

THE NUTRITIVE VALUE OF SKIM-MILK POWDERS, WITH SPECIAL REFERENCE TO THE SENSITIVITY OF MILK PROTEINS TO HEAT¹

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INTRODUCTION

In the commercial dehydration of raw liquid skim milk the immediate practical objective is the production of a dry powder that may be conveniently stored and readily transported. The agency of dehydration is heat. In the merchandising of these powders emphasis is placed upon their high nutritive value, particularly as sources of protein, of calcium, and of vitamin G. It is commonly, if tacitly, assumed that they are equal in nutritive value to the solids of raw liquid skim milk. However, there is more than a remote possibility that the heat treatment to which they have been subjected has lowered their nutritive value, although little definite evidence to this effect has been found in the literature.

In the drying of liquid skim milk, there are times when the resulting products are scorched. This scorching is caused in different ways, and under careful management it may be prevented. The point of interest here is that these scorched products are offered for sale through the various merchandising channels. In such cases there is ocular evidence that processing has changed the milk solids, but to what extent the nutritive value has deteriorated there is again no available evidence.

While many types of equipment are used in the manufacture of dry skim-milk powders, only two distinct processes of dehydration are in general use, one the roller process, and the other the spray process. Powders made by the roller process are largely merchandised as animal feed, while the powders sold for human consumption are largely prepared by the spray process. Whether the powders produced by the two processes are of equivalent nutritive value is not definitely known.

Of the 288,000,000 pounds of dry skim-milk powder produced in the United States in 1933, about 60 percent went into human consumption through such media as bakery goods, ice cream, confectioneries, and breakfast cereals. The remaining 40 percent was sold as animal feed. The former channels of consumption obviously represent the most particular and fastidious of sales distribution, in which a scorched product is frowned upon because of its off color and taste. The animal-feed buyer is not opposed to the scorched product because of color or taste, but he does, and should, view it with some doubt on the score of its nutritive value.

¹ Received for publication July 11, 1935; issued February 1936. The experimental data reported in this article have been taken in part from the thesis of B. W. Fairbanks submitted in 1935 to the Graduate School of the University of Illinois in partial fulfillment of the requirements for the degree of doctor of philosophy in animal husbandry, and in part from an experiment station project. The investigation carried on by the senior writer was made possible by the donation of funds and samples of skim-milk powders to the University of Illinois by the American Dry Milk Institute, Inc.

The investigation reported in the following pages was planned to solve these problems, in part at least, but mainly as they relate to the changes in the nutritive value of milk solids induced by the heat processes employed in the commercial manufacture of skim-milk powders.

REVIEW OF LITERATURE

Nevens and Shaw (13)² showed that the digestibility of the proteins of whole-milk powder was appreciably less than that of the proteins of liquid whole milk. They did not observe any significant differences in the digestibility of the proteins of whole-milk powders produced by the roller and the spray processes, although Miyawaki, Kanazawa, and Kanda (11) claim, on the basis of *in vitro* digestion experiments, that the digestibility of the proteins of roller-process powder is somewhat less than that of spray-process powder.

Chick (2) and Goldblatt and Moritz (5) compared the growth-promoting values of unheated and heated casein, obtaining no evidence of a deleterious effect of heat. In these tests the casein was heated for 36 to 72 hours at 110° to 130° C. As high-protein diets were used in both investigations, a possible slight inferiority of the heated casein may well have been masked. The more rapid resumption of growth in rats following depletion of vitamin A when unheated casein was used than when casein heated at 105° C. for 7 days was used, noted by Coward, Key, Morgan, and Cambden (3), may well have been a result of heat upon the casein itself rather than upon any vitamin A originally present in it. Morgan (12) obtained consistently lower biological values for toasted (150° for 30 minutes) than for raw casein, and in later work with Greaves (6) an attempt was made to trace the heat (140° for 30 minutes) deterioration to the destruction of certain of the amino acid components of casein; i. e., lysine, histidine, tyrosine, cystine, and tryptophane. From growth experiments they concluded that the nutritive value of both heated and unheated casein was improved by a supplement of cystine and that that of the heated casein (but not of the unheated casein) was improved by supplements of either lysine or of histidine; in nitrogen-balance experiments they found that the biological value of unheated as well as of heated casein was increased by a cystine supplement, while that of the heated casein was improved also by a supplement of lysine. Such results, indicating a supplementation of proteins simultaneously by more than one amino acid indispensable to life, are contrary to prevailing conceptions of the relation between the composition of proteins and their value in promoting protein synthesis in animals. Block, Jones, and Gersdorff (1) found that the lysine yielded by the acid hydrolysis of casein was not lowered by previous treatment of the casein with dilute sodium hydroxide or by dry heat (150° for 65 minutes).

