

VITAMIN CONTENT OF HONEY AND HONEYCOMB¹

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INTRODUCTION

Honey has been considered a valuable food since earliest times. Consisting as it does of a mixture of dextrose and levulose it is easily digested, and this fact may account in part for the good results obtained when it is used in the diet, especially in infant feeding. Since honey can be used to advantage in the diet of infants the question arose as to whether it might not be a source of some or all of the vitamins. A review of the literature revealed the fact that very little work had been done in the way of determining quantitatively the vitamin content of honeys.

REVIEW OF LITERATURE

Dutcher (3)² determined the vitamin B content of honey obtained while basswood and white clover were in full bloom, using pigeons in his work. The tests were made by absorbing the vitamin of the honey on Lloyd's reagent and feeding amounts equivalent to 45 gm. of honey. Nectar was tested in the same manner. Dutcher concluded that the strained honey contained a negligible amount of vitamin B and that there was little evidence of its presence in nectar.

In 1919 Bachman (1) found that 25 c. c. of a strained honey added to 75 c. c. of water and used in Nagel's solution did not furnish the vitamin necessary for the growth of yeast.

Faber (4) in 1920 made a study of the antiscorbutic value of a white-sage comb honey which was extracted before using. Guinea pigs were used, and all of them exhibited characteristic scurvy symptoms when fed a solution of 1 part of honey to 15 parts of water, which was later increased to 1 part of honey to 5 parts of water. The quantity of honey consumed ranged from 0.88 to 5.58 c. c. of honey per 100 gm. of initial body weight. Faber concluded that it was "probable" that honey contained no antiscorbutic vitamin.

Hawk, Smith, and Bergeim (5) determined the vitamin A, B, and C content of blended honey, white-clover honey, and honeycomb. For vitamin B their method consisted in feeding three groups of rats, respectively, (1) a diet free from vitamin B; (2) one in which blended honey replaced part of the starch; and (3) one in which white-clover honey replaced part of the starch. At the end of four weeks the diets were changed. Group 1 was divided and half the rats were given blended honey and the other half white-clover honey. After another two weeks all were given milk. From the results obtained, Hawk and his associates concluded that there was a small amount of vitamin B present in these honeys. Following a similar procedure for the

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² Reference is made by number (italic) to "Literature cited," p. 366.

vitamin A determination, they found that strained honey contained no vitamin A, whereas a definite but minimal amount was present in the comb honey. For the vitamin C test they fed three groups of guinea pigs in the same way; that is, they gave one group the scorbutic diet, a second group the same diet with blended honey to replace a part of the starch, and a third group the same diet with clover honey to replace the starch. All developed scurvy within two weeks, showing that the honeys contained no vitamin C.

In 1922 Luttinger (6) gave a general report of his findings on the use of honey in infant feeding in which he states that he found vitamins A, B, C present in 82 per cent of the honey examined. No experimental evidence was presented to bear out this statement.

Scheunert, Schieblich, and Schwanebeck (8), in 1923, examined three samples of honey for vitamins A, B, C, and concluded that none of the samples contained vitamins.

Caillas (2), in 1925, reported work done with pigeons which seemed to show that fresh honey contained vitamin B. The number of birds used, however, was too small to make the results very convincing.

DESCRIPTION OF HONEYS TESTED

Since it was out of the question to make an exhaustive examination of honeys of all the principal floral sources, three samples representing the extremes of color variation were chosen for investigation. None of the honeys had been heated as is often the case with extracted honey. Honey No. 1 was a white-clover honey from Grover Hill, Ohio. This was in a granular state when received. Honey No. 2 was a buckwheat honey, very dark in color, produced near Varysburg, N. Y. Honey No. 3 was a light-colored white-clover honey from Middlebury, Vt. It was drained from the comb and the comb was pressed as free from adhering honey as possible and was also used in feeding tests.

EXPERIMENTAL DATA

VITAMIN A DETERMINATIONS

The method used for vitamin A determinations was essentially that of Sherman and Munsell (11) with a few modifications.

The basal diet consisted of casein (purified), 18 per cent; starch, 67 per cent; brewery yeast, 10 per cent; Osborne and Mendel salts, 4 per cent; table salt, 1 per cent. The diet was irradiated with the light from a mercury vapor quartz lamp to insure an abundance of vitamin D. The rats were fed the vitamin-A-free diet until stationary or declining weight and appearance of symptoms due to vitamin A deficiency indicated that their body stores of vitamin A were depleted. As soon as the rats were in a suitable condition to be used for tests they were weighed and placed in individual cages. A weighed amount of the vitamin-A-free food was given to each rat and the honey was fed as a daily supplement to this diet.

