past decade. The USDA nutrient database gave average caffeine contents of 29 and 43 mg/12 oz beverage for regular and diet cola products, respectively (USDA 2006). However, the USDA database is impractical to use due to the wide range of caffeine values in private-label store-brand colas (4.9 to 61.9 mg/12 oz).

Private-label store-brand pepper-type beverages

The caffeine contents of 18 private-label store-brand pepper-type beverages are reported in Table 6. The caffeine contents of this group ranged from 18.2 to 59.8 mg/12 oz. The lowest and highest caffeine concentrations were found in Ingle's Diet Dr Lynn and Dr IGA, respectively. The caffeine contents of the samples were distributed evenly within this range. The distribution of this group was different from national pepper-type beverages, all of which contained around 40 mg caffeine per 12 oz. Dr IGA was found to contain more caffeine than any pepper-type beverage, national or store-brand, while several store-brand beverages contained less than half the caffeine of the national-brand products. The caffeine contents of Kroger's regular and diet Dr K were much higher (> 150%) than those analyzed by Grand and Bell (1997), indicating that the products have been reformulated. Similarly, the caffeine content of Winn-Dixie's Dr Chek analyzed in the present study was 33% higher than that reported previously (Grand and Bell 1997). The USDA nutrient database gave average caffeine contents of 37 and 43 mg/12 oz for regular and diet pepper-type drinks, respectively, which again does not adequately represent the wide distribution of the current results.

Table 6—Caffeine contents (mean ± standard deviation) of private-label store-brand pepper-type and citrus beverages

Beverage	Caffeine content (mg/12 oz)
Pepper-type beverages	
Dr IGA[a] ($n = 2$)[m]	59.8 ± 3.7
Diet Dr Pop[b] ($n = 2$)	56.8 ± 2.0
Dr Pop[b] ($n = 5$)	47.5 ± 11.0
Dr K[c] ($n = 2$)	41.2 ± 2.8
Diet Dr K[c] ($n = 2$)	40.7 ± 2.5
Dr Topper[d] ($n = 2$)	34.0 ± 2.7
Dr Publix[e] ($n = 2$)	31.6 ± 2.0
Dr Bob[f] ($n = 2$)	31.3 ± 1.6
Mr. Pig[g] ($n = 2$)	31.2 ± 2.3
Diet Dr Bob[f] ($n = 2$)	30.9 ± 0.6
Dr Thunder[h] ($n = 2$)	30.6 ± 1.3
Dr Chill[i] ($n = 2$)	29.9 ± 1.8
Diet Dr Thunder[h] ($n = 2$)	29.9 ± 0.8
Dr Chek[j] ($n = 2$)	24.4 ± 1.3
Diet Dr Chek[j] ($n = 2$)	22.3 ± 1.3
Dr Lynn[k] ($n = 2$)	19.3 ± 0.9
Dr Perky[l] ($n = 2$)	18.8 ± 1.5
Diet Dr Lynn[k] ($n = 2$)	18.2 ± 1.0
Citrus beverages	
Chek Kountry Mist[j] ($n = 2$)	55.1 ± 4.9
Ramp Red[f] ($n = 2$)	54.6 ± 1.0
IGA Spring Mist[a] ($n = 2$)	54.2 ± 4.4
Publix Citrus Hit[e] ($n = 2$)	54.1 ± 1.0
Ramp[f] ($n = 2$)	53.8 ± 1.0
Mountain Chill[i] ($n = 2$)	53.5 ± 0.8
Chek Red Alert[j] ($n = 2$)	53.2 ± 1.8
Mountain Holler[b] ($n = 2$)	53.1 ± 0.4
Mountain Yeller[g] ($n = 2$)	53.1 ± 0.1
CloverValley Citrus Drop[d] ($n = 2$)	52.0 ± 0.3
Sam's Mountain Lightning[h] ($n = 2$)	46.5 ± 1.0
Chek Diet Kountry Mist[j] ($n = 4$)	46.3 ± 7.7
Mountain Lion[l] ($n = 2$)	30.9 ± 0.1
Laura Lynn Mountain Moon Drops[k] ($n = 4$)	27.5 ± 7.2
Big K Citrus Drop[c] ($n = 2$)	26.2 ± 0.5
Big K Diet Citrus Drop[c] ($n = 2$)	25.1 ± 0.5

[a]IGA; [b]Save-a-Lot; [c]Kroger; [d]Dollar General; [e]Publix; [f]Bruno's; [g]Piggly Wiggly; [h]Wal-Mart; [i]Supervalu; [j]Winn-Dixie; [k]Ingle's; [l]Food Lion; and [m]*n*: number of lots.

Private-label store-brand citrus beverages

The caffeine contents of 16 private-label store-brand citrus beverages are also reported in Table 6. The caffeine contents of this group ranged from 25.1 to 55.1 mg/12 oz. The lowest and highest caffeine concentrations were found in Kroger's Big K Diet Citrus Drop and Winn-Dixie's Chek Kountry Mist, respectively. Ten beverages within this group contained over 50 mg caffeine per 12 oz. The USDA nutrient database gave an average caffeine content of 55 mg/12 oz for lemon-lime (citrus) products (USDA 2006). Most of this group's results were similar to the value from USDA. Kroger's Big K products contained approximately half the caffeine of the value listed by USDA. The amounts of caffeine existing in Chek Kountry Mist, Sam's Mountain Lightning (from Wal-Mart), Big K Citrus Drop, and Big K Diet Citrus Drop were similar to the values reported by Grand and Bell (1997).