The above-cited experiments on heated versus unheated casein are of no definite value in predicting the effect on the nutritive value of milk solids of dehydrating by the prevailing commercial processes, mainly because the time of exposure to heat is so much shorter in the latter case. Thus, in the roller process, the milk film is in contact with the drying roll for only about 4 seconds, and for only a fraction of this time is it exposed to the maximum roller temperature, estimated at

² Reference is made by number (*italic*) to Literature Cited, p. 1120.

134° C.³ In the spray process the raw milk is preheated at 60° to 63°, concentrated in a vacuum pan, and sprayed into a current of air at temperatures ranging from 93° to 149°.

OBJECT, MATERIALS, AND METHODS

The object of the experiment was to compare the digestibilities and the biological values of the proteins of a series of skim-milk powders prepared by different methods and processes with each other and with those of a sample of raw skim milk which had not been subjected to any heat treatment whatever. Albino rats were used as subjects. Somewhat incidentally, the net-energy values of the two roller-process powders subjected to the severest and the mildest heat treatment were compared. A study was also made of the amino acids limiting the nutritive values of the proteins of the roller-process powders, for the purpose of determining the location of the heat injury.

Six samples of dry skim-milk powders were prepared in the Dairy-men's League plant at Massena, N. Y., from the same supply of milk. Four of the samples were made upon twin-cylinder atmospheric rolls and the remaining two were prepared by the continuous spray process. These six samples were compared to raw liquid skim milk.

Raw liquid skim milk.—This sample was obtained from the Division of Dairy Manufactures of the University of Illinois and was drawn before any heat was applied. While this sample of skim milk was not taken from the liquid skim milk from which the powders were prepared, the discrepancy is not serious, as the proteins of skim milk are comparatively constant in amount and proportions.

Low-temperature powder, roller process (r. p.).—The steam gage at the rolls registered 50-pound pressure, and the film of milk delivered to the roll was thinner than in common practice. This powder was exposed to a temperature less than that commonly employed in commercial drying.

Choice commercial powder, roller process (r. p.).—Ordinary commercial plant procedures were employed, including 87 pounds of steam pressure and a milk film of ordinary thickness. This sample was a high-quality product, and would receive the highest grade adopted by the standards committee of the American Dry Milk Institute.

Slightly scorched powder, roller process (r. p.).—The steam pressure was increased to 90 pounds and a thin film of milk was delivered to the rolls. This sample was representative of the scorched milk powder frequently encountered in the trade.

Scorched powder, roller process (r. p.).—The knives were lifted intermittently from the rolls, so that the milk solids made more than one revolution of the rolls. The product was representative of the extreme scorching occasionally found in market samples.

Not preheated powder, spray process (s. p.).—The usual preheating process was omitted. Such a powder is not frequently encountered in the trade.

Preheated powder, spray process (s. p.).—The usual preheating process which is considered necessary for the preparation of a high quality good-baking spray-process powder was included.

³ No estimate of the maximum temperature to which the milk film is exposed on the roll can possess a high degree of accuracy nor be generally applicable under all conditions.

All samples were shipped to Urbana in tin containers with tight-fitting lids, except the choice commercial powder, which was shipped in a well-made barrel lined with paper. On arrival the latter sample was transferred to 50-pound lard pails. All samples were stored in a dry place.

For purposes of precise description, the chemical composition, solubility, and color analysis of the skim-milk powders are given in table 1.

TABLE 1.—*Chemical composition, solubility, and color analysis of the experimental dry skim-milk powders*

Sample of powder	Chemical composition					Solubility		Color analysis		
	Dry substance	Nitrogen	Crude protein (N×6.25)	Fat	Gross energy per gram	Dry substance in solution	Nitrogen in solution	Brightness	Dominant wave length	Purity
	Percent	Percent	Percent	Percent	Calories	Percent	Percent	Percent	m μ	
Low-temperature, r. p.	96.12	5.41	33.81	0.91	4.06	71.3	34.6	84.5	600	1
Choice commercial, r. p.	98.06	5.50	34.38	.99	4.02	67.7	32.4	84.6	560	23
Slightly scorched, r. p.	99.87	5.41	33.81	1.07	4.02	62.4	27.9	77.1	580	17
Scorched, r. p.	98.90	5.55	34.69	.90	4.25	64.6	24.9	62.0	587	18
Not preheated, s. p.	98.32	5.40	33.75	.69	4.13	95.1	92.3	95.5	590	1
Preheated, s. p.	98.40	5.50	34.38	.62	4.12	92.6	91.6	94.5	580	4

The method for determining solubility was adapted from the method of Wright (17). Twenty grams of skim-milk powder were transferred to a beaker, to which was added 200 cc of nitrogen-free water at 20° C. The beaker was placed in a water bath of 20°, and the contents stirred mechanically for exactly 30 minutes. Fifty cubic centimeters of the mixture were transferred to each of two centrifuge tubes and whirled at 1,850 revolutions per minute for exactly 15 minutes. Three 10-cc samples of the supernatant fluid were removed from one centrifuge tube for moisture determination, while from the second centrifuge tube three 10-cc samples were taken for the determination of nitrogen.

