THE INFLUENCE OF COOKING AND CANNING ON THE VITAMIN B AND G CONTENT OF LEAN BEEF AND PORK [1]

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INTRODUCTION

As knowledge of vitamins and their importance in nutrition has broadened, it has become increasingly important to determine, not only the distribution and relative quantities of the several vitamins in raw foodstuffs, but also their vitamin potency as commonly prepared for human or animal consumption. It is well known that such processes as drying, cooking, and canning may alter markedly the vitamin potency of foods, the extent of alteration depending on the processes used and the particular vitamins contained in the food.

Meats have an important place in the American diet. Properly prepared, they furnish protein, energy, certain minerals, notably iron, and vitamins in a palatable and satisfying form. The importance of meat as a source of the vitamin G is well known.

The principal vitamins in lean meat are the antineuritic vitamin B (B_1) and the vitamin G (B_2) complex. Vitamin B is rather easily destroyed by heat, whereas vitamin G is stable, even at fairly high temperatures. Since beef and pork are commonly cooked or canned for human consumption, it seemed worth while to determine what effects, if any, these processes have on their vitamin potency.

PREVIOUS WORK ON VITAMINS B AND G IN MEAT

Comparatively little work had been done on the vitamin content of meats when this project was planned. Most of the work reported had been on raw meats, and involved the vitamin B complex, rather than the separate vitamins B and G.

With pigeons as test animals, Hoagland (*6*)[3] had found the muscle tissues of the mature ox, calf, and lamb relatively poor to fair in vitamin B, when fed in the form of dried-meat preparations. He found also that hog muscle was richer in vitamin B than ox or sheep muscle, 15 percent in the diet being sufficient to protect pigeons from polyneuritis. Baked pork tenderloin was practically as rich in vitamin as the uncooked muscle, but cooked ham had a somewhat lower value than the raw product. Later Hoagland (*7*) reported that dried hog muscle added to a diet of polished rice, as 5, 6.6, and 10 percent of the diet, prevented polyneuritis in pigeons, showing pork to be a good source of vitamin B.

[1] Received for publication Sept. 5, 1935; issued October, 1936. The vitamin terminology used is from the committee report of the American Biological Chemists (*13*)[3]. Paper no. 20, Journal Series, North Dakota Agricultural Experiment Station. The vitamins are also designated according to the English system in paragraph 3.

[2] The authors had the technical assistance of Mrs. Eleanor Bue, Bernice Emmons, and Dorothy Berrigan in feeding the rats and in the tabulation of data, and of L. L. Nesbitt, assistant agricultural chemist, in the preparation of food materials.

[3] Reference is made by number (italic) to Literature Cited, p. 432.

If, as indicated by Seidell (*12*), pigeons do not need vitamin G, but are very sensitive to vitamin B deficiency, then the foregoing work was, in reality, a measure of the relative amounts of the antineuritic factor vitamin B only, and not a measure of vitamin G or the vitamin B complex.

In 1926 Goldberger and his associates (*5*) published results of extensive experiments on both man and animals. In their studies on man, they found that a daily allowance of 200 g of fresh, lean beef, freed from visible fat, gristle, and tendons, when finely chopped and mixed with a little water, seasoned with salt and quickly brought to a boil, gave full protection against pellagra.

Their studies on pellagra in man were supplemented by extensive experiments with albino rats. The tests with rats furnished evidence that the so-called "water-soluble B" consisted of at least two factors. One was the antineuritic heat-labile factor and the other, a heat-stable factor, was probably the same as the pellagra-preventing factor (P-P) found in meat and yeast.

Shortly after the present project was planned, but before work was started, Hoagland (*8*) (1929) published a report of tests with both pigeons and rats. He found that 5 percent of dried lean pork protected pigeons against polyneuritis and loss in weight for 8 weeks and longer. Both fresh and smoked hams were found to be equally good sources of the antineuritic vitamin B. Beef was less potent in the antineuritic vitamin B, since it required 35 to 40 percent of the dried beef in the diet to protect pigeons against polyneuritis for 8 weeks.

In his work with rats, Hoagland did not test for vitamins B and G separately, but measured the combined effect of the "water-soluble vitamins." His experiments indicated that pork was a good source of the water-soluble vitamins, but not so good as brewer's yeast. He states (*8, p. 446*):

> Lean beef contained much less water-soluble B vitamins than lean pork. From 40 to 70 percent of dried fresh beef was required for excellent growth in rats as compared with 15 to 25 per cent of dried lean pork or 5 per cent of dried brewer's yeast.

Later, Hoagland and Snider (*10*) reported work on the relative quantities of growth-promoting vitamin G in beef, pork, lamb, beef spleen, beef liver, pork liver, and beef kidney. On the basis of air-dried, fat-free material, beef, pork, and lamb appeared to contain approximately the same quantities of vitamin G; 15 to 20 percent of either in the diet being sufficient for excellent growth in rats.

BASAL DIET FOR VITAMIN B DETERMINATIONS

After considering a number of commonly used basal diets for vitamin B determinations, it appeared that the basal diets used by Goldberger and his associates (*5*), and by Hoagland (*8*) were essentially alike and satisfactory for this work.

The following basal diet, free from vitamin B, was adopted and found satisfactory:

	Percent
Purified casein	20
Crisco	8
Cornstarch	51
Cod-liver oil	2
McCollum's salt mixture no. 185	4
Autoclaved baker's yeast	15

The casein, a lactic acid-precipitated product, was extracted with dilute acetic acid, essentially according to the recommendations of Steenbock,[4] but as tests showed that the first lot of casein prepared still contained some vitamin B, the method was modified to insure complete extraction. The procedure found satisfactory and adopted was to extract the casein (15 pounds in a large muslin sack) with dilute acetic acid (about 100 cc of glacial acetic acid to 60 liters of water) in a 25-gallon wooden tub for 12 days. The casein was thoroughly drained and the solution changed once a day. Following this period, the casein was washed with distilled water for 4 days. Each day the casein was drained and fresh water added. The extracted casein was then dried in an electric oven at 130° F. and ground to pass through a 20-mesh sieve.

Crisco was purchased on the market as required. The cornstarch was the ordinary household product and was used without further treatment. One lot of cornstarch was extracted with dilute acetic acid similarly to the casein, but as no difference in results was observed, no further extractions were made. The cod-liver oil was a high-grade product of tested potency.

A pure dehydrated bakers' yeast was used throughout. It was treated and autoclaved according to the method of Chase (*2*) and Chase and Sherman (*3*). It was dried in a cool air-drying apparatus (*11*). According to Chase, 15 percent of the dried autoclaved yeast in the basal diet would furnish an adequate amount of vitamin G without vitamin B. This was confirmed in feeding tests by the authors.