Honeys No. 1 and No. 2 were fed in amounts of 1, 2, and 3 gm. per day. The plan of feeding daily portions of honey to the rats required a great deal of time. For this reason honey No. 3 and the honeycomb were incorporated in the basal diet in place of 30 per cent of the starch. In each litter one or more animals were designated as controls and received only the basal diet during the test period. The test period was continued for eight weeks, or until it was terminated by

the death of the rat. If the rat did not live out the eight weeks the last recorded weight is that of the dead rat. Autopsies were performed on all animals to determine whether the gross pathological lesions shown by animals confined to a vitamin-A-free diet were present. Table 1 gives the weights and survival periods of the rats used for these tests. Curves showing the changes in weight made by averaging results from the groups of test animals are presented in Figure 1.

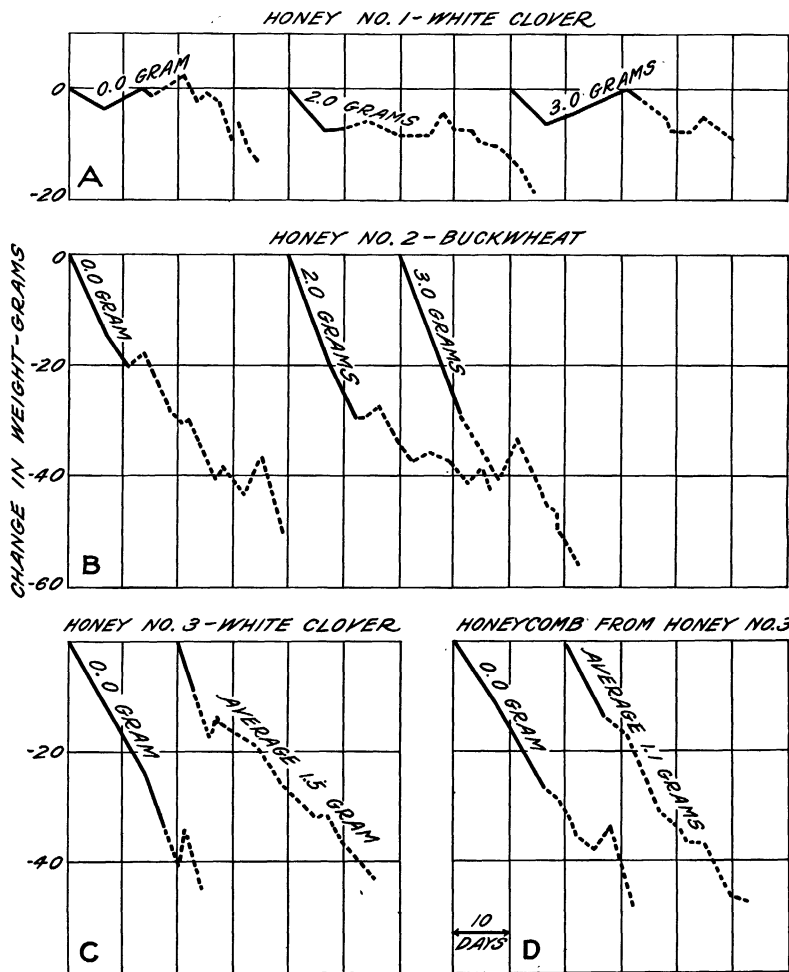


FIGURE 1.—Curves showing changes in weight made during the test period by groups of rats fed honey or honeycomb as the sole source of vitamin A. Each curve is the average result of several tests. The amount of honey or honeycomb received by each rat six times per week is indicated on each curve. The change in weight for the group is represented by a solid line to the point where the death of the first animal occurred. The broken line represents the averages for the surviving animals until all had died

None of the rats receiving the honey or honeycomb lived out the full eight weeks of the test period, nor did they live on an average any longer than the control rats. In all cases the rats fed honey and honeycomb exhibited as severe pathological lesions as those that received no honey in addition to the basal diet.

TABLE 1.—Weight records of rats fed honey and honeycomb as the sole source of vitamin A—Continued

LIGHT-COLORED WHITE-CLOVER HONEY (NO. 3)

Quantity of honey fed per rat per day, 6 days per week (grams)	Rat No.	Weight of rats at age of 4 weeks	Weight of rats when feeding of honey was begun	Weight of rats at end of successive weeks of test period								Period of survival
				1	2	3	4	5	6	7	8	
0	2908	58	114	102	93	80						65
	3086	57	131	108	95	79						65
	3089	52	114	105	93	80	69					69
	3092	49	110	105	92	70						65
	Average											
Average, 1.5 (30 per cent of diet)	2903	63	137	101								54
	2904	62	120	113	109	89						67
	2906	57	134	94								54
	2907	58	98	87								51
	2909	55	118	109	96	94	76					76
	2910	54	115	85	95	85	84	72				83
	2911	50	106	82								54
	3085	57	125	118	98	94	80					70
	3087	50	104	87	84							53
	3088	56	123	105	108	90	92					72
	3090	51	105	95	90	78	76	64				76
3091	50	117	100	87	80						66	
3093	40	100	94	91	83	63					72	
Average												65.2