The colors were analyzed with a Keuffel and Esser color analyzer or spectrophotometer. For these analyses the samples were ground in a mortar until all material passed through a 100-mesh sieve. The three characteristics of color—brightness, dominant wave length, and purity—are expressed numerically. As most of these powders were white or very nearly so, it is believed that the figures for brightness are the most significant.

The data of table 1 show that, with a single exception for solids and none for nitrogen, the solubility of the roller-process powders decreased as the severity of the heat treatment increased. The spray-process powders were much more soluble than the roller-process powders, and of the two former the preheated was the less soluble. Also the spray-process powders possessed a greater brightness rating than the roller-process powders, while among the latter, brightness decreased as the intensity of heat processing increased.

In many of the experiments the paired-feeding method (9) was used, while for the determination of the biological values of protein the nitrogen-balance method developed in this laboratory (7, 10) was followed.

EXPERIMENTAL RESULTS

THE DIGESTIBILITY OF ENERGY

The apparent digestibility of energy was determined upon two of the roller-process powders, the low-temperature powder and the scorched powder, representing the two extremes of heat treatment. This determination was made upon eight pairs of rats which were being used in a feeding experiment designed to compare the net-energy values of these powders. The results of the net-energy test are presented below. The rations compared contained about 63 percent of the respective powders. The collection periods were of 7 days duration and a feces marker was used.

The average coefficients of apparent digestibility of the energy of these two diets were 90.5 for the low-temperature powder and 89.2 for the scorched powder. In all pairs a higher coefficient was obtained with the low-temperature powder. Such a consistent result would be produced by chance only once in 128 trials, so that its significance is established without further statistical analysis. It may be concluded that the extreme heat employed in preparing the scorched powder depressed the digestibility of its energy in the animal body. Although the average depression for the ration as a whole was 1.375 percent, that of the powder itself, making up only 62.3 percent of the ration, must have been $1.375 \div 0.623 = 2.21$ percent.

THE DIGESTIBILITY OF PROTEIN

The digestibility of the protein in raw liquid skim milk and in the various skim-milk powders was determined from the nitrogen metabolism data secured for the purpose of computing the biological values of these protein mixtures. In these metabolism trials the various samples were compared in turn with the low-temperature (r. p.) powder, the same rats being used in each comparison in groups of 5, 8, or 10. The average coefficients of true digestibility (including due allowance for the metabolic products in the feces) are given in table 2.

TABLE 2.—Average coefficients of true digestibility of protein for the various experimental skim-milk samples

Rats (number)	Raw liquid skim milk	Roller-process powders				Spray-process powders	
		Low temperature	Choice commercial	Slightly scorched	Scorched	Not preheated	Preheated
8	94.8	92.7					95.3
5		90.6	93.4			92.0	
5		91.7		88.8		92.0	
5		92.1			81.4		

¹ Average of 10 determinations, 2 on each rat.

If digestion coefficients obtained for any two samples on the same rat are considered as paired observations, then Student's (15) method for the statistical analysis of small groups of such data may be applied. Such an analysis of the individual data shows that the proteins of liquid skim milk are significantly more digestible than the proteins of

the low temperature (r. p.) powder, and that the latter proteins are significantly better digested than those of the slightly scorched (r. p.) and scorched (r. p.) powders. All other differences are statistically insignificant.

These results are only in partial agreement with those of Nevens and Shaw (13), who were able to demonstrate a significantly greater apparent digestibility of the proteins of fresh whole milk than of the proteins of whole-milk powder prepared by either the spray process or the roller process. In these studies the sole diet of the experimental rats consisted of the milk products under test, while in the investigations reported in this article the experimental diets contained less than 25 percent of milk solids. The former dietaries would be more favorable to the detection of differences in the digestibility of milk products than would the latter.

THE NET-ENERGY VALUE

The two skim-milk powders representing the two extremes in heat treatment; i. e., the low-temperature and the scorched roller-process powders, were compared with reference to their availability as sources of energy in animal nutrition. They were incorporated into rations containing other sources of protein, minerals, and vitamins in presumably adequate amounts, so that the milk solids need supply only energy-yielding nutrients. The composition of the rations is given in table 3. The water was added to diet 2 in order to equalize the moisture content of the two powders (table 1).