Numerous tests indicated that this diet was satisfactory for the quantitative estimation of vitamin B. The negative control rats, on the basal B-free diet alone, became depleted in 13 to 15 days with an average gain of 20 to 25 g, developing polyneuritis, which was followed by death, in 18 to 28 days after depletion. The positive controls fed the B-free diet plus a known source of vitamin B made normal gains over an indefinite period.

BASAL DIET FOR VITAMIN G DETERMINATIONS

In choosing a basal diet for the vitamin G determinations, it appeared that the alcoholic extract of white corn as used by Goldberger and his associates (*5*) and by Hoagland and Snider (*9*) would be satisfactory as a source of vitamin B, free or practically free from vitamin G.

As different investigators had used alcohols of varying strength, it seemed desirable to conduct a number of preliminary tests. These tests confirmed the findings of others in showing that extracts of white corn made either with 80- or 85-percent alcohol, by weight, were potent in vitamin B and free, or practically free, from vitamin G. Extracts made with 95-percent alcohol contained no appreciable amounts of vitamin B.

These tests indicated also that the extract of corn made with 85-percent alcohol, by weight, and dried on cornstarch so that 1 g of extract was equivalent to 4 g of corn, furnished enough vitamin B for normal growth when included as 7 percent of the diet. Feeding the extract at levels of 10 and 13 percent did not increase the rate of gains.

[4] Steenbock, H. Personal communication to authors.

While these tests were in progress, a commercial vitamin B preparation appeared on the market. In this preparation the vitamin extract was dried on cornstarch so that 5 percent in the diet should furnish adequate amounts of vitamin B. However, the results of feeding tests indicated that 5 percent of the preparation in the diet did not always furnish enough vitamin B, but that 10 percent not only furnished adequate amounts but allowed a margin of safety.

Comparative tests showed that 1 g of the commercial vitamin B preparation was equivalent to 1 g of the 85-percent alcohol extract of corn in which 1 g of extract represented 4 g of corn, and that 7 percent of either in the diet furnished enough vitamin B for normal growth. Since the commercial preparation was found satisfactory, and its use simplified the procedure, it was adopted in preference to the alcoholic extract of corn and was employed in all the tests of the meat preparations.

On the basis of these preliminary tests, the basal vitamin G-free diet adopted was the same as the basal vitamin B-free diet except that 10 percent of the commercial vitamin B preparation and 5 percent of added cornstarch replaced the 15 percent of autoclaved yeast in the vitamin B-free diet.

EXPERIMENTAL PROCEDURE

GENERAL METHOD

The general plan followed in this series of experiments, to determine the vitamin B and G potency of raw, cooked, and autoclaved meats, was the usual rat-growth procedure.

Young albino rats 28 to 30 days old, from known parentage, were placed in large cages having wire-screen bottoms, and fed as a group until growth ceased. The basal diet used for feeding was adequate, so far as known, in all food factors except the specific vitamin to be studied.

The preliminary depletion period was followed by an 8-week experimental period, during which the animals were kept in individual cages with wire bottoms, and fed the basal diet with the meat preparations included in varying percentages or in definite amounts as supplements to the basal diet. The rats had free access to distilled water. Weekly records were kept of live weights, food intake, food waste, and symptoms of vitamin deficiency.

At the end of the 8-week experimental period, all surviving animals were etherized and autopsied, and notations were made of both external and internal conditions.

Two series of feeding tests were made for vitamin B in testing the meat preparations for vitamin potency. In the first series, the dried meats were fed as varying percentages of the basal diet. This was to determine approximately how much of the dried meat was required to maintain an average growth rate of 3 g per week over a period of 8 weeks, which is the growth rate suggested by Chase and Sherman (*3*) and Borquin and Sherman (*1*) for the quantitative determination of vitamins B and G.

In the second series, the different meat preparations were fed in definite amounts separately from the basal diet and not as a percentage of the diet. A modification of the paired system of feeding was used, whereby the rats were selected and matched in sets of three, as

nearly as possible, of the same sex, age, and weight. The rats of each set of three were distributed, one each, to the groups fed the raw, cooked, and autoclaved meats. All three rats in the same set were fed a weighed amount of basal diet, restricted to the level of the rat eating the least, plus supplements of the raw- cooked, and autoclaved meat preparations, respectively. Although this adaptation of the paired feeding method has the disadvantage of keeping the food intake of some rats below the amount they would otherwise eat, it has the distinct advantage of limiting the errors in the comparison of the three meat preparations. Differences in growth of the rats on raw, cooked, and autoclaved meats can be attributed to differences in the quantitative vitamin potency of the meats, if all variables in food intake are controlled.

THE MEAT PREPARATIONS

The meats used for testing were fresh hams and fresh beef round. Enough meat was prepared at one time to carry through an entire unit of the experiment. No attempt was made to determine the origin of the meat. Two lots of both beef and pork were tested. The lean meat was separated by hand from the visible fat and connective tissue and was then ground three times through a meat grinder of the plate type and thoroughly mixed. The ground meat was divided into three portions and treated as follows:

(1) Raw meat: Dried and finely ground.

(2) Cooked meat: The ground meat was heated and stirred in a double boiler until it reached a constant temperature of approximately 90° C., then dried and finely ground.

(3) Autoclaved meat: The raw ground meat was sealed in no. 2 tin cans without water and processed for 70 minutes at 10 pounds pressure in a pressure cooker or autoclave.[5] The cans were packed as they would be packed for home canning, and not evacuated. After processing, the cans were opened and the meat was dried and finely ground. No determinations were made of the temperature attained by the meat inside the cans.

All meat samples were air-dried in glass plates on a cool air-drying apparatus designed and described by Hopper (*11*). The meat juices which escaped from the meat upon cooking were dried with the meat.

The temperature of the meat samples during drying usually ranged from 9° to 14° C. In 20 to 22 hours the meat was sufficiently dry for grinding. It was then finely ground and further dried in vacuo over sulphuric acid. The dried meats were placed in half-pint glass fruit jars, the rubbers moistened with glycerin, and the covers clamped in place. The fruit jars were then placed in vacuum desiccators and thoroughly evacuated. When the desiccators were opened, the covers automatically clamped tight, thus sealing the dried meat under partial vacuum. The meat preparations so sealed were kept in an electric refrigerator until used. At the low temperature and with the speed at which the drying was accomplished, it is believed that the minimum oxidation took place and that storage under vacuum prevented deterioration upon standing.

When the jars were opened for use, the raw dry meat was found to have retained its deep reddish-brown color and the odor of raw meat.

[5] The processing time and pressure were erroneously stated as 60 minutes and 15 pounds, respectively, in the preliminary report 4.

The cooked and autoclaved meats had a brownish-gray color and the odor of a good roast. The flavor of the different preparations was retained in the dried product.