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0	3128	58	114	107	107	83	72					65
	3129	57	117	110	88	74						62
	3131	54	112	100	94	86	88	69				74
	3141	70	153	144	132	116	97					68
	3142	64	136	128	119	101	100	83				75
	3145	57	111	105	102	75	65					66
	3444	59	145	126	97	90						58
	3445	57	124	101	90	84						60
	3448	52	110	105	89	85	69					70
	3450	44	102	94	84	67	66					64
Average												66.2
Average, 1.1 (20 per cent of diet)	3125	61	134	112	100	86	84					64
	3127	59	122	110	107	86	72					70
	3130	56	114	105	92	78	74					67
	3132	47	106	96	82	63						59
	3144	60	130	119	96	95	84	76				73
	3143	54	111	107	94	74						62
	3146	57	117	110	92		85	70				76
	3446	55	120	80								49
	3447	52	114	95	86							53
3449	52	103	100	85	79	65					67	
Average												64.0

These results indicate that no one of these three samples of honey nor the honeycomb contained an amount of vitamin A that could be detected by the method used for measuring this factor.

VITAMIN B DETERMINATIONS

The determination of the vitamin B content of the three samples of honey was completed before the multiple nature of vitamin B had been generally recognized. The method used was that of Sherman and Spohn (12), which makes no distinction between the two vitamin B factors. All rats were kept in cages having raised screen bottoms and were given a basal diet of casein (purified) 18 per cent, starch 68 per cent, butterfat 8 per cent, cod liver oil 2 per cent, Osborne and Mendel salts 4 per cent. Honey No. 1 was fed in amounts of

TABLE 2.—Weight records of rats fed honey and honeycomb as the sole source of vitamin B—Continued

BUCKWHEAT HONEY (NO. 2)

Quantity of honey fed per rat per day, 6 days per week (grams)	Rat No.	Weight of rats at age of 4 weeks	Weight of rats at end of successive weeks of test period								Period of survival	
			1	2	3	4	5	6	7	8		
0	1961	45	41	41	36	30	26					34
	1965	41	40	36	31	25						26
	1972	41	39	37	33	27						28
	1985	35	34	31	27	25						24
	1989	32	32	29	26	21	19					30
	Average											
Average, 0.7 (30 per cent of diet).	1962	44	41	42	38	33	27	26				38
	1963	42	41	38	35	31	25					34
	1964	48	45	44	41	35	29	27				38
	1965	46	41	37	35	31	28	28				39
	1966	45	40	39	35	30	26					33
	1967	42	40	38	35	27	25					31
	1969	32	30	28	27	23	20					35
	1970	44	39	37	33	25	22					32
	1971	44	41	39	35	29	24	24				36
	1973	41	38	36	33	29	25	25				36
	1974	39	38	34	31	26	22		25			33
	1984	35	30	31	27	23	21					30
	1986	34	33	34	30	25	21					30
	1987	33	30	30	27	21						28
	1988	32	32	30	28	24	23					32
1990	37	35	32	29	28	23					35	
1991	35	31	31	27	23	21					31	
1992	34	31	32	29	24	21					30	
Average												33.4

LIGHT COLORED WHITE-CLOVER HONEY (NO. 3)

0	3010	49	49	45	41	34	30					31
	3014	45	46	43	38	32	27					31
	3015	45	45	42	36	31	29					30
	3072	52	50	48	40	36	32					32
	3075	52	48	44	43	38	33	28				38
	3079	45	41	38	38	30	29					29
	3081	45	41	37	36	30	26					31
	Average											
Average, 0.9 (30 per cent of diet).	3009	50	52	49	44	38	33					31
	3011	48	50	49	41	34	33					30
	3012	47	47	45	40	33	30					30
	3013	49	50	47	42	35	31					30
	3016	45	47	44	40	32	30					30
	3071	55	56	51	46	39	36					35
	3073	52	52	50	44	38	35					31
	3074	53	49	49	43	35	32					30
	3077	48	46	44	42	38	33					31
	3078	48	43	42	38	33	29					31
3080	45	40	39	34	31	27					31	
Average												30.9

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Average, 0.5 (20 per cent of diet).	3173	50	48	45	39	34	31	29				40
	3174	50	48	45	38	33	29					34
	3175	47	43	42	35	32	28					34
	3176	45	44	41	36	30	28					33
	3177	48	45	42	35	31	28					33
	3178	47	45	41	34	31						27
	3179	46	44	39	34	31						28
Average												32.7

No one of the honey samples nor the honeycomb enabled the rats to live any longer or to make any greater gains in weight than the rats that received no additions to the basal diet. These results show that the honeys and honeycomb examined contained no vitamin B.