TABLE 3.—Composition of the diets used in the comparison of the net-energy values of low-temperature (r. p.) skim-milk powder and scorched (r. p.) powder

Constituent	Diet 1		Diet 2	
	Percent	Percent	Percent	Percent
Low-temperature (r. p.) powder.....	63.50			
Scorched (r. p.) powder.....		62.34		
Casein.....	25.00	25.00		
Modified Osborne and Mendel salts ¹	4.50	4.50		
			Dried yeast ²	5.00
			Cod-liver oil.....	2.00
			Water.....	1.16

¹ See Wesson (16).

² Obtained from the Northwestern Yeast Co.

The rations were compared with respect to growth-promoting power by means of the paired feeding method, using eight pairs of rats. The growth results are assembled in table 4.

TABLE 4.—Comparison of the net-energy value of low-temperature powder (r. p.), diet 1, and scorched powder (r. p.), diet 2, as determined by the paired-feeding method during a feeding period of 56 days

Pair no. and powder used in diet	Total food	Initial weight	Final weight	Gain	Pair no. and powder used in diet	Total food	Initial weight	Final weight	Gain
Pair 1, males:					Pair 5, males:				
Low-temperature powder.....	Grams 527	Grams 49	Grams 155	Grams 106	Low-temperature powder.....	Grams 446	Grams 40	Grams 131	Grams 91
Scorched powder.....	527	51	145	94	Scorched powder.....	446	40	113	73
Pair 2, males:					Pair 6, females:				
Low-temperature powder.....	557	50	160	110	Low-temperature powder.....	469	46	122	76
Scorched powder.....	557	47	157	110	Scorched powder.....	469	46	126	80
Pair 3, females:					Pair 7, females:				
Low-temperature powder.....	527	48	134	86	Low-temperature powder.....	423	46	109	63
Scorched powder.....	527	50	144	94	Scorched powder.....	423	53	109	56
Pair 4, females:					Pair 8, males:				
Low-temperature powder.....	479	41	124	83	Low-temperature powder.....	499	52	143	91
Scorched powder.....	479	40	123	83	Scorched powder.....	499	51	151	100

After the feeding period was well under way, it appeared that both diets were deficient in vitamin B. In order to maintain appetite, each rat was given daily by mouth from 3 to 6 drops of tikitiki extract, pair mates being treated exactly alike.

The gains in weight of pair mates reveal no superiority of diet 1 over diet 2. In 3 pairs the rat on diet 1 gained the faster, in 3 pairs the reverse was true, while in 2 pairs the gains were equal.

Since it was possible that a difference in net-energy value between diets 1 and 2 produced a difference in the energy balance rather than in the rate of gain of the rats, the carcasses of four pairs, nos. 1, 3, 5, and 8, were analyzed for gross-energy content, using the Parr oxygen bomb calorimeter. Their contents of nitrogen were also determined. The results are summarized in table 5.

TABLE 5.—Nitrogen and energy contents of the carcasses of 4 pairs of rats reared on diets 1 and 2¹

Pair no.	Dry skim-milk powder in diet	Empty body weight	Nitrogen content		Energy content	
					Per gram	Total
		Grams	Percent	Grams	Calories	Calories
1	{ Low temperature	150	2.86	4.29	1.32	198
	{ Scorched	139	2.77	3.85	1.32	183
3	{ Low temperature	129	2.94	3.79	1.32	170
	{ Scorched	138	2.44	3.37	1.32	182
5	{ Low temperature	127	2.79	3.54	1.33	169
	{ Scorched	108	2.86	3.09	1.32	143
8	{ Low temperature	138	3.04	4.20	1.31	181
	{ Scorched	145	2.96	4.29	1.31	190

¹ Diet 1 contained the low temperature and diet 2 the scorched powder.

The energy content per gram of empty carcass was remarkably constant for all rats examined, and the total energy content of pair mates showed no consistent differences induced by the two diets. In 2 pairs the rat subsisting on diet 1 stored the greater amount of energy in its body, while in the remaining 2 pairs the reverse was true.

The nitrogen content of the carcass of the rat receiving the low-temperature powder (diet 1) was higher, both on the percentage and the absolute basis, in 2 of the 4 pairs than that of the rat receiving the scorched powder, but these differences are not sufficiently large or consistent or numerous to indicate with any great degree of certainty, according to statistical analysis, that they were the result of the difference in diet consumed rather than the result of chance.