CHEMICAL ANALYSES OF MEATS

Analyses were made of the dried pork and beef preparations to determine the water, protein, and fat content. The average composition of all samples, on a water-free basis, is given in table 1.

TABLE 1.—*Average percentages of protein and fat in water-free meat preparations raw, cooked, and autoclaved*

Meat and condition	Protein	Fat
Pork:	*Percent*	*Percent*
Raw	84.98	10.90
Cooked	85.98	10.34
Autoclaved	86.35	10.38
Beef:		
Raw	92.60	3.85
Cooked	91.75	3.74
Autoclaved	91.64	3.64

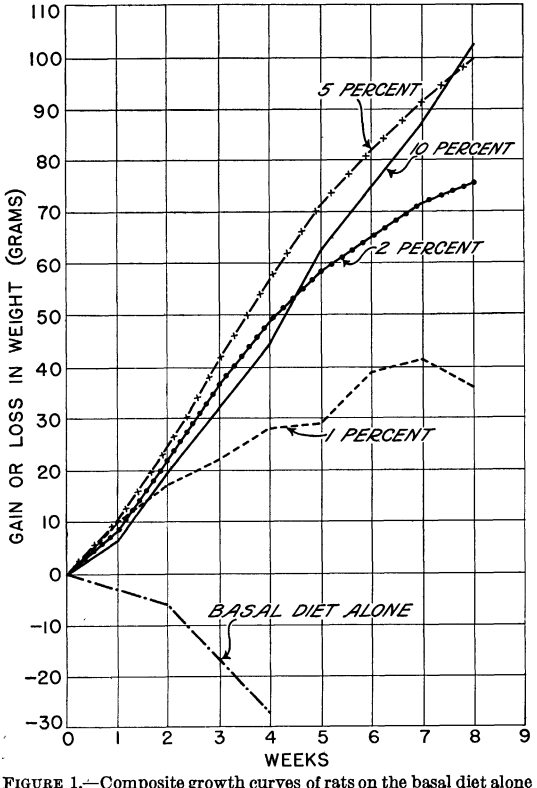

FIGURE 1.—Composite growth curves of rats on the basal diet alone and on the basal diet supplemented with different percentages of dried raw lean pork to provide vitamin B. Two rats were fed the basal diet alone, six received 1 percent, six received 2 percent, nine received 5 percent, and two received 10 percent of pork in their diet. Series 1.

Through a misunderstanding, the water content of the fresh meats in relation to the dried preparations was not determined on all preparations, but determinations on several of the different lots indicated that 1 part of dried product was equal to 3.3 to 4 parts, by weight, of the fresh meat.

VITAMIN B IN DRIED RAW, COOKED, AND AUTOCLAVED LEAN PORK

SERIES 1: DRIED PORK FED AS A PERCENTAGE OF THE DIET

To determine at what level the dried lean pork should be fed to produce the desired gain of 3 g per week during the test period, four groups of rats were fed the dried raw, cooked, and autoclaved pork as 1, 2, 5, and 10 percent of the total food intake. Adjustments were made in the protein and fat to maintain approximately a constant level of 20 percent of protein and 8 percent of fat in the diet. These adjustments for protein and fat were not made in later experiments

where the supplements were fed separately from the basal diet. Wherever the meat preparations were fed as a percentage of the diet, they replaced equal amounts of starch in the basal diet.

The composite growth curves for the groups on the raw dried pork are shown in figure 1.

It is worthy of note that the group on 10 percent of pork made no better gains than the group on 5 percent. The rats on 2 percent of pork gained approximately twice as much as those on 1 percent, but considerably less than those on 5 percent. All the rats fed the different levels of uncooked pork survived the entire 56-day period without any definite polyneuritic symptoms.

Table 2 gives the average weights, gains, and food consumption of all the different groups of rats.

TABLE 2.—*Average weights, gains in weight, and food consumption of groups of rats fed various percentages of dried lean pork, raw, cooked, or autoclaved, as a source of vitamin B in series 1*

Percentage of dried pork in diet	Rats	Average time on diet	Initial weight	Final weight	Total gain	Average per week				Gain per gram of pork fed	Gain per gram of total food
						Gain	Basal diet	Pork fed	Total food		
	No.	*Days*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
1, raw	6	56	85.5	122.0	36.5	4.6	46.1	0.47	46.6	9.79	0.099
1, cooked	[1] 5	56	85.8	118.4	32.6	4.1	46.1	.47	46.5	8.72	.088
1, autoclaved	[1] 5	56	92.0	127.2	35.2	4.4	47.3	.48	47.7	9.17	.092
1, cooked	[2] 6	52.5	87.2	108.3	21.1	2.8	43.1	.44	43.5	6.36	.064
1, autoclaved	[2] 6	52.7	91.3	114.0	22.7	3.0	43.2	.44	43.6	6.82	.069
2, raw	6	56	78.8	154.5	75.6	9.5	51.1	1.04	52.2	9.13	.182
2, cooked	6	56	76.8	150.8	74.0	9.2	51.6	1.05	52.7	8.76	.175
2, autoclaved	6	56	78.5	151.2	72.7	9.1	50.8	1.04	51.9	8.75	.175
5, raw	9	56	87.6	187.7	100.1	12.5	54.8	2.89	57.7	4.33	.217
5, cooked	8	56	80.5	180.6	100.1	12.5	55.9	2.94	58.8	4.25	.213
5, autoclaved	6	56	79.0	170.3	91.3	11.4	54.6	2.88	57.5	3.96	.198
10, raw	2	56	114.5	218.0	103.5	12.9	57.5	6.39	63.8	2.02	.202
10, cooked	2	56	74.0	176.5	102.5	12.8	53.0	5.89	58.9	2.17	.217

[1] Average for the rats that completed the full 56-day period. 1 rat in each group on the 1-percent level of cooked and autoclaved pork died of polyneuritis.
[2] Average for all rats on test.

The data of table 2 show no definite destruction of vitamin B as a result of cooking or autoclaving the pork. However, on 1 percent of dried pork in the diet, one rat in each group fed the cooked and autoclaved pork died of polyneuritis near the middle of the 56-day period. One or two other rats in each of these two groups had rough coats and showed signs of nervousness. This was interpreted as possibly due to a partial destruction of vitamin B.

The average weekly intake of pork on the 1-percent level was approximately 0.45 g, and this amount was chosen for the tests of series 2 on pork.

SERIES 2: DRIED PORK FED SEPARATELY FROM THE BASAL DIET

Eight sets of three matched rats each were fed weighed amounts of the basal diet plus 0.45 g weekly of the dried raw, cooked, or autoclaved pork. To facilitate the weighing of these small amounts of pork, it was mixed with cornstarch, so that 1.5 g of the mixture represented 0.45 g of meat. The weekly portion of supplement was weighed out at the first of the week and fed in three approximately equal feedings. The comparative gains of the rats on the different preparations are shown in figure 2.

