Preservation of Cocoa Antioxidant Activity, Total Polyphenols, Flavan-3-ols, and Procyanidin Content in Foods Prepared with Cocoa Powder

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ABSTRACT: Little is known about the effects of common cooking processes on cocoa flavanols. Antioxidant activity, total polyphenols (TP), flavanol monomers, and procyanidin oligomers were determined in chocolate frosting, a hot cocoa drink, chocolate cookies, and chocolate cake made with natural cocoa powder. Recoveries of antioxidant activity, TP, flavanol monomers, and procyanidins ranged from 86% to over 100% in the chocolate frosting, hot cocoa drink, and chocolate cookies. Losses were greatest in the chocolate cake with recoveries ranging from 5% for epicatechin to 54% for antioxidant activity. The causes of losses in baked chocolate cakes were investigated by exchanging baking soda with baking powder or combinations of the 2 leavening agents. Use of baking soda as a leavening agent was associated with increased pH and darkening color of cakes. Losses of antioxidant activity, TP, flavanol monomers, and procyanidins were associated with an increased extractable pH of the baked cakes. Chocolate cakes made with baking powder for leavening resulted in an average extractable pH of 6.2 with essentially complete retention of antioxidant activity and flavanol content, but with reduced cake heights and lighter cake color. Commercially available chocolate cake mixes had final pHs above 8.3 and contained no detectable monomeric flavanols after baking. These results suggest that baking soda causes an increase in pH and subsequent destruction of flavanol compounds and antioxidant activity. Use of an appropriate leavening agent to moderate the final cake pH to approximately 7.25 or less results in both good leavening and preservation of cocoa flavanols and procyanidins.

Keywords: antioxidant, cocoa powder, flavanol, leavening, polyphenol

Introduction

Cocoa and chocolate products have long been known to contain high levels of flavan-3-ol (flavanol) polyphenolic compounds, which have significant antioxidant activity (Adam and others 1931; Knapp and Hearne 1939; Forsyth 1955). Recent research has indicated that the flavanols found in cocoa and chocolate products are associated with short- and long-term health benefits including reduced oxidation of LDL cholesterol, reduced platelet aggregation, increased arterial blood flow, and decreased blood pressure (Ariefdjohan and Savaiano 2005; Engler and Engler 2006). Epidemiological data show a 50% lower rate of death due to cardiovascular disease and stroke in older men who consumed the highest amounts of chocolate and/or cocoa compared to those with the lowest tertile of cocoa consumption (Buijsse and others 2006). A recent meta-analysis of 133 clinical studies on wine, black and green tea, soy products, and cocoa/chocolate indicates that while green tea and black tea are beneficial for LDL-cholesterol lowering and long-term arterial flow mediated dilation (FMD), only chocolate and cocoa contributed to a significant reduction in blood pressure and acute and long-term improvements in FMD (Hooper and others 2005). Further, the treatment of cocoa powder with alkali reduces the content of flavanols and procyanidins (Miller and others 2008). However, little is known about the effects of subsequent processing such as boiling, baking, and cooking of cocoa-containing foods.

This study measured the antioxidant activity, total polyphenols (TP), the flavanol monomers ([-]epicatechin and [+]-catechin), and their oligomers also known as procyanidins in typical cocoa-containing recipes and in several commercial cake mixes.

Materials and Methods

Preparation of cocoa-containing recipes

Recipes using natural cocoa powder were selected from published cookbooks to include a chocolate fudge frosting (Anonymous 1965), hot cocoa beverage made with milk (Hurry-up...
Cocoa flavanols after baking . . .

Hot Cocoa, Anonymous (1979), chocolate cookies (Rich Cocoa Crinkle Cookies, Anonymous 2007), and chocolate cake (Collector’s Cocoa Cake, Anonymous 1987). Recipes were followed as published with regard to preparation, including time and temperature for baking. All ingredients were added by weight. A single lot of Hershey’s Natural Cocoa (The Hershey Co., Hershey, Pa., U.S.A.) was used in the recipes previously mentioned. All recipes were prepared and analyzed in triplicate.