The digestibility coefficients above discussed indicate clearly that the energy of the scorched skim-milk powder was somewhat less digestible than that of the low-temperature powder. The failure to demonstrate a difference in the net-energy values of the two powders by the growth experiment may be the result of (1) a greater degree of activity of the rats on the diet containing the low-temperature powder, (2) a greater specific dynamic effect of this diet or (3) a biological error in the growth experiment sufficient to obscure the effect of the greater digestibility of the low-temperature powder.

In all probability any difference in net-energy value between the low-temperature skim-milk powder and the scorched powder is considerable. It may be concluded further that powders subjected to

heat insufficient in intensity to produce more than a barely perceptible scorching are for all practical purposes equally valuable as sources of energy in the animal body.

BIOLOGICAL VALUE OF THE PROTEINS

The general plan of experimental procedure in determining the biological value of the proteins was that developed by Mitchell (7, 10), except that in these metabolism studies feces markers⁴ were employed. The compositions of the various diets are presented in table 6. In order to improve the consumption of the experimental diets, Harris yeast vitamin powder was fed. This vitamin concentrate was fed in some instances as part of the diet, in others as part of the diet supplemented by additional amounts weighed into individual feed dishes, and later by feeding all of it as an addition to the mixed diet, which accounts for the presence of more than 1 percentage of this constituent in a given diet. As the amount of vitamin powder was changed in the diet, a corresponding and equal change was made in the amount of starch.

TABLE 6.—Composition of the diets used in studying the relative biological values of the proteins of raw liquid skim milk and the various dry skim-milk powders

Constituents	Stand- ardiz- ing diet	Raw liquid skim milk	Low- tem- per- a- ture powder (r. p.)	Choice com- mer- cial powder (r. p.)	Slightly scorched powder (r. p.)	Scorched powder (r. p.)	Not pre- heated powder (s. p.)	Pre- heated powder (s. p.)
	Percent	Percent	Percent	Percent	Percent	Percent	Percent	Percent
Dried fat-free whole egg.....	{ 5.47 or 5.35 }							
Raw liquid skim milk.....		1 22.55						
Low-temperature powder (r. p.).....			23.66					
Choice commercial powder (r. p.).....				23.27				
Slightly scorched powder (r. p.).....					23.66			
Scorched powder (r. p.).....						23.06		
Not preheated powder (s. p.).....							23.70	
Preheated powder (s. p.).....								23.27
Modified Osborne-Mendel salts.....	4.5	4.5	4.5	4.5	4.5	4.5	4.5	4.5
Filtered butterfat.....	8.0	8.0	8.0	8.0	8.0	8.0	8.0	8.0
Cod-liver oil.....	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Sucrose.....	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
NaCl.....	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Harris yeast vitamin powder.....	{ .5 or .0 }		{ .3 or .5 or .0 48.54 or 48.34 or 48.84 }	.5	.5	.5		
Starch.....	{ 66.53 or 67.15 }	49.95		48.73	48.34	48.94	48.80	49.20
Pigment.....	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0

¹ Dry substance.

The calculations of the biological values of the proteins of the various milk samples was made from the nitrogen metabolism data according to the usual procedure followed in this laboratory (10); hence it seems unnecessary to give them in detail here. The biological

⁴ On the first day of each experimental period the diet fed contained 2 percent of ferric oxide, which marked the feces red. On the remaining days of the experimental period, in most cases of 7 days' duration, an equal percentage of barium sulphate was included in the diet. This produced a progressively lighter color in the feces. On the first day following an experimental period, or the first day of a transition period, which was always 4 days in length, the diet contained 2 percent of chromic oxide to mark the feces green.

value is the percentage of the absorbed nitrogen that is retained for both maintenance and growth under the standard conditions adopted for the determination. The estimate of the absorbed nitrogen makes due allowance for the metabolic nitrogen excreted in the feces, the ratio between metabolic fecal nitrogen and dry-matter intake being measured in the standardizing periods. The estimate of the total storage of nitrogen in the body makes due allowance for the endogenous nitrogen excreted in the urine, the ratio between endogenous nitrogen and body weight being also determined in the standardizing periods.

The various milk samples were compared in turn with respect to the biological value of their protein components with the low-temperature (r. p.) powder, groups of 5, 8, or 10 rats being used in each comparison. The average biological values for each group of rats and for each milk sample are assembled in table 7.

TABLE 7.—Average biological values of protein for the various experimental skim-milk samples

Rats (number)	Raw liquid skim milk	Roller-process powders				Spray-process powders	
		Low temperature	Choice commercial	Slightly scorched	Scorched	Not preheated	Preheated
8	89.8	89.1					82.3
5		89.6	81.6			87.2	
5		89.3		68.0		88.6	
5		87.9			69.8		

¹ Average of 10 determinations, 2 on each rat.