It appears from figure 2 that cooking destroyed some of the vitamin B and that autoclaving destroyed more, but not all, of the vitamin.

All of the 24 rats fed in series 2 survived the full experimental period and none showed any definite symptoms of vitamin B deficiency. As a rule, the rats on the autoclaved pork ate the least of the basal diet. Since the food intake in each set of three matched rats was restricted to the rat eating the least, the food intake of the rats on the raw and cooked pork was thereby largely restricted to the level of those fed the autoclaved pork. The principal data for series 2 are given in table 3.

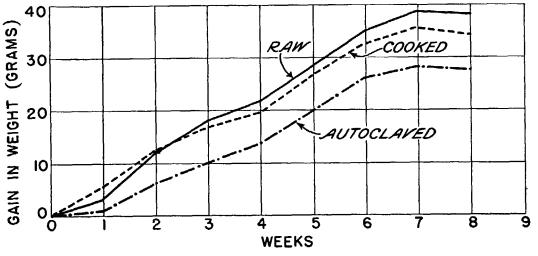

FIGURE 2.—Composite growth curves of three groups of eight rats each receiving 0.45 g of raw, cooked, and autoclaved canned lean pork, respectively, per rat weekly, fed separately from the basal diet in three equal portions to provide vitamin B. Series 2.

Table 3 shows that the food intake was virtually identical on the raw and cooked pork, but averaged about 2 g less per week on the autoclaved pork. The smaller food intake of the rats fed the autoclaved pork was probably due to a partial destruction of the appetite-stimulating vitamin B or some unknown factor. The smaller gains made by the rats on the autoclaved pork are only in part accounted for by the smaller food intake.

Since the total food intake was more accurately controlled in series 2, more weight is given to the results of these tests than to those in which the meat was fed as a percentage of the diet.

TABLE 3.—*Average weights, gains in weight, and food consumption of groups of eight rats fed three equal portions of dried pork, raw, cooked, or autoclaved, separately from their basal diet and at a level of 0.45 g per rat per week, as a source of vitamin B, in series 2 tests lasting 56 days*

Kind of pork fed	Initial weight	Final weight	Total gain	Average per week				Gain per gram of pork fed	Gain per gram of total food
				Gain	Basal diet	Pork fed	Total food		
	Grams	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
Raw	77.4	115.0	37.6	4.7	39.3	0.45	39.8	10.44	0.118
Cooked	77.4	110.8	33.4	4.2	39.7	.45	40.2	9.33	.104
Autoclaved	79.3	106.9	27.6	3.5	37.5	.45	38.0	7.78	.092

VITAMIN B IN DRIED RAW, COOKED, AND AUTOCLAVED LEAN BEEF

SERIES 1: DRIED BEEF FED AS A PERCENTAGE OF THE DIET

Thirty-seven rats were fed the vitamin B-free basal diet until depleted and then separated into groups and fed the dried raw lean beef at levels of 2, 5, 7, 10, and 15 percent of the diet to determine the amount of meat required to produce the desired gain of 3 g per week. No cooked or autoclaved beef was fed in this series.

The composite curves of gains or losses by weeks are shown in figure 3, based on the averages of the rats in each group that survived the full 8-week period.

The response of the individual rats to the different levels of beef in the diet varied considerably, especially in some groups, in spite of the fact that the rats were from the same stock and had received the same treatment. These variations are not apparent from the graphs or tables, but are significant in the interpretation of the data and, consequently, are given in some detail.

All the rats fed 2 and 5 percent of dried beef in the diet developed polyneuritis, which resulted in the death of one rat in the 2-percent group after 26 days, and two rats in the 5-percent group. One of these rats died after 8 and the other after 27 days on the test. The remaining rats in each group recovered rapidly when fed yeast.

Three of the nine rats that were started on the 7-percent level of dried beef died after 11, 16, and 19 days, respectively, on the diet. One of the remaining six was fed yeast from the fifth week, but it made only fair recovery. The five rats that survived the full 8-week period made small, but rather uniform, gains during the first 5 weeks, followed by accelerated gains during the last 3 weeks. No explanation for the accelerated gains is apparent.

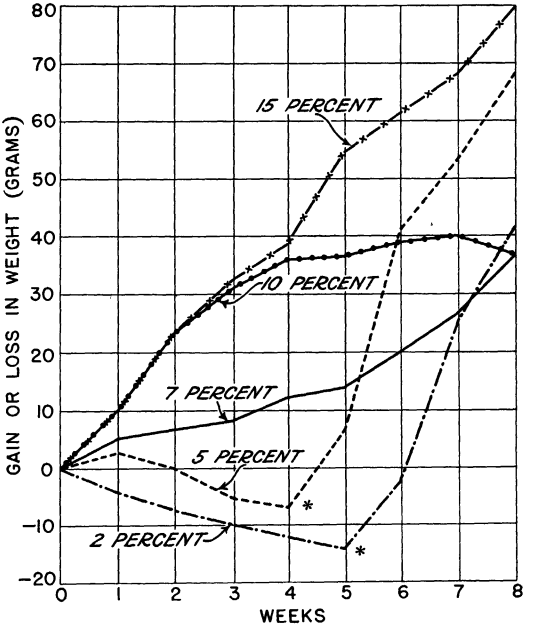

FIGURE 3.—Composite growth curves of rats fed the basal diet supplemented with different percentages of dried raw lean beef to provide vitamin B. Two rats received 2 percent, three received 5 percent, five received 7 percent, seven received 10 percent, and five received 15 percent of beef in their diet. The rats fed at the 2- and 5-percent levels were near death when yeast was added to their diets as indicated by the asterisk (*) on the graph.

Three of the twelve rats started on the 10-percent level of dried beef died after 14, 23, and 45 days, respectively, and two rats after reaching a weakened condition, improved markedly when fed yeast. The remaining seven rats, on 10 percent of beef, made virtually identical gains with those on 15 percent, during the first 4 weeks of the test period, but, as a group, they made practically no gains during the last 4 weeks. An inspection of the individual data shows that, for no apparent reason, four of the seven rats gained more or less consistently throughout the test period, reaching an average weight of 126 g as compared to 108 g reached by the five rats on 7 percent of beef and 145 g reached by the five rats on 15 percent of beef. One of the three remaining rats made only a small gain and the other two lost weight during the last half of the test period, thus making the average weight of these three only 74 g at the end of the test.

Six rats were placed on the 15-percent beef diet, but one died at 31 days, leaving five that completed the full period, with fairly steady and uniform gains throughout.

Table 4 gives a summary of the average weights, gains or losses, and food consumption of the rats at the different levels of beef feeding.