VITAMIN C DETERMINATIONS

The method used to determine the vitamin C content of honey was that described by Sherman, LaMer, and Campbell (10). No tests were made on the honeycomb. The basal diet described by Sherman (9) consisted of skim-milk powder heated at 110° C. for 4 hours, 30 per cent; a mixture of equal parts commercial rolled oats and wheat bran, 59 per cent; butterfat, 10 per cent; table salt, 1 per cent. The guinea pigs were somewhat heavier at the beginning of the test period than the standard animal described by Sherman. Some difficulty had been experienced in other work in getting the smaller guinea pigs to eat the basal diet satisfactorily. In these tests the preliminary period was continued until it was ascertained beyond a doubt that the guinea pigs would eat the basal diet.

The honey was fed apart from the basal diet, and the test period was continued in each case until the guinea pig died. Honey is not relished by guinea pigs, and the feeding required a great deal of time and patience. The intake of honey was calculated as grams per day per 300 gm. of initial body weight. Table 3 gives the results of the feeding tests. For purposes of comparison each plus (+) under autopsy findings has been given a value of 1 and these have been totaled for each animal.

On an average the guinea pigs that had the honey did not live appreciably longer than those that had none. Before death and at autopsy all showed symptoms of scurvy that were as severe as the symptoms shown by the control animals. It is evident from these results that the three samples of honey examined contained no appreciable amounts of vitamin C.

VITAMIN D DETERMINATION

The method for testing for vitamin D is identical with that described in a previous paper (7). Young rats approximately 60 grams in weight were placed on the Steenbock low phosphorus diet consisting of yellow corn, 76 per cent; wheat gluten, 20 per cent; calcium carbonate, 3 per cent; and sodium chloride, 1 per cent, for 21 days, at which time they showed evidences of a rachitic condition. The honey was then fed during a test period of not less than 6 nor more than 15 days. At the end of the test period a line test was made according to the method described by McCollum (15). While this method may not be entirely satisfactory for quantitatively measuring vitamin D it seemed to us to be preferable to any other method worked out to date.

The three honeys tested were incorporated in the basal diet to the amount of 30 per cent, 10 per cent, and 20 per cent, respectively, and the honeycomb as 10 per cent. In each litter of rats used for testing honey No. 1 there was one or more control rats which were given 0.5 per cent cod-liver oil during the test period instead of honey. This plan of having positive controls was not considered necessary in the tests with the other samples.

Summaries of the results of these tests are given in Table 4. As all of the line tests with the honeys and the honeycomb were negative, the results with individual rats are not given. X-ray photographs were also made of the rats used in these determinations. These pictures check the line tests made on the corresponding rats in that all showed severe rickets. From these results it would seem that none of the honeys examined nor the honeycomb contained any amount of vitamin D that would cause calcium deposition in rats which had been maintained for 21 days on the Steenbock low-phosphorus-yellow-corn diet.

TABLE 4.—Summary of tests made to determine the vitamin D content of honey and honeycomb as compared with a cod-liver oil supplement

WHITE-CLOVER HONEY (NO. 1)

Test food in the diet (per cent)	Duration of test period	Number of cases	Average intake of honey per 100 gm. of rat per day	Average value of line test
	Days		Grams	
0.....	0	11	-----	0
	11	5	-----	0
	13	6	-----	0
	15	1	-----	0
30.0.....	11	13	2.36	0
	12	2	1.98	0
	13	13	2.39	0
	15	1	2.49	0

COD-LIVER OIL

0.5.....	{ 9	15	0.04	4
	{ 11	2	.04	4

BUCKWHEAT HONEY (NO. 2)

0.....	{ 0	4	-----	0
	{ 13	2	-----	0
	{ 15	4	-----	0
10.0.....	{ 11	2	0.70	0
	{ 13	3	.67	0
	{ 15	3	.70	0
20.0.....	{ 13	2	1.45	0
	{ 15	2	1.42	0

LIGHT-COLORED WHITE-CLOVER HONEY (NO. 3)

0.....	{ 0	3	-----	0
	{ 15	3	-----	0
20.0.....	{ 13	12	1.49	0
	{ 15	12	1.50	0

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0.....	{ 0	1	-----	0
	{ 13	1	-----	0
10.0.....	{ 9	3	0.80	0
	{ 13	3	.79	0

CONCLUSION

The three samples of honey examined were produced in widely separated localities and represented extremes of color variation. No detectable amounts of vitamins A, B, C, or D were found in any of the honeys or in the honeycomb.

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