The average biological value of 89.8 for the proteins of liquid skim milk may be compared with the average of 95 obtained by Shiftan (14) with two pigs fed a ration containing about 12 percent of protein. The average of 82.3 for preheated skim-milk powder (s. p.) is very close to the value of 85 obtained in this laboratory in two investigations (8, 10) with milk powders of this description, and also with the value of 86 reported by Boas Fixsen and Jackson (2), relating to a whole-milk powder manufactured by the roller process.

The biological values (single or average) obtained for different samples on the same rats have been considered as paired observations, and the differences between them have been subjected to statistical analysis according to the method of Student (15). The statistical results are summarized in table 8. From these analyses it may be concluded that the nutritive value of the digestible proteins of fresh skim milk is not depressed by drying by the roller process at the lowest feasible temperature, nor by drying by the spray process provided the preheating is dispensed with. However, if drying is accomplished by the prevailing commercial processes, yielding products represented by the choice commercial (r. p.) sample and the preheated (s. p.) sample, a definite lowering of nutritive value occurs, equal to about 8 percent. If drying by the roller process is so poorly controlled that even slight scorching occurs, then a much greater decrease in nutritive value is effected, amounting to more than 20 percent.

TABLE 8.—Analysis according to Student's method of the differences in biological values between the proteins of dry skim-milk powders and of liquid skim milk

Statistical item	Raw liquid skim milk <i>v.</i> low-temperature powder (r. p.)	Low-temperature powder (r. p.) <i>v.</i> not preheated powder (s. p.)	Raw liquid skim milk <i>v.</i> preheated powder (s. p.)	Low-temperature powder (r. p.) <i>v.</i> preheated powder (s. p.)	Low-temperature powder (r. p.) <i>v.</i> choice commercial powder (r. p.)	Low-temperature powder (r. p.) <i>v.</i> slightly scorched powder (r. p.)	Low-temperature powder (r. p.) <i>v.</i> scorched powder (r. p.)
Mean of differences, <i>M</i>	0.625	1.7	7.5	6.875	7.8	21.6	18.2
Standard deviation of differences, <i>s</i>	4.22	4.24	3.54	3.14	1.6	2.32	3.25
Probability, <i>P</i>35	.13	.0004	.0003	<.0019	<.0019	.0019

TOTAL EFFECT OF DRYING UPON THE NUTRITIVE VALUE OF SKIM-MILK PROTEINS

The relative protein values of the various milk samples tested, which take account of differences in digestibility of protein as well as differences in biological value, have been computed and the results are presented in table 9. These values, listed in the last column of the table, are expressed in percentages, the value of raw liquid skim milk being taken as 100. From these values it appears that skim milk may be dried with a loss in protein value of only 5 percent or less, but that by the ordinary processes of commercial drying losses of 9 to 11 percent occur, and if there is perceptible scorching through careless management the loss may be 30 percent.

TABLE 9.—Relative protein values of the different milk samples tested ¹

Sample	Average true digestibility of protein	Average biological value	Relative protein value	Sample	Average true digestibility of protein	Average biological value	Relative protein value
	Percent	Percent	Percent		Percent	Percent	Percent
Raw liquid skim milk.....	95	90	100	Spray-process powders:			
Roller-process powders:				Not preheated.....	92	88	95
Low temperature.....	91	89	95	Preheated.....	95	82	91
Choice commercial.....	93	82	89				
Slightly scorched.....	89	68	71				
Scorched.....	81	70	66				

¹ Expressed as percentages with the relative protein value for raw liquid skim milk taken as 100.

Thus, the data obtained demonstrate a point of tremendous practical importance, namely, that the proteins of milk are very sensitive to heat with respect to their value in nutrition, and that considerable losses in protein efficiency may be incurred unless the time of exposure to heat and the intensity of the heat are carefully controlled. In this respect the requirements for the highest engineering efficiency may conflict with the requirements for the highest quality of product, and the dry-milk industry must decide which requirements are paramount.

SEAT OF INJURY TO THE PROTEINS OF MILK PRODUCED BY DRYING IN TERMS OF THE CONSTITUENT AMINO ACIDS

Having established the existence of definite deterioration of skim-milk proteins during commercial drying processes, as well as the

extent of such deterioration, the next step in the investigation was to discover, if possible, which of the constituent amino acids was involved in the successive decreases in nutritive value as the severity of the drying process was intensified. Such information would be afforded by a study of the amino acids limiting the nutritive values of the various samples of skim-milk powders.