TABLE 4.—*Average weights, gains or losses in weight, and food consumption of groups of rats fed various percentages of dried raw beef as a source of vitamin B in series 1*

Raw beef in diet (percent)	Rats	Average time on diet	Initial weight	Final weight	Total gain or loss	Average per week				Gain or loss per gram of beef	Gain or loss per gram of total food
						Gain or loss	Basal diet	Beef fed	Total food		
	Number	*Days*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
2	[1] 3	29.7	73.0	55.0	−18.0	−4.3	29.0	0.6	29.6	−7.17	−0.145
5	[2] 7	23.9	63.4	58.4	−5.0	−1.9	32.2	1.7	33.9	−1.12	−.056
7	[3] 5	56.0	70.0	107.6	37.6	4.7	42.3	3.2	45.5	1.47	.103
10	[4] 7	56.0	65.9	102.4	36.5	4.6	39.6	4.4	44.0	1.05	.105
15	[5] 5	56.0	65.5	145.0	79.4	9.9	44.0	7.8	51.8	1.27	.191

[1] Average of all rats in group up to time of feeding yeast; 1 of the 3 rats died before yeast was fed.
[2] Average of all rats in group up to time of feeding yeast; 2 of the 7 rats died before yeast was fed.
[3] 9 rats started; 3 died and 1 was fed yeast; averages are for remaining 5 rats.
[4] 12 rats started; 3 died and 2 were fed yeast; averages are for remaining 7 rats.
[5] 6 rats were started but 1 died; averages are for remaining 5 rats.

From the results obtained, it was difficult to determine how much beef would be required to furnish enough vitamin B to maintain an average gain of 3 g per week, but, apparently, 7 percent in the diet, or 3.2 g of dried beef per week, was about the lower limit.

SERIES 2: DRIED BEEF FED SEPARATELY FROM THE BASAL DIET

Sets of three matched rats each were fed on equal weighed portions of dried raw, cooked, and autoclaved beef, fed separately from the basal diet to insure more uniform consumption of the beef. The basal food intake was also kept as uniform as possible, but it was found that the rats on the autoclaved beef almost invariably decreased their food intake as the period progressed. Whenever a rat on the autoclaved beef ate less than 3 g of food per day, it was dropped from the set and the food intake equalized between the rats on the raw and cooked beef. All the rats relished the raw and cooked beef, but those on the autoclaved beef showed a consistent dislike for it. Some of the autoclaved beef was frequently left uneaten or wasted from the food cup. It was, therefore, impossible to keep the food intake alike for all the rats.

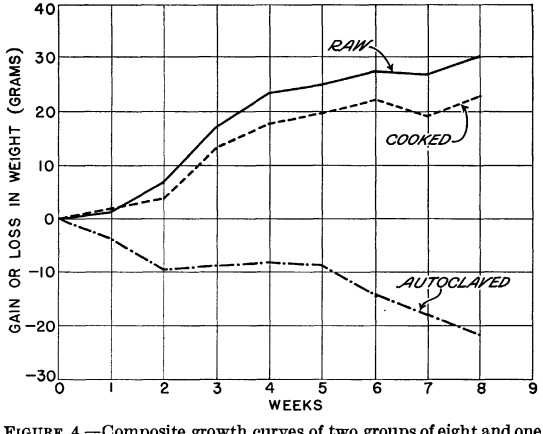

FIGURE 4.—Composite growth curves of two groups of eight and one group of nine rats receiving 4.5 g of raw, cooked, and autoclaved canned lean beef, respectively, per rat weekly, fed separately from the basal diet in three equal portions to provide vitamin B. Series 2.

No such difficulty existed with the pork, possibly due to the fact that the pork was fed in small amounts (less than 0.5 g per week) mixed with cornstarch, or to the larger amounts of vitamin B contained in the autoclaved pork as compared to the beef, or to a loss of the appetite-stimulating factor, vitamin B. The last-mentioned seems the most likely explanation, for when adequate amounts of

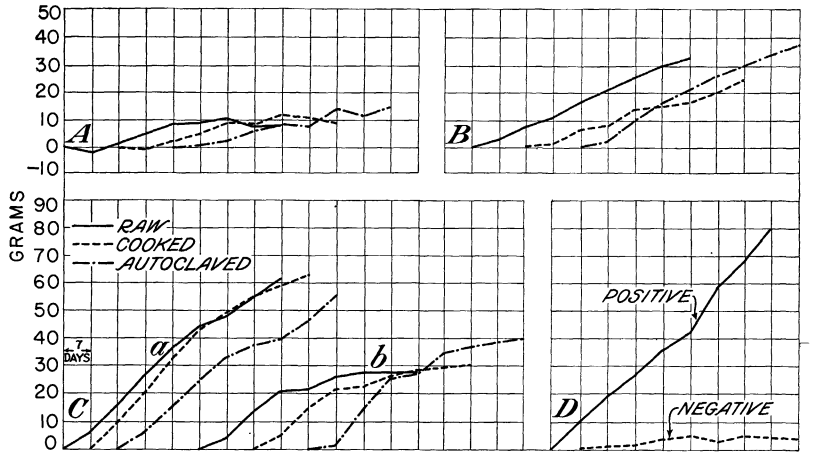

FIGURE 5.—Composite growth curves of rats receiving various quantities of dried pork added separately to their diets to provide vitamin G; *A*, three groups of eight rats each, fed 1 g of pork per rat per week; *B*, three groups of five rats each, fed 1.5 g of pork per rat per week; *C*, three groups of six rats each in series 1 (*a*) and three rats each in series 2 (*b*), fed 2 g pork per rat per week; *D*, groups of four rats on positive and six on negative control diet.

vitamin B were furnished, as in testing for vitamin G, the rats ate the autoclaved beef readily.

Because of the results of series 1, levels of 4, 4.5, and 6 g of dried beef weekly were chosen for series 2 in testing the relative potency of the three preparations for vitamin B.

The composite weight curves of the rats fed 4.5 g of beef per week are shown in figure 4.

TABLE 5.—*Average weights, gains or losses in weight, and food consumption of groups of rats fed three equal portions of dried beef, raw, cooked, or autoclaved, separately from their basal diet and at levels of 4, 4.5, and 6 g per rat per week, as a source of vitamin B in series 2*

Amounts and kind of beef fed weekly	Rats	Average time on diet	Initial weight	Final weight	Total gain or loss	Average per week				Gain or loss per gram of beef fed	Gain or loss per gram of total food
						Gain or loss	Basal diet	Beef fed	Total food		
	Number	*Days*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
4 g:											
Raw	5	56	89.0	112.0	23.0	2.9	32.5	4.0	36.5	0.73	0.079
Cooked	5	56	88.2	118.4	30.2	3.8	33.4	4.0	37.4	.95	.102
Autoclaved	5	39	87.6	60.6	−27.0	−5.7	21.2	2.7	23.9	−2.11	−.238
4.5 g:											
Raw	8	54.3	80.9	111.0	30.1	3.9	33.3	4.5	37.8	.87	.103
Cooked	8	54.3	81.4	104.4	23.0	3.0	31.9	4.5	36.4	.67	.082
Autoclaved	9	50.0	84.3	67.3	−17.0	−3.8	23.1	3.9	27.0	−.97	−.141
6 g:											
Raw	2	56.0	72.5	118.5	46.0	5.8	35.2	6.0	41.2	.97	.141
Cooked	2	56.0	69.5	112.5	43.0	5.4	33.7	6.0	39.7	.90	.136
Autoclaved	2	50.5	71.0	57.0	−14.0	−2.1	20.9	4.2	25.1	−.50	−.084

Figure 4 shows that the rats on the cooked beef did not gain as well as those on the raw beef, and also that the ones on the autoclaved beef lost weight from the start. The average results for all the groups at the different levels of beef feeding are shown in table 5.