Quality control of store-brand beverages

Based upon the standard deviations listed in Table 3 to 6, the quality control of national-brand beverages appeared generally better than that for the store-brand beverages. Additional lots were obtained and analyzed for some beverages whose duplicate lots had quite different caffeine values. Products displaying large variations between lots included Rite Aid's Big Fizz Cola, Walgreens Cola, Walgreens Diet Cola, Dollar General's CloverValley Cola, CloverValley Diet Cola, Save-a-Lot's Dr Pop, Winn-Dixie's Chek Diet Kountry Mist, and Ingle's Laura Lynn Mountain Moon Drop. In addition, 1 lot of Food Lion's Mountain Lion was found to contain no caffeine (this sample was not included in the data analysis). Thus, there appears to be less stringent quality control with store-brand products than with the national-brand products.

Mean caffeine contents in different beverage types

The average amounts of caffeine existing in each beverage classification are tabulated in Table 7. The average caffeine values for national-brand cola and pepper-type beverages were similar. The national-brand citrus beverages contained more caffeine than cola and pepper-type beverages. National-brand diet colas contained, on average, more caffeine than the regular colas. One may suggest that this result is due to caffeine being diluted by the bulk from added sugar. However, the 9% to 11% sugar added to regular colas causes the solution volume to increase by less than 7% (Weast 1972). Therefore, adding 11% sugar to a beverage containing 42 mg caffeine/12 oz would only dilute the caffeine content to 39 mg/12 oz. Furthermore, the data in Table 1 do not show a pattern with regard to the differences between caffeine levels in regular and diet colas. Thus, specific formulation changes, not simply dilution from the sugar, account for

Table 7 — Mean caffeine contents (mg/12 oz) with standard deviation by beverage classification

Beverage	National brand	Store brand	Overall average	USDA (2006)
Cola with sugar	35.8 ± 8.6 ($n = 16$)[a]	26.6 ± 12.2 ($n = 21$)	30.6 ± 11.6 ($n = 37$)	29
Diet cola	42.1 ± 6.7 ($n = 15$)	28.0 ± 14.3 ($n = 20$)	34.0 ± 13.5 ($n = 35$)	43
Pepper-type	41.0 ± 1.2 ($n = 5$)	33.3 ± 11.6 ($n = 12$)	35.6 ± 10.3 ($n = 17$)	37
Diet pepper	41.9 ± 1.5 ($n = 5$)	33.1 ± 14.0 ($n = 6$)	37.1 ± 10.9 ($n = 11$)	43
Citrus	52.3 ± 17.7 ($n = 6$)	47.7 ± 10.8 ($n = 14$)	49.1 ± 12.9 ($n = 20$)	55
Diet citrus	64.0 ± 10.1 ($n = 4$)	35.7 ± 15.0 ($n = 2$)	54.6 ± 17.9 ($n = 6$)	53

[a] *n*: number of beverage types.

the different caffeine levels. With respect to store-brand beverages, their caffeine contents were, on average, lower than the national-brand counterparts. In addition, the variation between store brands was generally greater than between national brands.

The USDA data in Table 7 represent the average caffeine values for a given beverage type. Interestingly, the overall caffeine averages for the regular colas, regular pepper-type beverages, and diet citrus beverages were close to the USDA values. Contrary to this result, the average caffeine contents of the diet colas, diet pepper-type beverages, and regular citrus beverages were lower than listed by USDA. When the beverages were categorized into national and store brands, additional discrepancies appeared. For the diet cola, diet pepper, and regular citrus beverage categories, the mean national-brand data were similar to the USDA values. However, for the national-brand regular cola, regular pepper-type, and diet citrus beverages, the average caffeine values were greater than those reported in the USDA database. The average caffeine data from store-brand beverages were all lower than those listed by USDA. The USDA classifications of caffeinated carbonated beverages should be broadened to differentiate between national-brand and store-brand categories, recognizing there remains a wide distribution within each beverage type.

Conclusion

The caffeine data collected in the present study suggest that consumers concerned about limiting daily caffeine ingestion from carbonated beverages may select the lower caffeine-containing store-brand beverages; however, a limited number of these beverages actually contain substantially more caffeine than national-brand products. In addition, although the store-brand beverages are less expensive, their caffeine levels tend to vary more between brands, and in some cases between different lots of the same brand, than the national-brand beverages. Consumers desiring caffeine may likewise select from higher caffeine-containing beverages. Because of the wide range of caffeine values (5 to 74 mg/12 oz.), broad generalizations about the caffeine contents of national and store-brand carbonated beverages are difficult to make. Our data may be used to update and expand the USDA nutrient database so that consumers have more current and accurate information. However, the best way for universal access to caffeine data is to place values on food labels so all consumers can be better informed about the amount of caffeine they are ingesting. Consistent with this recommendation, the Coca-Cola Co. and PepsiCo announced in February 2007 their intent to place caffeine contents on the labels of various carbonated beverages (IFT 2007). If all manufacturers placed caffeine contents on food labels, consumers would have the ability to instantly compare products, enabling them to make more informed purchasing decisions.

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