Effect of leavening agents

Using the “Collector’s Cocoa Cake” recipe, the effect of leavening agents was further investigated by (1) substituting baking powder (Calumet Double Acting baking powder, Kraft Foods North America, Tarrytown, N.Y., U.S.A.) for baking soda (Arm and Hammer, Princeton, N.J., U.S.A.), (2) varying the proportions of baking soda and/or baking powder, and (3) testing 2 different commercial baking powders. The 2 commercial baking powders tested were Calumet Double Acting baking powder (baking soda, cornstarch, sodium aluminum sulfate, calcium sulfate, and monocalcium phosphate; Kraft Foods North America) and Rumford baking powder (monocalcium phosphate, baking soda, cornstarch; Claber Girl Corp., Terre Haute, Ind., U.S.A.). In addition, chocolate cakes were made using 2 commercially available cake mixes purchased locally (Duncan Hines Moist Deluxe Devil’s Food Premium Cake Mix, Pinnacle Foods Corp., Cherry Hill, NJ, USA 08002; Pillsbury Devil’s Food Moist Supreme Premium Cake Mix, distributed by J.M. Smucker Co., Orrville, Ohio, U.S.A.). All cakes were prepared and tested in triplicate.

Sample analysis

Antioxidant activity was measured using oxygen radical absorbance capacity (ORAC), a widely used fluorescent method for assessing antioxidant capacity in biological samples. The current method allows for the determination of lipophilic and hydrophilic antioxidant capacities and is based on the inhibition of a peroxo-radical induced oxidation initiated by the thermal base decomposition of azo compounds like 2,2′-azobis-2-methylpropanimidamide (AAPH) using fluorescein as a fluorescent probe and Trolox as a standard substrate (Ou and others 2001; Huang and others 2002). Samples (0.5 g) were extracted with 20 mL of 1:1 acetone/water containing 0.5% acetic acid by sonication at 50 °C for 10 min. The ORAC assay was conducted on a BioTek Synergy HT fluorescence plate reader using excitation and emission filters of 485 and 528 nm, respectively. ORAC values were expressed in micromoles of Trolox equivalents (TE) per gram of product.

The total polyphenol (TP) colorimetric assay initially was developed as a method for the measurement of proteins based on the reagent’s ability to react with hydroxyl constituents and later adapted by Singleton and Rossi (1965) to measure phenolic compounds in wine. It is a widely used measure of reducing capacity. Gallic acid (Sigma Chemical Co., St. Louis, Mo., U.S.A.) was used as the standard. Samples were defatted with hexane, and the defatted material (1 g) was then extracted with 10 mL of 70:29:5.0:5 acetone/water/acetic acid. The assay was conducted with 1 mL of sample extracts or gallic acid standard in 15 mL of water and acetone/water/acetic acid. The assay was conducted using 1 mL of distilled water and filtered through a 45 μm film for HPLC injection. Chromatographic separation was performed on an Agilent 1100 HPLC system using a Phenomenex Luna Phenyl Hexyl column (250 × 4.6 mm, 5 μm) and a phenyl hexyl guard column. The mobile phase consisted of (A) water/acetonitrile/acetic acid (89:9:2, v/v/v) and (B) acetonitrile/water (80:20, v/v). The gradient was as follows: 0% B from 0 to 10 minutes; 0% to 40% B from 10 to 25 minutes; 40% to 100% B from 25 to 32 minutes; 100% B from 32 to 35 minutes at a flow rate of 1.0 mL/min. The column was equilibrated for 10 minutes before the next injection. Column temperature was set at 37 °C. (+)-Catechin and (−)-epicatechin standards were purchased from Sigma Chemical Co. (+)-Catechin and (−)-epicatechin eluted at 10.2 and 16 minutes, respectively, and were monitored using a UV detector at 280 nm.

