

# Effects of Processing and Usage Level on Performance of Bovine Plasma as an Egg White Substitute in Cakes<sup>1</sup>

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## ABSTRACT

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The effects of processing and level used on the performance of bovine plasma replacing egg whites in cakes were investigated. Plasma was evaluated in high-ratio, white layer cakes in different forms (fresh, frozen, and spray-dried) at five levels (50, 75, 90, 100, and 110% of the normal protein level when using egg whites). An enzyme-hydrolyzed 90/10 blend of plasma and beef stock was also evaluated. Cakes made with plasma had slightly less volume, less crowning profile, and darker crust color

than cakes made with equivalent amounts of egg white protein. Also, those cakes were softer, less chewy, and less springy but were similar in cohesiveness and gumminess to those made with egg whites. Nearly equivalent performance was achieved by replacing 1.0 parts of egg whites with 1.1 parts of plasma. There were few differences in performance among fresh, frozen, and spray-dried plasma. Hydrolyzed plasma-beef stock blends gave cakes with higher volumes but darker crust colors than plasma.

Low-cost egg substitutes have long been sought by the baking industry because egg products are often the most expensive ingredients. Egg proteins, especially egg albumin (whites), are essential for desirable volume and texture in baked goods because of their unique solubility, foaming, emulsifying, and heat coagulation properties (Pylar 1973). Bovine plasma, a by-product of the animal slaughtering industry, is the only other native protein known to have properties similar to those of egg whites (Tybor 1973, Tybor et al 1975). Enzymatically modified plant proteins, often promoted as good egg substitutes, have similar foaming properties but different heat coagulation characteristics (Eldridge et al 1963).

Spray-dried plasma protein concentrates are now commercially available in food-grade forms that currently sell for only about one-third the cost of spray-dried egg whites. The fractionation of bovine blood into food ingredients is well established (Pals 1970, Gordon 1971, Halliday 1973). Blood is collected in a sanitary manner, quickly mixed with anticoagulant solution (isotonic mixture of sodium chloride and sodium citrate), chilled, and held until the animals pass U.S. Department of Agriculture inspection. The stabilized blood is centrifuged to separate red blood cells from plasma.

As early as World War I, plasma was used in Germany for protein fortification of bread (Kobert 1915) and during World War II was used as an egg substitute in mayonnaise, soups, and sauces (Shenstone 1953). Brooks and Ratcliff (1959) reported that blood plasma could replace 33% of the whole egg in European low-ratio cakes. Johnson et al (1979) demonstrated that fresh lyophilized plasma can replace egg whites in high-ratio cakes, but, upon storage, the baking potential of plasma deteriorates. Khan et al (1979) also reported that bovine plasma protein isolate can replace up to 25% of the egg white protein in angel food cakes.

Recently, improvements in the processing and quality of plasma have been made. Microbiological contamination and the level of anticoagulation salt have been reduced. The color has also been improved. To extend shelf life and facilitate marketing, liquid plasma is now being spray-dried or frozen. In recent years, bovine plasma has been processed on a commercial scale, and substantial quantities of food-grade plasma are now used as a binder in

comminuted meats and for flavoring. The objectives of this study were to evaluate plasma processed on commercial scale by these improved technologies and different forms of preservation as egg white replacers in cakes.

## MATERIALS AND METHODS

### Cake Ingredients

Fresh liquid plasma, spray-dried plasma, and a spray-dried enzymatically hydrolyzed blend of 90% plasma and 10% beef stock were obtained from the American Meat Protein Corp., Ames, IA. Half of the fresh liquid plasma was frozen, and the other half was used fresh. All frozen samples were stored at  $-29^{\circ}\text{C}$  until used. Fresh plasma was stored at  $5^{\circ}\text{C}$  and used within 48 hr after collection.

