

These residues contained about 100 mg/lb of carotene and 250 mg/lb of xanthophyll and are high in nitrogen. The residues from the other presses had a protein, fat, and ash content similar to that of the original alfalfa. All residues contain more fiber than is accepted for standard grades of Dehy. The maximum crude fiber in 17 and 20% protein grade Dehys is 27 and 22%, respectively. These residues could meet the standards for good quality Dehy if the nitrogen-containing brown juice from the PRO-XAN process were added back. The addition of brown juice solids would dilute the fiber without significantly lowering the protein. Even without the brown juice added back, the pressed cakes could be used as a ruminant feed. Hollo and Koch (1970) also felt that such residues would be satisfactory for ruminants.

There was almost no loss of crude protein in any of the presses. However, there were losses of carotenoids. In sugar cane rolls, there was a loss of about 10% in a single pass and a loss of about 30% during three rollings. The overall loss of carotenoids in single and double passes through the twin-screw press was 20 and 40%, respectively. A possible explanation for the larger loss of carotenoids in the twin-screw press is that increased rupture of cells would result in the release of increased amounts of enzymes, such as lipoxigenase, which induce carotenoid oxidation (Ben Aziz *et al.*, 1968).

ACKNOWLEDGMENT

The authors express their appreciation to R. H. Edwards, K. V. Smith, T. L. Greer, and E. J. Vandiver for assistance in operating the equipment; to H. M. Wright and associates for the feed analysis; to Reitz Manufacturing Co. for supplying the V-press; and to Dixon Dryer Co. for supplying the alfalfa and the vertical screw press.

LITERATURE CITED

- Aughtry, J. D., *Feedstuffs* **43**, 22 (1971).
 Ben Aziz, A., Grossman, S., Budowski, P., Ascarelli, I., Bondi, A., *J. Sci. Food Agr.* **19**, 65 (1968).
 Casselman, T. W., Green, V. E., Jr., Allen, R. J., Jr., Thomas, F. H., Technical Bulletin 694, Agricultural Experiment Station, University of Florida, August 1965.
 Chayen, I. H., Smith, R. H., Tristram, G. R., Thirkell, D., Webb, T., *J. Sci. Food Agr.* **12**, 502 (1961).
 Davys, M. N. G., Pirie, N. W., *Engineering* **190**, 274 (1960).
 Davys, M. N. G., Pirie, N. W., *J. Agr. Eng. Res.* **10**, 142 (1965).
 Davys, M. N. G., Pirie, N. W., Street, G., *Biotechnol. Bioeng.* **11**, 529 (1969).
 Halverson, A. W., *J. Agr. Food Chem.* **10**, 419 (1962).
 Hartman, G. H., Jr., Akeson, W. R., Stahmann, M. A., *J. Agr. Food Chem.* **15**, 74 (1967).
 Hollo, J., Koch, L., *Proc. Biochem.* **37** (October, 1970).
 Huang, K. H., Tao, M. C., Boulet, M., Riel, R. R., Julien, J. P., Brisson, G. J., *J. Can. Inst. Food Technol.* **4**, 85 (1971).
 Knuckles, B. E., Spencer, R. R., Lazar, M. E., Bickoff, E. M., Kohler, G. O., *J. Agr. Food Chem.* **18**, 1086 (1970).
 Knuckles, B. E., Witt, S. C., Miller, R. E., Bickoff, E. M., *J. Ass. Offic. Anal. Chem.* **54**, 769 (1971).
 McDonald, A. N. C., Hamblin, H. J., Cashmore, W. H., *Rep. 43 Natl. Inst. Agr. Eng.* **30** (1954).
 Official Method of Analysis, 11th Ed., A.O.A.C., Washington, D.C., 1970.
 Pirie, N. W., *Proc. Nutr. Soc.* **15**, 154 (1956).
 Pirie, N. W., *Science* **152**, 1701 (1966).
 Pirie, N. W., *Plant Food Human Nutr.* **1**, 237 (1969).
 Spencer, R. R., Bickoff, E. M., Kohler, G. O., Witt, S. C., Knuckles, B. E., Mottola, A., *Trans. Soc. Agr. Eng.* **13**, 198 (1970).
 Spencer, R. R., Mottola, A. C., Bickoff, E. M., Clark, J. P., Kohler, G. O., *J. Agr. Food Chem.* **19**, 504 (1971).
 Witt, S. C., Spencer, R. R., Bickoff, E. M., Kohler, G. O., *J. Agr. Food Chem.* **19**, 162 (1971).