Procyanidins were measured by an HPLC method based on the separation of the flavanol oligomers from N = 1 to 10 (Gu and others 2004). Samples were defatted with hexane, and the defatted material (1 to 2 g) extracted with 5 mL of 70:29:5.0:5 acetone/water/acetic acid using sonication at 37 °C for 10 minutes. Sample extracts were clarified by centrifugation, filtered, and analyzed by HPLC using fluorescence detection with 276 nm excitation and 316 nm emission filters. Separation was performed on a Phenomenex Luna Silica (250 × 4.6 mm, 5 μm) column at 37 °C. A gradient consisting of solvent A (84:14:2:2 methylene chloride/methanol/water/acetic acid [v/v/v]) and solvent B (96:2:2 methanol/water/acetic acid [v/v/v]) was applied at a flow rate of 0.8 mL/min: 0% to 18% B linear from 0 to 30 minutes, 18% to 31% B linear from 30 to 45 minutes, 31% to 88% B linear from 45 to 50 minutes, 88% B isocratic from 50 to 65 minutes, followed by washing and re-equilibration of the column. Flavanols were quantified using procyanidin standards and response factors provided by Dr. Ron Prior (USDA, ARS, Arkansas Children’s Nutrition Center, Little Rock, Ark., U.S.A.). Standards for monomers through decamers were isolated from cocoa while the standard for polymers was isolated from blueberries.

The extractable pH of the products was determined by suspending 1 part of the sample in 9 parts deionized water at room temperature and measuring with an Orion pH (Orion Research Inc., Cambridge, Mass., U.S.A.) calibrated at pH 4 and 10 on the day of use. Baked cakes were cut in half vertically, and the resulting height was measured at the midpoint using a laboratory ruler calibrated in 1 mm increments. Approximately one half of each cake was broken into 1 to 2 inch pieces and 200 g transferred into a food processor. Cake samples were ground for 5 s using a serrated blade to produce a uniform crumb particle size with a free flowing consistency. The entire 200-g ground sample was transferred to an Agtron sample dish. The color was determined by a Hunter Tristimulus colorimeter, model d251 dp-9000 unit (Reston, Va., U.S.A.) equipped with a standard area 3.5-inch diameter viewing port, using the “L” scale, which measures the degree of lightness from 100 = light to 0 = black.

Recoveries of the measured chemicals were calculated by comparing the concentration in the finished products with the concentration based on the amount of cocoa used in the recipe. For cookies and cakes, amounts were corrected for moisture loss.
Cocoa flavanols after baking . . .

Statistical evaluation of data
All recipes were prepared and analyzed in triplicate. The mean ± standard deviation for each recipe was reported using standard statistical calculations for these parameters.

Results and Discussion
Cocoa-containing foods: antioxidant activity, total polyphenols, flavanols, and procyanidins
Table 1 shows the effects of preparation on the antioxidant activity, total polyphenols (TP), flavanol, and procyanidin content of cocoa-containing foods. The chocolate fudge frosting and hot cocoa beverage were prepared by combining cocoa with a heated ingredient (for example, milk or butter) and the remaining ingredients. Essentially 100% (86% to 109%) of the antioxidant activity, TP flavanol monomers, and procyanidins were retained in the chocolate frosting after preparation. The large procyanidin recoveries observed with the chocolate frosting and several other samples in this study are excessive, reflecting several issues with the procyanidin determination. Although the assay is based on HPLC, it is still a general assay for procyanidins. Due to the complex nature of this assay, it is possible that interfering peaks in the chromatograms resulted in elevated estimates of procyanidins.