Fresh eggs were obtained from a local grocery. The whites were separated and used fresh. Frozen and spray-dried egg whites (type p-20) were obtained from Henningsen Foods, Inc., Omaha, NE. Betrkake emulsified shortening (monoglycerides and diglycerides) was obtained from Durkee Industrial Foods Corp., Cleveland, OH. Softasilk cake flour, a cake flour for institutional food service use, was obtained from Iowa State University's Food Service Center and produced by General Mills, Inc., Minneapolis, MN. Breadlac nonfat dry milk solids were obtained from Galloway West Co., Fond du Lac, WI.

Protein contents were determined by macro-Kjeldahl method, using 6.25 g of protein per gram of nitrogen as the conversion factor and AACC method 46-13 (AACC 1983). Moisture was determined by AACC method 44-31, oven drying at  $130^{\circ}\text{C}$  for 2 hr (AACC 1983).

### Cake Baking Procedure

Cakes were prepared by modifying AACC method 10-90 (AACC 1983) to accommodate equivalent amounts of protein from the various forms of plasma and egg whites because this method specifies use of spray-dried egg whites. Water additions in first- or second-stage mixing, or both, were adjusted to account for different moisture contents of the various egg white and plasma products. The amounts of each cake ingredient used in the formula were as follows: 100 g of cake flour; 140 g of sugar; 50 g of shortening; 12 g of nonfat dry milk; 5 g of baking powder; 3 g of salt; 9 g of dried egg whites; and 87, 29, and 29 g of water for mixing stages 1, 2, and 3, respectively.

Fresh, frozen, and spray-dried plasma and fresh, frozen, and spray-dried egg whites were evaluated at different levels (50, 75, 90, 100, and 110% of the normal level of egg white protein). Precisely, 425 g of batter was scaled into each of two 8-in. cake pans and baked at  $190^{\circ}\text{C}$  ( $375^{\circ}\text{F}$ ) for 25 min in a rotary electric oven. Specific gravity of the batter was determined by weighing a known volume of batter. Baking trials were replicated three times. Cakes were cooled for 60 min, stored in plastic bags overnight, and evaluated the next day.

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### Cake Evaluation

Cake quality was evaluated by AACC method 10-91 (AACC 1983). Cake volume was evaluated by rapeseed displacement. Cake profile was evaluated by using the AACC cake profile template.

A Hunterlab Labscan Spectro colorimeter, L-5-5100 color difference meter, was used to measure color of crust and crumb samples. The instrument was standardized by using a white tile ( $X = 81.60$ ,  $Y = 86.68$ ,  $Z = 91.18$ ) and a 3-cm-diameter opening in the viewing aperture. Cakes were cut by using a 3.3-cm-diameter cookie cutter, and the pieces were placed into 3.5-cm-diameter petri dishes. Hunter  $L$ ,  $a$ , and  $b$  values were measured on duplicate samples from each cake. In the Hunter color system, the  $L$  value measures intensity of the color (lightness) with an  $L$  value of 0 indicating black and an  $L$  value of 100 indicating white. A positive  $a$  value indicates red and a negative value indicates green. A positive  $b$  value indicates yellow and a negative value indicates blue. The hue angle,  $\theta$ , ( $\tan^{-1}b/a$ ) was also calculated to define the actual color for the cakes.

An Instron Universal Testing Machine (model 1122) equipped with a 500-kg compression/tension load cell was used to measure texture properties of cakes. The crosshead speed was 200 mm/min, and the chart speed was 500 mm/min. Cake texture was evaluated by using the modified texture profile analysis (Bourne 1968, 1974). Ten 20-mm cubes from the center crumb of each

cake were evaluated. Each cube was compressed to 80% of its original height in each of two consecutive compressions. The force-distance curve was used to determine firmness, springiness, cohesiveness, gumminess, and chewiness.

All data were statistically analyzed by using analysis of variance and Duncan's multiple range test. Least significant differences were calculated at the 0.05 level.