Received for review March 3, 1972. Accepted April 21, 1972. Reference to a company or product name does not imply approval or recommendation of the product by the U.S. Department of Agriculture to the exclusion of others that may be suitable.

Composition of Commercial Wheys

Janis Cerbulis,* John H. Woychik, and M. Valerie Wondolowski

Commercial whey samples of single cheese types and of blended wheys were analyzed for their protein, carbohydrate, lipid, and ash contents which averaged 9.7, 71.7, 1.28, and 8.2%, respectively.

The amino acid compositions of the dialyzable and nondialyzable fractions of selected whey samples were compared to those of a control whey.

The 22–23 billion pounds of whey produced annually by the cheese industry pose a substantial challenge to feed and food technologists, and are a major problem in pollution control. Although approximately one-third of the whey produced is utilized in feed and food formulations, new product development is required for a greater utilization of this by-product. To facilitate this development, we have investigated the nitrogen distribution, protein, carbohydrate, and lipid contents and the amino acid compositions of the

dialyzable and nondialyzable nitrogen fractions of several commercial and laboratory wheys.

EXPERIMENTAL

Whey Samples and Fractions. Whey samples were obtained as dried solids or as pasteurized liquids which were subsequently freeze-dried. The whey samples were obtained from the following sources: sweet whey blends A and B, Kraft Foods, Chicago, Ill.; blend C, Foremost Foods Co., San Francisco, Calif.; blend D, Meinerz Creamery, Fredericksburg, Iowa; and blend E, Pollio Dairy, Campbell, N.Y.; Swiss whey, Star Valley Swiss Cheese Co., Thayne, Wyo.; 25% Cheddar–75% Swiss whey, Cache Valley Dairy Association, Smithfield, Utah; cottage whey A, Kraft Foods, Chicago, Ill.; cottage whey B, Cheddar and skim Cheddar

* Eastern Regional Research Laboratory, Eastern Marketing and Nutrition Research Division, Agricultural Research Service, U.S. Department of Agriculture, Philadelphia, Pennsylvania 19118.

Table I. Composition of Wheys

	Sweet whey blends				
	A	B	C	D	E
Total nitrogen, %	2.0	2.2	1.9	2.2	1.9
Nondialyzable nitrogen, % total N	76.0	83.7	82.2	76.2	80.5
Crude protein, % (total N × 6.38)	12.8	14.0	12.1	14.0	12.1
"True" protein, % (nondialyzable N × 6.38)	9.7	11.7	10.0	10.7	9.7
Lipids, %	1.2	1.0	1.0	0.8	1.2
Lipid nitrogen, % total N	0.7	0.4	0.5	...	0.4
Lactose, %	71.9	71.7	67.2	72.4	71.3
Ash, %	7.6	8.4	8.9	8.3	8.3
Water, %	3.0	2.9	3.1	3.2	2.3

wheys were obtained from the Dairy Products Laboratory EMNRD, USDA, Beltsville, Md. The control whey was prepared from raw skim milk by acid precipitation with 1 N HCl and the separation of curd and whey was achieved by filtration through a flannel bag. The whey fraction was freeze-dried.

Nondialyzable whey fractions were obtained by dialysis of 20 g of whey solids (dissolved in 200 ml of H₂O) against 15 l. of distilled H₂O for 4 days, with three changes of water daily. The nondialyzable fraction was freeze-dried.

Dialyzable fractions for amino acid analysis were prepared by dialyzing 10 g of whey solids against four changes of 500 ml of H₂O. The combined dialysates were adjusted to pH 1.7 with HCl and passed through a 3 × 30 cm column of Dowex 50 (H⁺) at a flow rate of 30 ml/hr. The column was washed with H₂O until free of lactose. No nitrogenous compounds were detected in the sample effluent or in the wash H₂O. Adsorbed nitrogenous compounds were eluted with 500 ml of 7% aqueous ammonia; the eluate was concentrated on a rotary evaporator and freeze-dried.

Analytical Methods. Nitrogen was determined by the standard micro-Kjeldahl method (AOAC, 1965a). A nitrogen conversion factor of 6.38 was used for calculation of total protein.

Lipids were extracted from nondialyzable solids with chloroform-methanol (2:1, v/v) as previously described (Cerbulis, 1967).

Lactose was determined by the method of Marier and Boulet (1959); ash content was determined by the official AOAC method (1965b).