The hot cocoa drink also retained high levels (92% to 156%) of these chemistries. After correction for moisture loss, recoveries in the chocolate cookies ranged from 88% for TP to 113% for antioxidant activity. Chocolate cakes showed the most dramatic loss in all measures with recoveries of 42% to 54% for ORAC and TP and 4% to 27% for the flavanol monomers and procyanidins. Flavanol monomers dropped close to the limit of detection (0.01 mg/g) in the baked cake. Results indicated that heating and baking of the frosting, cocoa drink, and cookies tested here did not result in destruction of antioxidant activity or the flavanol and procyanidin content. These results further suggested that some factor(s) in the cake baking process negatively affected the antioxidant activity, TP flavanols, and procyanidin content of the product and deserved further investigation.

Effect of temperature
Additional studies were done with the chocolate cake recipe to investigate the key factors that influence flavanol loss during baking. Since loss of polyphenols during roasting of cocoa beans has been reported (Robinson and others 1961; Wollgast and Anklam 2000; Zhu and others 2002), it was hypothesized that temperature (that is, baking) may be responsible for the losses in chemicals observed in the cookies and cake. To test this hypothesis, additional cakes were baked under 3 different conditions of time and temperature: 121 °C (250 °F) for 110 min, 149 °C (300 °F) for 60 min, 177 °C (350 °F) for 35 min. The duration of baking was determined by the time required for a toothpick inserted into the cake to come out clean. Regardless of time and baking temperature, the recoverable levels of ORAC activity, polyphenols, and monomers in the baked chocolate cakes were similar (data not shown), suggesting that oven temperature between 121 and 177 °C was not the key factor in the loss of antioxidant chemistries.

Effect of leavening agents
Review of a recent edition of the Betty Crocker Cookbook (Anonymous 1996) found that over 60% of the cakes and baked product recipes used baking soda as the sole leavening agent. The original cake recipe used in this study used baking soda as the sole leavening agent (data shown in Table 1). Starting with this recipe, modifications were made to the proportions of baking soda and baking powder ranging from 100% baking soda (in the original recipe) to 100% baking powder (Table 2). Chocolate cakes made with only baking soda had an average postbaked extractable pH of 8.81. By comparison, chocolate cakes made with only baking powder had a postcook extractable pH of 6.16. The height of the baked cakes also differed slightly with a height of 4.1 cm for the baking soda cakes and 2.9 cm for the baking powder cakes. The cakes differed visually in color from a very dark brown for the baking soda chocolate cakes (Hunter “L” value = 12.43 where the “L” scale measures the degree of lightness from 0 = black to 100 = light) to a medium brown (Hunter “L” value = 18.47) for the baking powder cakes.

Replacement of baking soda with increasing amounts of baking powder led to changes in the final baked pH, color, and the measured antioxidant and flavanol-related chemistries (Table 2). Baking soda is 100% pure sodium bicarbonate—an alkaline compound, whereas baking powder is a mixture of baking soda and various acidic ingredients. More specifically, the pH of the baked cakes.

Table 1 – Comparative recovery of antioxidant activity, total polyphenols, and procyanidins in homemade cocoa-containing products.*

<table>
<thead>
<tr>
<th>Sample</th>
<th>Moisture loss (%)</th>
<th>pH</th>
<th>ORAC (micromole TE/g)</th>
<th>Total polyphenols (mg/g)</th>
<th>Flavanol monomers</th>
<th>Procyanidins N = 1 to 10 (mg/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chocolate frosting</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Calculated</td>
<td>–</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Prepared</td>
<td>0</td>
<td>6.2</td>
<td>73</td>
<td>5.90</td>
<td>0.12</td>
<td>0.31</td>
</tr>
<tr>
<td>Recovery, %</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hot cocoa drink</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Calculated</td>
<td>–</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Prepared</td>
<td>0</td>
<td>6.4</td>
<td>40</td>
<td>3.26</td>
<td>0.06</td>
<td>0.17</td>
</tr>
<tr>
<td>Recovery, %</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chocolate cookie</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baked</td>
<td>4.2 ± 1.6</td>
<td>6.4</td>
<td>56</td>
<td>4.54</td>
<td>0.09</td>
<td>0.24</td>
</tr>
<tr>
<td>Recovery, %</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chocolate cake</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Calculated</td>
<td>8.7 ± 1.0</td>
<td>8.8</td>
<td>42</td>
<td>3.41</td>
<td>0.07</td>
<td>0.18</td>
</tr>
<tr>
<td>Baked</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Prepared and baked values are the means (± standard deviation) of triplicates.