**TABLE I**  
Protein and Moisture Contents of Plasma Products and Egg Whites

Protein Source/ Form	Moisture (%)	Protein <sup>a</sup> (%)
Plasma		
Fresh	91.10	77.75
Frozen	91.10	77.75
Spray-dried	6.29	70.23
Enzyme hydrolyzed plasma + beef stock		
Spray-dried	6.29	70.19
Egg whites		
Fresh	89.02	92.35
Frozen	88.89	87.31
Spray-dried	6.28	78.43

<sup>a</sup> Conversion factor of 6.25 g of protein per gram of N; reported on moisture-free basis.

**TABLE III**  
Qualities of Cakes Made with Different Levels of Spray-Dried Bovine Plasma

Cake Quality Factor	Percent of Normal Egg White Protein <sup>a</sup>				
	50%	75%	90%	100%	110%
Batter					
Specific gravity, g/cm <sup>3</sup>	0.595 a	0.593 a	0.592 a	0.572 a	0.573 a
Size and shape					
Volume, cm <sup>3</sup>	888 c	927 b	932 b	937 b	970 a
Profile, mm	4.7 a	3.7 a	5.7 a	6.3 a	6.3 a
Color					
Crust color, $L$ value	45.28 a	41.40 c	43.42 abc	44.28 ab	41.68 bc
Crumb color					
$L$ value	71.09 a	70.17 ab	70.07 ab	70.07 ab	69.27 b
$a$ value	-1.14 b	-1.07 b	-1.00 ab	-1.04 ab	-0.92 a
$b$ value	11.12 ab	10.44 b	11.06 ab	11.06 ab	11.22 a
$\theta$ value	95.86 a	95.86 a	95.17 a	95.37 a	94.69 b
Texture					
Firmness, kg	3.52 c	3.57 c	3.85 bc	4.10 ab	4.30 a
Springiness, mm	8.50 c	9.50 bc	10.10 bc	10.93 ab	12.53 a
Cohesiveness	0.26 c	0.27 bc	0.30 ab	0.32 ab	0.32 a
Gumminess, kg	0.90 c	0.98 c	1.15 b	1.32 a	1.39 a
Chewiness, kg·mm	7.67 d	9.30 cd	11.17 bc	14.43 ab	17.47 a

<sup>a</sup> Means within a row followed by the same letter are not significantly different by Duncan's multiple range test at the 0.05 level.

**TABLE II**  
Qualities of Cakes Made with Different Levels of Spray-Dried Egg Whites

Cake Quality Factor	Percent of Normal Egg White Protein <sup>a</sup>				
	50%	75%	90%	100%	110%
Batter					
Specific gravity, g/cm <sup>3</sup>	0.595 a	0.580 a	0.573 a	0.578 a	0.575 a
Size and shape					
Volume, cm <sup>3</sup>	957 b	990 a	992 a	1,013 a	1,017 a
Profile, mm	11.7 a	12.7 a	10.7 a	13.3 a	13.3 a
Color					
Crust color, $L$ value	47.95 a	48.76 a	51.24 a	46.97 a	46.02 a
Crumb color					
$L$ value	72.97 a	72.55 a	73.25 a	73.51 a	73.46 a
$a$ value	-1.34 a	-1.38 ab	-1.43 c	-1.40 bc	-1.37 ab
$b$ value	8.73 a	8.70 a	8.84 a	8.65 a	8.68 a
$\theta$ value	98.73 a	99.02 a	99.19 a	99.19 a	98.96 a
Texture					
Firmness, kg	3.97 c	4.99 b	5.53 b	5.52 b	6.30 a
Springiness, mm	13.80 d	18.50 c	21.10 bc	22.73 ab	26.09 a
Cohesiveness	0.29 c	0.31 ab	0.33 ab	0.33 ab	0.36 a
Gumminess, kg	1.14 a	1.55 b	1.72 b	1.84 b	2.27 c
Chewiness, kg·mm	15.65 d	28.67 c	36.12 bc	42.22 b	59.38 a

<sup>a</sup> Means within a row followed by the same letter are not significantly different by Duncan's multiple range test at the 0.05 level.