The small average weekly gains made by the rats on 4 g of dried raw beef per week are mainly due to the fact that one rat wasted food and lost weight; the other four averaged 3.7 g gain per week, which is practically the same as the gains made by the rats on the cooked beef. Of the five rats on the autoclaved beef, one made a small gain of 3 g in 8 weeks; the other four lost from 32 to 38 g.

Of the eight rats fed 4.5 g of raw beef weekly, one lost 2 g, but the others gained from 28 to 40 g each. Similarly, one of the rats on the cooked beef lost 5 g, while the others gained from 9 to 37 g. If the ones that lost weight are omitted, the average weekly gains per rat are 4.5 g on the raw, and 3.5 g on the cooked beef.

All rats fed 6 g of raw and cooked beef weekly survived the entire 8-week period and made larger average weekly gains than the groups fed at the lower levels. Both of the rats fed the autoclaved beef lost weight. One of the rats died at the end of 45 days on the diet.

With one or two exceptions on the 4- and 4.5-g levels, all the rats on the autoclaved beef lost weight, but evidently the autoclaved beef still contained some vitamin B, since the losses in weight decreased and the survival period increased in the successive groups with the increase in level of beef feeding.

UNITS OF VITAMIN B IN DRIED LEAN PORK AND BEEF

The units of vitamin B in the dried lean pork and beef, computed according to the method of Chase and Sherman (*3*), are presented in table 6.

UNITS OF VITAMIN B PER GRAM OF DRIED LEAN PORK

The units of vitamin B in pork, as given in table 6, were computed only at the 1- and 2-percent levels and 0.45 g weekly, as the gains on the 5- and 10-percent levels were too large to furnish a reliable measure.

In series 1, where the pork was fed as a percentage of the diet, there is no clear-cut or definite indication of a destruction of vitamin B as a result of either cooking or autoclaving. However, in series 2, where the pork was fed separately from the basal diet, and where the food was equalized more closely in the different sets of three, there appears to be a rather definite indication that some of the vitamin was destroyed as a result of cooking and autoclaving. From the values found in series 2 alone, it appears that approximately 10 percent of vitamin B was destroyed as a result of cooking and 25 percent as a result of autoclaving the pork. Slightly different results are obtained when the rats on the 1-percent level of pork are included. The weighted average for the rats fed at the 1-percent level of dried pork and those fed 0.45 g per week, separately from the basal diet, is 24 units per gram of dried raw pork. This is equivalent to approximately 7 units per gram of fresh pork. For the cooked pork, the weighted average is 21 units and for the autoclaved pork, it is 19 units per gram of dried product.

TABLE 6.—*Units of vitamin B in variously treated dried pork and beef, as computed from the data by the method of Chase and Sherman*

PORK

Series, treatment of meat used, and quantity fed	Rats	Average time on diet	Average weekly gains or losses in weight	Vitamin B per gram of meat
Series 1:	*Number*	*Days*	*Grams*	*Units*
1 percent, raw	[1] 6	56.0	4.6	22.8
1 percent, cooked	[2] 5	56.0	4.1	20.5
1 percent, autoclaved	[2] 5	56.0	4.4	21.2
1 percent, cooked	[1] 6	52.5	2.8	14.8
1 percent, autoclaved	[1] 6	52.7	3.0	15.9
2 percent, raw	6	56.0	9.5	21.2
2 percent, cooked	6	56.0	9.2	20.2
2 percent, autoclaved	6	56.0	9.1	20.3
Series 2:				
0.45 g, raw, weekly	8	56.0	4.7	24.5
0.45 g, cooked, weekly	8	56.0	4.2	21.9
0.45 g, autoclaved, weekly	8	56.0	3.5	18.2

BEEF

Series 1:				
7 percent, raw	[3] 5	56.0	4.7	3.4
10 percent, raw	[4] 7	56.0	4.6	2.4
15 percent, raw	[2] 5	56.0	9.9	3.0
Series 2:				
4.0 g, raw, weekly	5	56.0	2.9	1.7
4.0 g, cooked, weekly	5	56.0	3.8	2.2
4.0 g, autoclaved, weekly	5	39.0	−5.7	----------
4.0 g, raw, weekly	[5] 4	56.0	3.7	2.2
4.5 g, raw, weekly	8	54.3	3.9	2.0
4.5 g, cooked, weekly	8	54.3	3.0	1.6
4.5 g, autoclaved, weekly	9	50.0	−3.8	----------
4.5 g, raw, weekly	[5] 7	56.0	4.5	2.3
4.5 g, cooked, weekly	[5] 7	56.0	3.5	1.8
6.0 g, raw, weekly	2	56.0	5.8	2.3
6.0 g, cooked, weekly	2	56.0	5.4	2.1
6.0 g, autoclaved, weekly	2	50.5	−2.1	----------

[1] Averages for all rats in the group.
[2] Average, omitting 1 rat that died in each group.
[3] Average of 5 rats out of 9 that lived through entire period without modification of diet.
[4] Average of 7 rats out of 12 that lived through entire period without modification of diet.
[5] Average, omitting 1 rat in each group that lost weight.

UNITS OF VITAMIN B PER GRAM OF DRIED LEAN BEEF

The units per gram of dried beef in series 1 are slightly higher than the units as determined in series 2. In series 2, at the level of 4 g of dried beef per week, there is no definite indication of a destruction of vitamin B, but at the 4.5-g level there appears to be a destruction of vitamin B as a result of cooking. At the 6-g level, there is also no definite indication of a destruction of vitamin B as a result of cooking, but at all three levels, there is definite indication that autoclaving destroyed at least a part of the vitamin contained in the beef. The dried lean raw beef contained an average of about 2.5 units per gram, which is equivalent to 0.6 unit per gram of fresh beef. From this it is evident that the lean beef contained only about one-tenth as much vitamin B as the pork.