Amino Acid Analyses. Dialyzable and nondialyzable nitrogenous fractions were hydrolyzed with glass-distilled 6 N HCl in sealed evacuated tubes for 24 hr at 110°. Amino acid compositions were determined from duplicate samples analyzed on an automatic amino acid analyzer. Tryptophan was determined by the colorimetric procedure of Spies and Chambers (1949).

Table II. Composition of Wheys

	Type of whey							
	Swiss	25% Cheddar		Skim milk		Cottage		Control
		75% Swiss	Cheddar	Cheddar	Cheddar	A	B	
Total nitrogen, %	2.3	2.4	1.8	1.9	2.0	2.0	1.8	
Nondialyzable nitrogen, % total N	69.0	70.5	77.2	71.5	79.0	64.4	72.2	
Crude protein, % (total N × 6.38)	14.7	15.3	11.5	12.1	12.8	12.8	11.5	
"True" protein, % (nondialyzable N × 6.38)	10.1	10.8	8.9	8.7	10.1	8.2	8.3	
Lipids, %	4.3	0.8	2.7	0.4	0.5	0.5	0.4	
Lipid nitrogen, % total N	0.7	0.3	
Lactose, %	69.2	72.5	74.4	74.6	68.2	74.3	72.4	
Ash, %	9.4	8.8	7.4	7.7	11.5	11.3	11.3	
Water, %	2.6	6.0	4.8	7.1	4.0	5.1	5.8	

Table III. Amino Acid Composition^a of Whey Fractions

Amino acid	Nondialyzable					Dialyzable				
	Swiss	Cheddar	Cottage	Blend	Control	Swiss	Cheddar	Cottage	Blend	Control
Aspartic acid	10.4	11.3	11.1	11.3	10.9	6.8	6.7	6.0	5.8	6.2
Threonine	8.0	8.7	6.3	8.0	5.7	3.5	3.8	3.3	3.3	3.6
Serine	4.4	4.1	4.0	4.2	4.0	3.0	3.4	2.7	2.5	2.7
Glutamic acid	18.5	15.0	16.3	16.0	15.6	16.3	15.9	19.3	13.8	19.4
Proline	4.8	4.9	4.1	4.8	4.1	7.8	12.0	7.3	8.8	8.2
Glycine	1.6	1.5	1.6	1.6	1.7	2.3	1.9	1.2	2.8	2.2
Alanine	4.2	4.1	4.0	4.3	3.9	2.3	3.1	2.1	2.3	2.3
Cystine	1.7	2.1	2.2	2.1	2.2
Valine	5.0	5.0	4.3	5.3	4.7	4.7	4.8	4.3	4.7	4.6
Methionine	1.2	2.1	1.7	2.0	2.0	1.4	4.8	1.5	0.6	1.3
Isoleucine	5.6	5.9	5.2	5.7	4.9	4.5	5.6	4.9	4.6	5.0
Leucine	11.6	11.2	12.6	11.5	12.6	8.1	9.0	6.4	6.5	8.2
Tyrosine	3.0	3.0	3.5	3.2	3.7	3.7	3.0	4.6	2.7	3.9
Phenylalanine	3.0	2.8	3.2	3.0	3.3	5.6	3.6	4.6	6.0	5.1
Histidine	2.7	2.7	2.9	2.7	2.9	5.2	3.7	5.4	6.7	4.3
Lysine	10.5	11.0	11.5	10.1	11.7	15.0	14.5	19.0	16.7	15.0
Arginine	2.9	2.6	3.1	2.8	3.2	9.9	4.1	7.3	12.5	7.9
Tryptophan	0.8	1.9	2.3	1.2	2.7

^a Percent of total amino acids recovered; uncorrected for hydrolytic destruction.

RESULTS AND DISCUSSION

Whey Composition. The value of whey in feed and food uses is based primarily on the protein content, which is usually determined by multiplying the total nitrogen value by the factor 6.38. Protein values determined by this procedure fall in the range of 12–14%. Although this method provides a rapid approximate value, it is recognized that the inclusion of the dialyzable nitrogen in the calculations results in values 20–25% higher than the actual protein content.

A large part (30% or more) of the whey protein fraction was not soluble at pH 7–9; in addition, considerable protein precipitation occurred upon acidification of the whey solution to pH 4.6. The insoluble protein fraction could be solubilized by use of 2-mercaptoethanol and urea, which reflects considerable heat denaturation of whey proteins during the commercial drying process. No protein denaturation was evident in the freeze-dried whey samples.