## RESULTS AND DISCUSSION

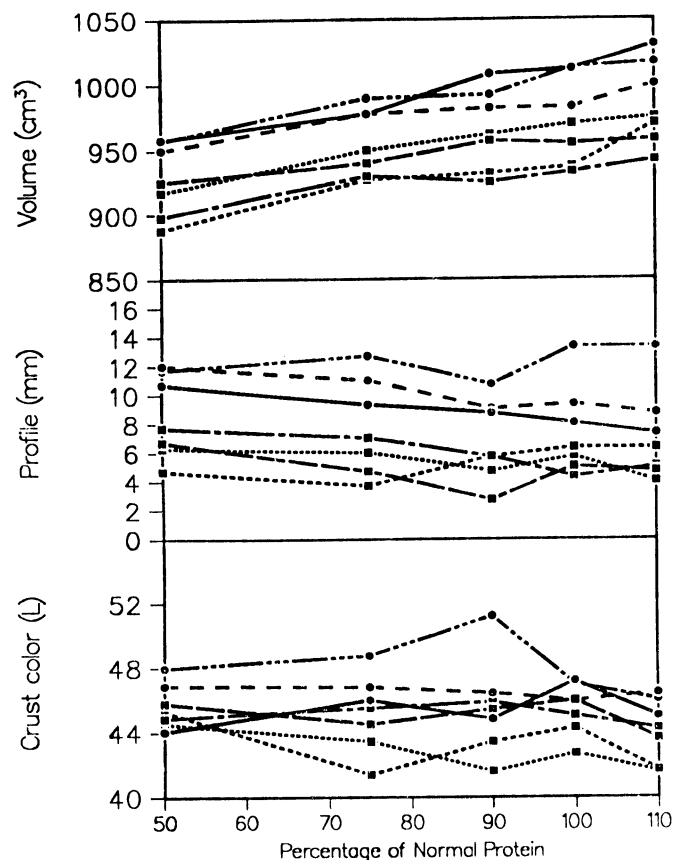
### Protein Contents of Plasma Products Compared to Egg Whites

Protein and moisture contents for the plasma and egg white products are shown in Table I. Generally, the protein contents of the plasma products were 10% less than their egg white counterparts. This is largely due to the added salt used to chelate calcium,

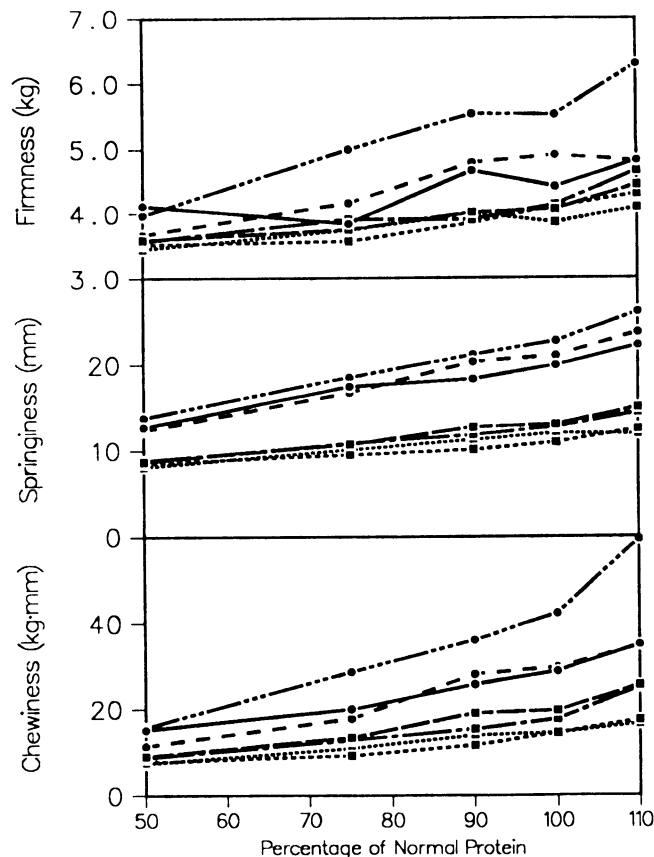
which is important in blood clotting. That protein contents of the products used in this study were higher than those observed by Johnson et al (1979) reflects the lower salt concentrations now used to prevent coagulation.