It is not possible from the data to explain why autoclaving destroyed the vitamin B more completely in the lean beef than in the lean pork. The data of table 1 show that the lean pork contained about three times as much fat as the lean beef, and it is suggested that the fat may possibly have aided in protecting the vitamin from destruction.

VITAMIN G IN DRIED RAW, COOKED, AND AUTOCLAVED LEAN PORK

Pork was fed at levels of 1, 1.5, and 2 g per week, as preliminary tests had shown, these levels to be suitable for testing for vitamin G. The pork for the entire week was weighed out in advance and fed as a supplement separately from the basal diet in approximately three equal portions during the week.

Composite growth curves for the different groups are shown in figure 5, and a summary of the data by groups, including the controls, is given in table 7.

TABLE 7.—*Average weights, gains or losses in weight, and food consumption of groups of rats fed dried pork, raw, cooked, or autoclaved, separately from their basal diets and at levels of 1, 1.5, and 2 g per rat per week, as a source of vitamin G*

Amounts and treatment of pork fed weekly for period and series indicated	Rats	Average time on diet	Initial weight	Final weight	Total gain or loss	Average per week				Gain per gram of pork fed	Gain or loss per gram of total food
						Gain or loss	Basal diet	Pork fed	Total food		
1 g, September to November 1932:	*Number*	*Days*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
Raw	8	56	67.9	75.9	8.0	1.0	33.7	1.0	34.7	1.00	0.029
Cooked	8	54	66.1	75.7	9.6	1.2	33.6	1.0	34.6	1.20	.034
Autoclaved	8	56	67.5	81.5	14.0	1.7	33.1	1.0	34.1	1.70	.050
Negative controls	3	56	50.3	73.3	23.0	2.9	33.3	.0	33.3	-------	.087
Positive controls	2	56	53.5	117.0	63.5	8.0	43.6	1.0	44.6	8.00	.179
1.5 g, April to July 1933:											
Raw	5	56	38.8	70.8	32.0	4.0	30.6	1.5	32.1	2.67	.125
Cooked	5	56	38.6	62.2	23.6	3.0	27.7	1.5	29.2	2.00	.103
Autoclaved	5	56	38.2	75.0	36.8	4.6	30.9	1.5	32.4	3.07	.142
Negative control	1	56	57.0	69.0	12.0	1.5	29.8	.0	29.8	-------	.050
2 g, series 1, June to August 1932:											
Raw	6	56	60.1	121.5	61.3	7.7	37.8	2.0	39.8	3.85	.193
Cooked	6	56	59.3	121.8	62.5	7.8	38.6	2.0	40.6	3.90	.192
Autoclaved	6	56	60.5	116.0	55.5	6.9	36.4	2.0	38.4	3.45	.180
Negative controls	2	56	56.0	55.5	−.5	−.1	26.6	.0	26.6	-------	−.004
Positive controls	2	56	45.0	139.5	94.5	11.8	38.2	3.5	41.7	3.37	.283
2 g, series 2, July to September 1933:											
Raw	3	56	63.3	90.6	27.3	3.4	29.2	2.0	31.2	1.70	.109
Cooked	3	56	63.0	93.2	30.3	3.8	31.1	2.0	33.1	1.90	.115
Autoclaved	3	56	63.0	103.0	40.0	5.0	31.2	2.0	33.2	2.50	.151
Negative control	1	56	70.0	75.0	5.0	.6	32.7	.0	32.7	-------	.018

It was not possible to conduct all the feeding trials at the different levels simultaneously. Series 1 at the level of 2 g of dried pork weekly was carried out during the first summer, followed by the group on 1 g of pork weekly during the fall of the same year. The following spring the tests at the 1.5-g level were made, followed by series 2 on the 2-g level during the summer.

The data of table 7 show that 1 g of the dried pork preparations failed to support a unit gain of 3 g per week, and that 1.5 g gave slightly more than a unit rate of gain per week. The gains on 2 g of pork in series 1 were nearly twice as great as the gains on 1.5 g, but in series 2 at the same level of pork as in series 1, the gains were similar to those on the 1.5 g of pork. At first it might appear that a loss in vitamin potency had occurred between the tests of series 1 the first summer, and series 2 the following summer; but this seems unlikely

since the tests at the 1.5-g level were made just prior to the tests of series 2. Moreover, no decrease in potency due to storage was observed in the beef under similar circumstances.

The larger food intake by the rats in series 1 as compared to series 2 may explain in part the differences in the gains of the rats in the two series, but it does not explain why the rats on 1.5 g of pork made virtually the same gains as those on 2 g in series 2 where the food intake was practically the same. Possibly differences in the state of depletion of the rats at the start was a factor.

In spite of some inconsistencies in the gains on the raw, cooked, and autoclaved pork in the different groups, and on the different levels of intake, the data fail to show any destruction of vitamin G in the pork when cooked or autoclaved by the methods here used.

VITAMIN G IN DRIED RAW, COOKED, AND AUTOCLAVED LEAN BEEF

The work of Hoagland and Snider (*10*) had indicated that pork and beef have about the same vitamin G potency. With this in

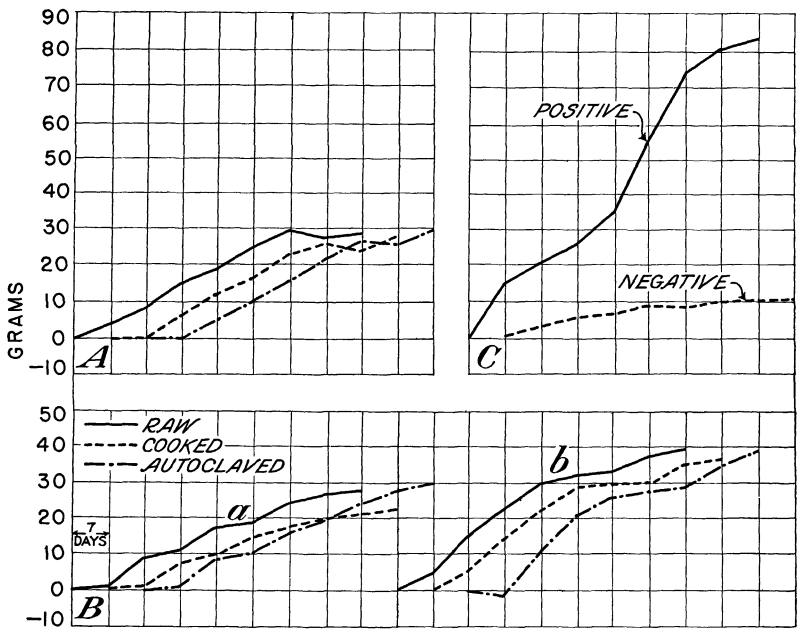

FIGURE 6.—Composite growth curves of rats receiving dried lean beef added separately to their diets to provide vitamin G: *A*, Three groups of 10 rats each fed 1.5 g of beef per rat per week; *B*, three groups of 10 rats each in series 1 (*a*) and 3 rats each in series 2 (*b*) fed 2 g of beef per rat per week; *C*, 1 rat on positive and group of 6 rats on negative control diet

mind the writers fed dried beef only at levels of 1.5 and 2 g per week, as 1 g of pork had been found inadequate.