In this study the paired-feeding method was used to compare the growth-promoting powers of rations containing the various milk powders as the sole source of protein and the same rations with small supplements (0.3 percent) of selected amino acids. The rations all contained enough of the milk powders (23 to 27 percent) to supply approximately 9 percent of protein, 4.5 or 5.0 percent of the Wesson (16) modification of the Osborne and Mendel salt mixture, 1 percent of NaCl, 10 or 12 percent of sucrose, 2 percent of cod-liver oil, 8 percent of butterfat, 10 or 12 percent of lard, and enough starch to make 100 percent. Vitamins B and G were supplied extra as Harris yeast vitamin powder.

The deficiency of the low-temperature (r. p.) powder in cystine was readily demonstrated in a 14-day feeding test with eight pairs of rats. In this short period the rat in each pair given the cystine supplement gained more in weight than its pair mate on the unsupplemented diet on the same amount of food and attained a greater body length. The average excess gain by the test rats was 7.37 g, the standard deviation of excess gains was 2.75 g, and the probability that fortuitous factors would have produced so consistent an outcome is only 0.0001, according to Student's (15) probability tables. The average difference in body length was 5.50 mm, the standard deviation of differences 3.94 mm, and the probability of a chance outcome only 0.0038.

That preheated skim-milk (s. p.) powder is also deficient in cystine had been previously demonstrated by Mitchell and Beadles (9), the skim-milk powder used being of this description.

The evidence thus far adduced indicates that the initial drop of about 9 to 11 percent in the nutritive value of skim-milk proteins during drying by prevailing commercial methods must be the result of a destruction of cystine, since cystine is still the amino acid limiting the biological value of commercial skim-milk powders. However, the growth-promoting value of the slightly scorched (r. p.) powder was not improved by a cystine supplement. Eight pairs of rats were used in this test, and at the end of 2 weeks the gains of pair mates were very nearly the same, being exactly equal in 2 pairs, only 1 g apart in 4 pairs, 2 g apart in 1 pair, and 5 g apart in the remaining pair. The rats receiving cystine supplements gained more than their control mates on the same amount of food in only 2 pairs. At this point in the feeding experiment the control rats, previously consuming the unsupplemented diet, were given a supplement of lysine dihydrochloride equal to 0.3 percent of the basal diet. The other rats in each pair continued on the basal diet plus cystine.

Three weeks after this change in plan was put into effect, the rats receiving the lysine supplement had exceeded in gain their pair mates receiving the cystine supplement in all of the 8 pairs and had also attained to greater body lengths in all pairs. The average difference in gain between pair mates was 6.37 g, the standard deviation of differences 2.19 g, and the probability that a purely random com-

combination of factors common to both pair mates would produce as consistent an outcome as this is negligible, amounting to less than 0.0001. The average difference in body length (from nose to root of tail) is 5.25 mm, the standard deviation of differences, 3.23 mm, and the desired probability only 0.0018, again entirely negligible. Body length measurements, somewhat more surely than body weight measurements, are reliable criteria of growth, and in this case demonstrate beyond question that a lysine supplement to the proteins contained in the slightly scorched skim-milk powder increased their growth-promoting value. The complete data of this test will be found in table 10.

TABLE 10.—*Effects of supplements of cystine alone for 2 weeks and of cystine and lysine for 3 weeks on the growth-promoting value of slightly scorched skim-milk powder (r. p.)*

Results of the first 2 weeks of feeding					Results of the last 3 weeks of feeding					
Pair no. and diet or supplement	Total food	Initial weight	Final weight	Gain	Pair no. and supplement	Total food	Initial weight	Final weight	Gain	Body length
Pair 1, females:					Pair 1, females:					mm
Basal only.....	109	63	87	24	Lysine.....	164	87	118	31	182
Cystine.....	109	58	82	24	Cystine.....	164	82	106	24	173
Pair 2, females:					Pair 2, females:					
Basal only.....	97	56	74	18	Lysine.....	180	74	111	37	175
Cystine.....	97	56	73	17	Cystine.....	178	73	108	35	169
Pair 3, females:					Pair 3, females:					
Basal only.....	77	48	62	14	Lysine.....	142	62	93	31	163
Cystine.....	77	48	61	13	Cystine.....	144	61	88	27	160
Pair 4, females:					Pair 4, females:					
Basal only.....	85	46	62	16	Lysine.....	148	62	91	29	163
Cystine.....	85	47	64	17	Cystine.....	148	64	86	22	159
Pair 5, females:					Pair 5, females:					
Basal only.....	103	51	74	23	Lysine.....	186	74	114	40	173
Cystine.....	103	49	67	18	Cystine.....	186	67	99	32	167
Pair 6, females:					Pair 6, females:					
Basal only.....	102	53	70	17	Lysine.....	170	70	103	33	168
Cystine.....	102	54	69	15	Cystine.....	170	69	93	24	166
Pair 7, females:					Pair 7, females:					
Basal only.....	108	53	75	22	Lysine.....	184	75	112	37	176
Cystine.....	108	51	73	22	Cystine.....	184	73	102	29	165
Pair 8, females:					Pair 8, females:					
Basal only.....	96	47	67	20	Lysine.....	204	67	115	48	172
Cystine.....	96	48	69	21	Cystine.....	204	69	111	42	171