### Effect of Level of Spray-Dried Egg Whites

Cakes containing 100 and 110% of the normal egg white level



**Fig. 1.** Effects of different forms and levels of bovine plasma and egg whites on volumes, profiles, and crust colors of high-ratio, white layer cakes. Fresh egg whites (●—●); frozen egg whites (●—●); spray-dried egg whites (○—○); fresh plasma (■—■); frozen plasma (■—■); spray-dried plasma (□—□); and spray-dried, enzymatically hydrolyzed plasma-beefstock blend (■—■). The least significant differences (0.05 level) were 9.4 cm<sup>3</sup> for volume, 0.8 mm for profile, and 1.04 for crust color.



**Fig. 2.** Effects of different forms and levels of bovine plasma and egg whites on crumb firmness, springiness, and chewiness of high-ratio, white layer cakes. Fresh egg whites (●—●); frozen egg whites (●—●); spray-dried egg whites (○—○); fresh plasma (■—■); frozen plasma (■—■); spray-dried plasma (□—□); and spray-dried, enzymatically hydrolyzed plasma-beefstock blend (■—■). The least significant differences (0.05 level) were 0.17 kg for firmness, 0.67 mm for springiness, and 3.3 kg·mm for chewiness.

**TABLE IV**  
Qualities of Cakes Made with Different Types of Egg Whites and Bovine Plasma at the Normal Egg White Protein Level

Cake Quality Factor	Bovine Plasma <sup>a</sup>				Egg White <sup>a</sup>		
	Fresh	Frozen	Dry	Hydrolyzed	Fresh	Frozen	Dry
<b>Batter</b>							
Specific gravity, g/cm <sup>3</sup>	0.588 ab	0.602 a	0.578 ab	0.572 b	0.593 a	0.595 a	0.572 b
<b>Size and shape</b>							
Volume, cm <sup>3</sup>	933 b	933 b	937 b	970 ab	1,012 a	983 ab	1,013 a
Profile, mm	5.0 b	4.3 b	6.3 ab	5.7 ab	8.0 ab	9.3 ab	13.3 a
<b>Color</b>							
Crust color, L value	45.98 a	45.04 a	44.28 a	42.70 a	47.17 a	45.95 a	46.97 a
<b>Crumb color</b>							
L value	69.24 cd	68.97 d	70.07 c	69.78 cd	72.98 ab	72.35 b	73.51 a
a value	-1.10 a	-1.06 a	-1.04 a	-0.97 a	-1.53 b	-1.54 b	-1.40 b
b value	10.87 a	10.91 a	10.83 a	11.06 a	9.01 b	8.99 b	8.65 b
θ value	95.78 b	95.55 b	95.49 b	95.01 b	99.64 a	99.72 a	99.19 a
<b>Texture</b>							
Firmness, kg	4.06 c	4.13 c	4.10 c	3.86 c	4.41 bc	4.90 b	5.52 a
Springiness, mm	12.97 b	12.70 b	10.93 b	12.00 b	19.87 a	21.03 a	22.73 a
Cohesiveness	0.37 a	0.34 ab	0.32 ab	0.31 b	0.33 ab	0.29 b	0.33 ab
Gumminess, kg	1.53 b	1.39 b	1.32 b	1.20 b	1.42 b	1.42 b	1.84 a
Chewiness, kg·mm	19.59 c	17.62 c	14.43 c	14.48 c	28.73 b	29.63 b	42.21 a