The composite growth curves of the rats fed in these tests are shown in figure 6, and a summary of the gains and food intake is given in table 8.

TABLE 8.—*Average weights, gains or losses in weight, and food consumption of groups of rats fed dried beef, raw, cooked, or autoclaved, separately from their basal diets and at levels of 1.5 and 2 g per rat per week, as a source of vitamin G*

Amounts and treatment of beef fed weekly for period and series indicated	Rats	Average time on diet	Initial weight	Final weight	Total gain or loss	Average per week				Gain per gram of beef fed	Gain or loss per gram of total food
						Gain or loss	Basal diet	Beef fed	Total food		
1.5 g, March to May 1933:	*Number*	*Days*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
Raw	10	56	52.0	80.6	28.6	3.6	33.9	1.5	35.4	2.40	0.102
Cooked	10	56	52.4	79.5	27.1	3.4	34.0	1.5	35.5	2.27	.096
Autoclaved	10	56	52.3	81.5	29.2	3.7	33.7	1.5	35.2	2.47	.105
Negative controls	4	56	52.5	65.0	12.5	1.6	31.0	.0	31.0	-------	.052
2 g, series 1, December to February 1932–33:											
Raw	10	56	63.0	90.1	27.1	3.4	34.4	2.0	36.4	1.70	.093
Cooked	10	56	61.7	84.2	22.5	2.8	33.9	2.0	35.9	1.40	.078
Autoclaved	10	56	61.4	90.2	28.8	3.6	33.8	2.0	35.8	1.80	.101
Negative controls	1	56	34.0	45.0	11.0	1.4	26.5	.0	26.5	-------	.053
Positive controls	1	56	47.0	130.0	83.0	10.4	51.1	2.0	53.1	5.20	.196
2g, series 2, July to September 1933:											
Raw	3	56	59.7	98.7	39.0	4.9	32.7	2.0	34.7	2.45	.141
Cooked	3	56	61.0	97.3	36.3	4.5	32.7	2.0	34.7	2.25	.130
Autoclaved	3	56	62.0	100.3	38.3	4.8	32.2	2.0	34.2	2.40	.140
Negative control	1	56	44.0	42.0	−2.0	−.3	21.3	.0	21.3	-------	−.014

The 10 sets of 3 matched rats each fed 1.5 g of dried beef per week separately from the basal diet (table 8) were fed simultaneously with the 5 sets of 3 rats on 1.5 g of dried pork (table 7). The data show that the rats on the raw, cooked, and autoclaved beef gained 3.6, 3.4, and 3.7 g, respectively, as compared to gains of 4.0, 3.0, and 4.6 g by the rats on the pork. These results indicate that the vitamin G potency of the beef and pork preparations was practically the same.

Although there were considerable variations in the rates of gain among the groups, the results indicate no loss of vitamin G potency as a result of either cooking or autoclaving.

UNITS OF VITAMIN G IN DRIED LEAN PORK AND BEEF

The units of vitamin G in the several feeding periods and at the different levels of feeding have been computed according to the method of Borquin and Sherman (*1*), and are given in table 9.

The units of vitamin G per gram of dried pork and beef in table 9 vary considerably between different levels of feeding and between series on the same level, but a comparison of the units per gram of meat fails to show any definite destruction of vitamin G as a result of cooking or autoclaving. Using the data from all the rats fed at the 1.5 and 2 g levels, the weighted average units of vitamin G per gram of dried meat are: 6.9 for the raw, 6.5 for the cooked, and 7.3 for the autoclaved pork, as compared to 4.9 for the raw, 4.4 for the cooked, and 5.1 for the autoclaved beef.

TABLE 9.—*Units of vitamin G in variously treated dried pork and beef as computed from the data by the method of Borquin and Sherman*

PORK

Series, treatment of meat used, and quantity fed weekly	Rats	Average time on diet	Average weekly gains in weight	Vitamin G per gram of meat
	Number	*Days*	*Grams*	*Units*
1 g, raw	8	56	1.0	2.3
1 g, cooked	8	54	1.2	2.8
1 g, autoclaved	8	56	1.7	4.0
1.5 g, raw	5	56	4.0	6.2
1.5 g, cooked	5	56	3.0	4.7
1.5 g, autoclaved	5	56	4.6	7.2
Series 1:				
2 g, raw	6	56	7.7	9.0
2 g, cooked	6	56	7.8	9.1
2 g, autoclaved	6	56	6.9	8.1
Series 2:				
2 g, raw	3	56	3.4	4.0
2 g, cooked	3	56	3.8	4.4
2 g, autoclaved	3	56	5.0	5.8

BEEF

1.5 g, raw	10	56	3.6	5.6
1.5 g, cooked	10	56	3.4	5.3
1.5 g, autoclaved	10	56	3.7	5.8
Series 1:				
2 g, raw	10	56	3.4	4.0
2 g, cooked	10	56	2.8	3.3
2 g, autoclaved	10	56	3.6	4.2
Series 2:				
2 g, raw	3	56	4.9	5.7
2 g, cooked	3	56	4.5	5.2
2 g, autoclaved	3	56	4.8	5.6

SUMMARY OF RESULTS

The main facts brought out in these studies on pork and beef as are follows:

(1) The dried raw lean pork used in the tests was a good source of vitamin B, containing about 24 units per gram of dried material. This is equivalent to about 7 units per gram of fresh pork. The dried raw lean beef contained 2.5 units of vitamin B per gram, equivalent to 0.6 unit per gram of fresh beef. On this basis the fresh lean beef contained less than one-tenth as much vitamin B as the pork.

(2) The lean pork cooked at a temperature not exceeding 90° C. contained 21 units of vitamin B per gram of dried material, representing a loss of 12 percent in potency. Lean beef cooked in the same way contained 2 units per gram of dried material, representing a loss in potency of 20 percent.

(3) Heating the pork in a steam autoclave or pressure cooker for 70 minutes at 10 pounds' pressure destroyed more of the vitamin B than did cooking. The autoclaved pork had a value of about 19 units per gram of dried material, representing a loss in potency of approximately 21 percent. Under similar treatment the vitamin B in the beef was almost completely destroyed.

(4) The vitamin G content of lean pork and beef is nearly the same. The average units per gram of dried material for pork are: Raw 6.9, cooked 6.5, and autoclaved 7.3. For dried beef the units are: Raw 4.9, cooked 4.4, and autoclaved 5.1. The data indicate no destruction of vitamin G as a result of cooking or autoclaving.

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