Several methods for protein precipitation were investigated for their applicability to the whey system. Protein coagulation, brought about by acidification to pH 4.6 and heating at 95–100° for 30 min, left about 5% of nondialyzable nitrogen in the supernatant. Trichloroacetic acid (12%) alone is not capable of precipitating the macropeptide liberated by rennin action on κ -casein (Nitschmann and Henzi, 1959). Although the total precipitable nitrogen increased when trichloroacetic acid was combined with phosphotungstic acid (deKoning *et al.*, 1966), 1–3% of nondialyzable nitrogen remained soluble. The above methods are not suitable for the determination of total protein in whey because of incomplete precipitation of nondialyzable nitrogen. The protein content was determined therefore by multiplication of the nondialyzable nitrogen values, obtained after prolonged dialysis, by the factor 6.38.

The composition of the wheys is reported in Tables I and II. Total nitrogen values for the whey samples ranged between 1.82 to 2.40%, with an average value of 2.03%. These total nitrogen values, when multiplied by the factor 6.38, yield crude protein values of 11.6–15.4% (average 12.9) in agreement with values reported by Watt and Merrill (1963). Dialysis studies revealed that the nondialyzable nitrogen constituted 64.4–83.7% (average 75.2%) of the total nitrogen and that the protein content fell in the range of 8.0 and 11.5% (average of 9.7%). Comparable values were obtained for typed or blended wheys.

The lipid content of the blended sweet wheys averaged about 1%. However, the typed wheys showed considerable variation. Lipid values of 0.36 and 0.46% were obtained from skim Cheddar and cottage cheese wheys, while values of 2.7 and 4.2% were obtained for Cheddar and Swiss wheys. The lipid nitrogen averaged 0.5% of the total nitrogen.

The lactose content showed some variation, but all were in the normal range with an average of 71.7% of the total solids.

Ash content of the acid wheys (11.3 and 11.5%) was considerably higher in comparison to the sweet wheys (8.23% average). This is to be expected since a large part of the calcium phosphate is liberated into the whey upon acidification. In sweet wheys, however, the largest part of the calcium phosphate remains bound to the casein curd formed through the action of rennin (Harwalker and Emmons, 1969).

Amino Acid Composition of Whey Fractions. The amino acid compositions of the dialyzable and nondialyzable whey fractions are reported in Table III, along with the composition of a whey control obtained following isoelectric precipitation of the casein. The composition of the nondialyzable control fractions is in excellent agreement with that reported by Finnish workers (Uusi-Rauva *et al.*, 1969). The amino acid compositions of the nondialyzable fractions of the commercial and control wheys were quite comparable, although some variation in values was evident. The variation in composition among the dialyzable fractions was substantially greater, possibly reflecting secondary rennin proteolysis. Cheddar whey showed the greatest difference in composition, particularly for proline, alanine, methionine, histidine, and arginine. No explanation is offered for these divergent values except to suggest that proteolysis or microbial contamination might have been causative factors.

ACKNOWLEDGMENT

The authors are indebted to M. T. Lukasewycz for the ash determinations.

LITERATURE CITED

- Association of Official Agricultural Chemists, "Official Methods of Analysis," 10th ed, 38.012, 1965a, p 744.
 Association of Official Agricultural Chemists, "Official Methods of Analysis," 10th ed, 15.016, 1965b, p 223.
 Cerbulis, J., *J. Agr. Food Chem.* **15**, 784 (1967).
 deKoning, P. J., Eisses, J., DeVries, H., *Neth. Milk Dairy J.* **20**, 204 (1966).
 Harwalker, V. R., Emmons, D. B., *Can. Inst. Food Technol. J.* **2**, 9 (1969).
 Marier, J. R., Boulet, M., *J. Dairy Sci.* **42**, 1390 (1959).
 Nitschmann, H., Henzi, R., *Helv. Chim. Acta* **42**, 1985 (1959).
 Spies, J. R., Chambers, D. C., *Anal. Chem.* **21**, 1249 (1949).
 Uusi-Rauva, E., Pajula, R., Antila, M., *Suom. Kemistilehti B* **42**, 328 (1969).
 Watt, B. K., Merrill, A. L., *Composition of Food, Agricultural Handbook No. 8*, USDA, Washington, D.C., Revised 1963.

Received for review March 15, 1972. Accepted June 8, 1972.
 Reference to a brand or firm name does not constitute endorsement by the U.S. Department of Agriculture over others of a similar nature not mentioned.