<sup>a</sup> Means within a row followed by the same letter are not significantly different by Duncan's multiple range test at the 0.05 level.



produced cakes with volumes in excess of 1,000 cm<sup>3</sup>, which we considered our standard for cakes made with flour sold to institutional baking operations (Table II). This standard cake volume is slightly smaller than an appropriate standard for cakes normally made from good commercial cake flours used by wholesale baking operations (approximately 1,050 cm<sup>3</sup>). Further studies are planned to compare performances using a commercial flour specially made for wholesale baking operations. As egg white level decreased, so did the volume. At 50% of the normal egg white level, the resulting cakes were denser than those produced with more egg whites. Reducing the egg white level from 100 to 50% reduced cake volume by about 6%. As egg white level decreased, specific gravity increased towards higher values, but differences were not significant at the 0.05 level.

The profiles of the cakes were highly crowned at all egg white levels, but crowning declined slightly as the level of egg whites decreased. Crumb and crust colors were little affected by the level of spray-dried egg whites. Texture was affected by level of spray-dried egg whites; cakes became less tender as the level of egg whites increased. According to the universal testing machine assessments, cakes became more firm, springy, cohesive, gummy, and chewy as egg white level increased.

The thermal denaturation of egg albumin acts as a water sink and binder between starch granules and other cake ingredients. Thus, along with the gelatinization of starch, protein denaturation

acts as a thermally triggered "setting" of cake structure. Simultaneous gelatinization of starch and denaturation of egg whites at about the same temperature as maximum expansion due to chemical leavening is thought to be responsible for achieving desirable volume and texture (Mizukoshi et al 1979, 1980; Mizukoshi 1985).

#### Effect of Eliminating Egg Whites

Volumes of cakes made without egg whites or plasma could not be measured because of extreme fragility but were estimated to be less than 600 cm<sup>3</sup>. Cakes fell in the middle, which resulted in negative values for profile crown.

#### Effect of Level of Spray-Dried Plasma

Cakes made with spray-dried plasma were slightly lower in volume, but similar in firmness, cohesiveness, and color to those made with egg white protein. They had 5–7% less volume than those made with equivalent amounts of egg white protein (Tables II and III). As with egg white levels, cake volumes declined as the level of spray-dried plasma declined (Table III). Cake volume declined by about 5% as the level of spray-dried plasma was reduced from 100 to 50%. The level of plasma could only be reduced to 75% of the normal egg white protein level, and volumes were not significantly different from cakes containing 50% egg whites. As with egg whites, the specific gravities of the batters



Fig. 3. Cakes made with fresh, frozen, and spray-dried egg whites (top) and bovine plasma (bottom) at a normal protein level.



increased towards higher values as the level of plasma was reduced, but differences were not significant at the 0.05 level.

Profiles of cakes made with spray-dried plasma were 30–50% less crowned than were cakes made with egg whites (Table III). Cakes with less crowning may be desirable because flatter cakes stack better and have more uniform crumb structure if volume is adequate.

Crumb colors of cakes made with plasma were largely unaffected by level of plasma, as indicated by the values of hue angle  $\theta$ , except that cakes containing 110% of the normal egg white level were slightly, but significantly, more yellow (Table III). Redness of crumb color was used for comparing negative  $a$  values because greenness seemed not to be an appropriate descriptor in a cake system. The crumb of cakes made with plasma was slightly darker, redder, and more yellow than the crumb of cakes prepared with egg whites as determined by colorimeter measurement.

As with egg whites, the level of spray-dried plasma significantly affected the texture of the cakes (Table III). All values for the five texture parameters declined as the level of plasma decreased.