**Recoveries for chocolate cookie and chocolate cake were adjusted to account for moisture loss during the baking process.

***Total polyphenols expressed as milligram of gallic acid equivalents (GAE).
Cocoa flavanols after baking . . .

Chocolate cakes changed in an orderly way as the ratio of baking powder to baking soda was changed. Color of the baked cakes as measured by Hunter “L” values was inversely linearly related to pH (Figure 1A; $R^2 = 0.9546$). Antioxidant activity (ORAC) and TP also showed inverse linear relationships to cake pH (Figure 1B; $R^2 = 0.9936$; Figure 1C; $R^2 = 0.7691$, respectively). Closer examination of the procyanidin content showed that the high pH generated by baking soda resulted in substantial loss of the flavanol monomers (Figure 2A; $R^2 = 0.9339$). The monomers and smaller procyanidin oligomers are the most readily absorbed and bioavailable flavanols (Scalbert and Williamson 2000; Manach and others 2004), reaching peak levels in the blood by 2 h after consumption (Richelle and others 1999). A similar pattern was observed with total procyanidins (Figure 2B; $R^2 = 0.9876$). Overall, the higher pH generated by baking soda resulted in a 71% to 94% decrease in the flavanol monomers and a 84% decrease in the procyanidins in contrast to essentially

### Table 2—Comparison of homemade chocolate cakes baked with baking soda (BS), baking powder (BP), and mixtures of baking powder/baking soda.

<table>
<thead>
<tr>
<th>Leavening</th>
<th>Baked pH</th>
<th>Height (cm)</th>
<th>Hunter “L” color</th>
<th>ORAC (micromole TE/g)</th>
<th>Total polyphenols (mg/g)</th>
<th>Flavanol monomers</th>
<th>Procyanidins $N = 1–10$ (mg/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calculated</td>
<td>8.81 ± 0.13</td>
<td>4.1 ± 0.0</td>
<td>12.43 ± 0.67</td>
<td>42</td>
<td>3.4</td>
<td>0.07</td>
<td>0.18</td>
</tr>
<tr>
<td>100% BS</td>
<td>8.09 ± 0.12</td>
<td>4.1 ± 0.0</td>
<td>13.37 ± 0.25</td>
<td>28 ± 1</td>
<td>1.76 ± 0.09</td>
<td>0.05 ± 0.00</td>
<td>0.02 ± 0.00</td>
</tr>
<tr>
<td>Recovery, %</td>
<td>67</td>
<td>52</td>
<td>12</td>
<td>49</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1/4 BP: 3/4 BS</td>
<td>7.44 ± 0.05</td>
<td>4.1 ± 0.0</td>
<td>14.37 ± 0.06</td>
<td>34 ± 3</td>
<td>2.54 ± 0.06</td>
<td>0.11 ± 0.01</td>
<td>0.08 ± 0.01</td>
</tr>
<tr>
<td>Recovery, %</td>
<td>81</td>
<td>75</td>
<td>171</td>
<td>43</td>
<td>64</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1/2 BP: 1/2 BS</td>
<td>6.86 ± 0.01</td>
<td>3.6 ± 0.0</td>
<td>16.17 ± 0.21</td>
<td>38 ± 2</td>
<td>2.41 ± 0.04</td>
<td>0.12 ± 0.01</td>
<td>0.16 ± 0.02</td>
</tr>
<tr>
<td>Recovery, %</td>
<td>89</td>
<td>71</td>
<td>176</td>
<td>86</td>
<td>79</td>
<td></td>
<td></td>
</tr>
<tr>
<td>100% BP</td>
<td>6.16 ± 0.03</td>
<td>2.9 ± 0.0</td>
<td>18.47 ± 0.23</td>
<td>42 ± 2</td>
<td>2.48 ± 0.03</td>
<td>0.09 ± 0.00</td>
<td>0.20 ± 0.00</td>
</tr>
<tr>
<td>Recovery, %</td>
<td>99</td>
<td>73</td>
<td>130</td>
<td>111</td>
<td>109</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Values are the means (± standard deviation) of triplicates.