The sample of scorched skim-milk powder (r. p.), by similar paired-feeding tests, was found not to be improved in growth-promoting properties by supplements of cystine, histidine, or tryptophane. However, when supplemented by lysine, this sample, like the slightly scorched sample, exhibited a clear improvement in its power to promote growth. Only four pairs of rats were used in this test, with the results summarized in table 11. In each pair, the rat receiving the lysine supplement gained the faster and attained the greater body length. In spite of the fact that only four pairs of rats were used, the results are quite highly significant. Thus, the average difference in gain between pair mates was 7.75 g, the standard deviation of differences 4.09 g, and the probability of a fortuitous outcome, only 0.026. The average difference in body length was 5.75 mm, the standard deviation of differences 2.86 mm, and the desired probability only 0.020.

TABLE 11.—Effect of a lysine supplement on the growth-promoting value of the proteins of scorched skim-milk powder (*r. p.*)

Pair no. and diet or supplement	Total food	Initial weight	Final weight	Gain	Body length
	<i>Grams</i>	<i>Grams</i>	<i>Grams</i>	<i>Grams</i>	<i>Millimeters</i>
Pair 1, females:					
Basal only.....	310	59	89	30	161
Lysine.....	307	57	88	31	163
Pair 2, females:					
Basal only.....	359	61	94	33	163
Lysine.....	359	61	105	44	172
Pair 3, females:					
Basal only.....	278	49	72	23	153
Lysine.....	278	49	80	31	157
Pair 4, females:					
Basal only.....	285	55	72	17	154
Lysine.....	285	55	83	28	162

From this series of paired-feeding experiments it appears that the initial stages of the destruction of milk proteins by heat, occurring during the drying according to prevailing commercial processes, involves, and is the direct result of, a destruction of cystine, but that the later stages, from the initiation of perceptible scorching to the production of a thoroughly scorched product, involve a more rapid destruction of lysine than of cystine. This is a finding of considerable practical importance, since the value of milk proteins in supplementing the proteins of the cereal grains is the result of the deficiency of the latter in lysine and of the presence in the former of abundant proportions of this amino acid. Hence, scorched skim-milk powders, merchandisable only for animal feeding, would not possess this supplementing capacity.

CONCLUSIONS

The proteins of milk are very sensitive to the intensities and durations of heat treatment employed in commercial drying. However, it is possible to dry skim milk with commercial equipment without appreciably affecting its energy value or the nutritive value of its proteins. In the preparation of choice commercial roller-process powders, or of preheated spray-process powders, the biological value of the protein is lowered about 8 percent (from 90 to 82), although its digestibility is not appreciably affected. If preheating is omitted in the spray-drying process this reduction in nutritive value of the milk proteins does not occur. Since cystine is the amino acid limiting the biological value of the proteins of choice commercial roller-process powder and preheated spray-process powder, as well as of the original milk, it may be concluded that this initial decline in biological value of milk proteins is due to a partial destruction of cystine.

As the temperature of drying in the roller process is increased until perceptible scorching occurs the biological value of the milk proteins is rapidly lowered from 82 to 70 or less. The scorched products thus obtained are no longer benefited by cystine additions, but they do respond to lysine additions in increased nutritive value of their proteins. Hence the rapid change in milk proteins at the scorching point (or earlier) is primarily a result of the destruction of lysine. Such products, therefore, are of no value as supplements to the proteins of cereal grains. The digestibility of the milk proteins is also lowered at the scorching point in the roller-drying process, the extent

of lowering increasing more rapidly with the degree of scorching than the reduction in biological value.

However, even with extreme scorching, the net-energy value appears to be but slightly (if at all) affected. Apparently the protein disintegration occasioned by scorching conditions does not impair appreciably the value of the protein as a source of energy to the body.

The solubility of the total solids and the nitrogen of dry skim-milk powders is greater for spray-process than for roller-process powders and decreases in the latter with the intensity of the drying conditions. The spray-process powders gave a higher percentage of brightness than the roller-process powders when colors were analyzed, and in general brightness decreased as the heat increased. These solubility differences and color differences, however, are not reliable criteria of the changes occurring in the nutritive value of the proteins.

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