#### Comparison of Cakes Made with Plasma or Egg Whites

Cakes made with all forms of native plasma at 100% egg white level or less were significantly smaller (about 7%) in volume and less crowned than those made with egg whites (Table IV and Fig. 1). The least significant differences (LSD) were 9.4 cm<sup>3</sup> for volume and 0.8 mm for profile at the 0.05 level of significance. However, volumes of cakes made with 1.1 part of plasma were similar to cakes with 1.0 part of egg white protein.

Crust colors of all plasma cakes were equivalent to those of cakes made with egg whites for all protein levels (Table IV and Fig. 1). The LSD was 1.04 for crust  $L$  values at the 0.05 level. Crumb colors of cakes made with plasma were slightly but significantly darker (lower  $L$  values) than those of cakes made with egg whites for all protein levels (Table IV). The values of hue angle  $\theta$  of cake crumb color were also lower, indicating that cakes made with plasma had lighter yellow crumbs (slightly to the green side) than the cakes made with egg whites.

Generally, cakes made with plasma were less firm, less springy, and less chewy than those made with egg whites for all protein levels (Table IV and Fig. 2). The LSDs at the 0.05 level were 0.17 kg, 0.67 mm, and 3.3 kg·m for firmness, springiness, and chewiness of cakes, respectively. Cakes made with plasma were equivalent to those made with egg whites in gumminess and cohesiveness (Table IV).

#### Effect of Freezing and Spray-Drying

Little effect on cake-baking potential due to method of preservation was observed. At the normal egg white protein level and also other levels, freezing and spray-drying did not affect the volumes, profiles, or crust colors of cakes made with either egg white or plasma (Table IV, Figs. 1 and 3). Crumb colors were slightly but significantly darker when the spray-dried form was used (Table IV and Fig. 3). Cakes made with spray-dried egg whites were firmest, springiest, and most chewy of all the cakes made (Fig. 2).

#### Effects of Enzymatic Hydrolysis on Performance of Spray-Dried Plasma

A blend of 90% plasma and 10% beef stock that was hydrolyzed by treating with trypsin and then spray-dried was also tested for cake-baking potential. This product is classified as a flavoring by the Food and Drug Administration and, when used for flavoring purposes, can be so labeled on ingredient statements. Cakes made with the hydrolyzed blend had higher volumes than cakes made with native plasma at the normal egg white protein level and at other levels—nearly 96% of the volume of cakes made with spray-dried egg whites (Table IV and Fig. 1). The crust colors of cakes made with the enzymatically hydrolyzed plasma-beef stock blend tended to be darker than those of other cakes, but these differences were not significant at the 0.05 level. How-

ever, the darker crust color was very uniform across the surface. Crumb colors of cakes made with the hydrolyzed blend were similar to those of cakes made with fresh and spray-dried plasma (Table IV). Texture values were not significantly different in cakes prepared with the enzymatically hydrolyzed plasma-beef stock blend from those made with other forms of plasma (Table IV and Fig. 2).

#### CONCLUSIONS

The use of fresh, frozen, and spray-dried plasma to replace similar forms of egg whites in high-ratio, white layer cakes resulted in cakes with similar volume, texture, and appearance. Nearly equivalent cake quality was achieved when replacing 1.0 part of egg white protein with 1.1 part of plasma protein. At equivalent protein levels, cakes made with plasma were softer, less springy, and less chewy. The replacement of egg whites with a spray-dried, enzyme-hydrolyzed blend of 90% plasma and 10% beef stock increased volume, darkened crust, and further softened texture compared with cakes made with spray-dried native plasma. Almost no significant differences in cake baking performance were noted among any of the fresh, frozen, and spray-dried products for either plasma or egg whites, except that crumb colors were slightly darker when spray-dried forms were used. Thus, we found that plasma preparations were suitable replacements for egg whites in high-ratio, white layer cakes. An extensive sensory study on the replacement of egg whites with plasma products was carried out and will be reported in a subsequent paper.

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