*Ratio of baking soda (BS) to baking powder (BP) used in recipe.
no loss in flavanol monomers or procyanidins with the lower pH generated by baking powder. Some of the cakes made with baking powder or mixtures of baking powder and baking soda indicated large recoveries of flavanol monomers. There are several possible factors that could account for these values. The samples being analyzed are at the limit of detection of the assay, therefore, small changes can make large differences when peaks are integrated at this level. It is also well known in both model and food-based systems that the monomeric flavanols undergo epimerization-causing catechins to become epicatechins and vice-versa (Wang and Hellier-well 2000). The rate of this conversion is dependent on temperature and is not unexpected in this study with the final result being the possibility of elevated recoveries in selected compounds.

Finally, the cake recipe was prepared using 2 different baking powders for comparison with the original baking soda recipe and with chocolate cakes made from commercial mixes (Table 3). Use of 2 different commercial baking powders, A and B, resulted in cakes with similar physical and chemical characteristics for baked pH (6.16 ± 0.65), color (18.47 to 20.27 Hunter “L” color), ORAC antioxidant activity (42 to 45 μmol TE/g), TP (2.48 to 3.26 mg/g), flavanol monomers (0.29 to 0.33 mg/g), and procyanidins (0.22 to 0.87 mg/g). Homemade chocolate cakes baked with baking soda had a much higher baked pH of 8.81, the largest rise in height (4.1 cm), the darkest color, but also much lower levels overall of the antioxidant-related chemistries. When compared to the cakes made with baking powder, antioxidant activity decreased 45% to 49%. Similarly, TP decreased 42% to 56%, flavanol monomers 90% to 91%, and total flavanols 61% to 85%.

Preparation of 2 commercially available cake mixes resulted in final pHs at or above 8.35 and with low antioxidant chemistry content similar to the homemade chocolate cakes leavened with baking soda (Table 3). The low antioxidant activity, TP and flavanol monomers in the cake mixes could be a result of several factors including use of Dutch-processed cocoa powder instead of natural cocoa powder as well as choice of leavening ingredients. These results suggest that representative commercial cake mixes are poor sources of cocoa flavanols.

Flavanol loss associated with use of baking soda

This is the 1st study focusing on the fate of antioxidant activity, TP, flavanol monomers, and procyanidins in heated or baked cocoa-containing products. The results show that the choice of leavening agent and the resultant effect on pH during baking is a key factor in the subsequent levels of antioxidant activity, total polyphenols, and flavanol compounds. Baked cake pH in excess of about 7.5 resulted in progressive loss of cocoa flavanol monomers as pH increased with almost complete loss at higher pH accompanied by loss of the procyanidin oligomers. Over 60% of home cookbook recipes contain baking soda as the sole leavening agent as do many, but not all, commercial cake mixes surveyed at local grocery stores. The original cake recipe tested in the current study also used 100% baking soda as the leavening

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<table>
<thead>
<tr>
<th>Sample/leavening</th>
<th>Baked pH (cm)</th>
<th>Height (cm)</th>
<th>Hunter “L” color</th>
<th>ORAC (µmol TE/g)</th>
<th>Total polyphenols (mg/g)</th>
<th>Flavanol monomers (mg/g)</th>
<th>Procyanidins (mg/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Homemade cake with baking powder</td>
<td>6.16 ± 0.03</td>
<td>2.9 ± 0.0</td>
<td>18.47 ± 0.23</td>
<td>42 ± 2</td>
<td>2.48 ± 0.03</td>
<td>0.09 ± 0.00</td>
<td>0.87 ± 0.11</td>
</tr>
<tr>
<td>A</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Homemade cake with baking powder</td>
<td>6.69 ± 0.03</td>
<td>3.4 ± 0.1</td>
<td>20.27 ± 0.42</td>
<td>45 ± 1</td>
<td>3.26 ± 0.03</td>
<td>0.11 ± 0.01</td>
<td>0.33 ± 0.02</td>
</tr>
<tr>
<td>B</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Homemade cake with baking soda</td>
<td>8.81 ± 0.13</td>
<td>4.1 ± 0.0</td>
<td>12.43 ± 0.67</td>
<td>23 ± 4</td>
<td>1.44 ± 0.03</td>
<td>0.02 ± 0.00</td>
<td>0.13 ± 0.05</td>
</tr>
<tr>
<td>Commercial cake mix A</td>
<td>8.35 ± 0.06</td>
<td>4.2 ± 0.1</td>
<td>12.37 ± 0.50</td>
<td>21 ± 4</td>
<td>1.00 ± 0.03</td>
<td>0.00 ± 0.00</td>
<td>0.19 ± 0.02</td>
</tr>
<tr>
<td>Commercial cake mix B</td>
<td>8.81 ± 0.08</td>
<td>4.3 ± 0.2</td>
<td>11.30 ± 0.17</td>
<td>9 ± 1</td>
<td>0.85 ± 0.03</td>
<td>0.00 ± 0.00</td>
<td>0.76 ± 0.07</td>
</tr>
</tbody>
</table>

*Values are the means (± standard deviation) of triplicates.

Figure 2—Relationship between pH and flavanol monomers (A) and procyanidins (B) in homemade baked chocolate cakes.
agent. The results of the our study demonstrate that there is significant loss of flavanols, especially the low molecular weight flavanols in the majority of cocoa-containing cakes and baked goods commercially available or made at home. The results indicate that the highest losses occur at pHs above 7.5 and that these losses are caused by the use of baking soda as the only leavening agent. However, the results also indicate that more of the flavanol antioxidant chemistries can be preserved using a combination of leavening agents to moderate pH changes while still achieving an acceptable leavening action. Common acidic ingredients, sometimes referred to as leavening acids, include cream of tartar, tartaric acid, monocalcium phosphate monohydrate, anhydrous monocalcium phosphate, sodium acid pyrophosphate, sodium aluminum phosphate, dicalcium phosphate dehydrate, sodium aluminum sulfate, and glucono delta lactone, which have different neutralizing strengths and times of action (Dubois 1981). Each can be used to moderate cake pH to neutral or even slightly acidic conditions (Table 2, Figure 1). Other ingredients such as buttermilk, fruits, fruit purees, or juices can be added to provide flavor and texture to the cake but may also serve to keep the cake recipe more acidic. In studies not shown here, we have found that acidic ingredients like applesauce will minimize increases in final baked cake pH and thus preserve more of the flavanol content in the baked cake. These results suggest that the effect of increased pH due to baking soda can be largely reversed by choosing the appropriate leavening acid in the form of a baking powder or by adding acidic ingredients to the recipe. The final pH of the baked cake needs to be pH 7.5 or less for preservation of antioxidant activity and flavanol content.

**Conclusions**

Beyond the loss of flavanols from cocoa-flavored foods, other flavanol-rich ingredients such as grapes, raisins, cranberries, blueberries, apples, and other fruits, and spices, such as cinnamon may suffer significant loss in their flavanol content in cakes and baked goods with high final pH. It is important to consider the impact of acidulants or basic leavening agents on the final pH and the impact these ingredients may have on the naturally occurring and healthful flavanol content.

**References**


