THE EFFECT OF EXPERIMENTAL HYPERTHYROIDISM IN RATS UPON THE RIBOFLAVIN, NICOTINIC ACID AND PANTOTHENIC ACID CONTENT OF VARIOUS TISSUES

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PREFACE

Experimentally induced or naturally occurring hyperthyroidism has been associated with an enhanced dietary requirement for several of the vitamins, particularly those of the B-complex. In the course of experiments in progress at the Oklahoma Agricultural Experiment Station on unknown growth factors for successful reproduction and lactation in rats and swine, an assay procedure was developed which used the growth-depressing effect of hyperthyroidism induced by feeding iodinated casein. A survey of the information available showed that inadequate information was available concerning the effect of such endocrine imbalance on the vitamin content of the tissue, even for those vitamins for which suitable assay procedures have been developed. As a forerunner of studies on the effect of hyperthyroidism on the vitamin B_{12} content of tissue, studies were made to determine the effect on riboflavin, nicotinic acid and pantothenic acid. This report deals with certain of these studies.

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INTRODUCTION

Various studies have been made on the effect of experimentally induced hyperthyroidism on vitamin metabolism and vitamin requirements. However, the data published on the effect of hyperthyroidism on the riboflavin, nicotinic acid and pantothenic acid content of rat tissues are very limited.

In 1932 Hemwich (13) demonstrated an increased requirement for vitamin B during hyperthyroidism in rats. At about the same time Cowgill and Palmieri (5) discovered that the vitamin B requirement for pigeons was greater during hyperthyroidism than under normal conditions. Drill and Sherwood (10) found in 1938 that rats fed a normal diet plus sufficient thyroid gland to produce a loss in weight would stop losing weight when thiamin was injected and would regain their lost weight when a source rich in the vitamin B_2 complex such as yeast was added. In a similar study Drill and Overman (9) demonstrated that injections of pyridoxine and calcium pantothentate could effectively replace the addition of yeast. Thus in addition to thiamine, both pyridoxine and pantothenic acid were required in larger amounts during experimental hyperthyroidism.

Chevremont and Combaire (4) reported that during the increase in cellular metabolism produced in rats by the injection of 20 mg per day of thyroxine, the dialyzable lactoflavin of all the tissues increased and the lactoflavin combined as yellow enzyme decreased. In an experiment carried out by Sure and Ford (19) subcutaneous injections of from 0.5 to 1.0 mg of synthetic thyroxine to paired

rats given 850 mg of riboflavin for 17 to 20 days resulted in the excretion of larger amounts of ribovlavin and only slightly larger excretions of thiamine in the urine of the experimental rats than of the controls. Tissue levels of both vitamins were decreased. They further stated that oral administration of the same amount of synthetic thyroxine produced only a small decrease in the riboflavin content of various tissues. No explanation was given for this observation since the animals showed symptoms of thyroid toxicity.

Katzenelbogen (14) investigated the relationship between hyperthyroidism and nicotinic acid requirement. He noted that feeding of desiccated thyroid to rats on a diet low in nicotinic acid resulted in a marked diminution of the coenzyme I content of both liver and kidney cortex. Increased dosage of nicotinic acid restored the normal coenzyme I content in the hyperthyroid animal.

Drill (6) demonstrated that rats receiving 12 grams of a normal diet plus 100 mg of thyroid gland per day showed a normal amount of thiamin in the spleen, a reduction in the kidney and a marked reduction in the liver when compared with normal rats on the same diet. Hyperthyroid rats receiving 12 mg of normal diet and later injected with 500 mcg of thiamin per day, while still receiving the thyroid gland showed normal amounts of thiamin in the spleen and muscle, slightly raised content in the heart and a definite reduction in the kidney and liver. The hyperthyroid rats injected with 500 mcg of thiamin per day eliminated the same amount of thiamine in the urine as the controls.

Ershoff reports (11) that toxic doses of desiccated thyroid increased requirements for at least one unknown nutrient in the immature female rat. Failure to provide this factor in adequate amounts resulted in deficiency symptoms, manifest in the thyroid-fed rats by retarded growth and early death. Whole liver completely counteracted the above retardation of growth, while both whole liver and yeast prolonged significantly average survival on thyroid-containing rations. The beneficial effects of whole liver on the growth of immature thyroid-fed rats were correlated with increased food consumption and increased efficienty of food utilization. The protective factor (s) in whole liver and yeast was distinct from any of the known vitamins since individual supplements of thiamine, riboflavin, pyridoxine, calcium pantothentate, nicotinic acid, inositol, p-amino benxoic acid, biotin-folic acid or ascorbic acid were without effect on the above symptoms. Betheil (2) and Lardy demonstrated that on a synthetic basal ration whole liver powder at a level of 10% completely reversed the growth retardation in the immature, hyperthyroid rat; a level of 3 to 5 per cent was partially effective.

Betheil and Lardy (2) state that vitamin B_{12} is effective in counteracting the growth retardation which results from the feeding of desiccated thyroid.

Drill and Shaffer (7) demonstrated that when hyperthyroid dogs were deprived of a yeast supplement, the caloric intake dropped below normal, and weight losses followed. In an experiment conducted by Sure and Ford (19) the experimental-hyperthyroid rats,

given daily doses of 0.59 mg of synthetic thyroxine by injection consumed more food than the control rats.

A sex difference in response to thyroid feeding in rats has been reported by Drill (8). He found that female rats receiving 100 mg of desiccated thyroid gland daily ceased to lose weight when given 50 mcg of thiamine and regained their lost weight when yeast was added to the diet. In male rats only 50 mg of thyroid gland could be so counteracted. Drill (6) also noticed that female rats were more resistant than male rats to thyroid feeding as judged by the difference in the loss of weight.

PROCEDURE

Rats of the Sprague-Dawley strain were used in these experiments. In Experiment I 16 male rats weighing approximately 175 grams, were placed on the basal ration (Table I) for an equilibration period of 10 days. They were then alloted according to weight into two groups. One group (designated C) was given the basal ration while the other (designated E) received the same ration plus 0.25 per cent iodinated casein (Protamone, Cerophyl Laboratory, Kansas City, Missouri). The basal ratinn included the mineral mix recommended by Hegsted (12) in order that these nutrients would not be limiting nutritional factors. The basal ration was supplemented with one half the usually accepted optimal amounts of vitamins in order that a presumably adequate but not excessive supply was available. In this way it was hoped that the vitamin content of the tissues would more nearly represent the amounts required for the metabolism of the cells rather than either a condition of storage following excess supply or a state of depletion. Rats were kept in individual cages and supplied food and water ad libitum. The rats were weighed daily. After ten days, the rats on the experimental ration lost approximately 12 per cent of their initial body weight while the controls gained weight. Symptoms of hyperthyroidism, (nervousness, hyperpnea, tachycordia) were evident. The animals were then sacrificed and the liver, kidneys, heart, brain and a section composed principally of the gastrocnemius muscle were removed from each rat. The samples were frozen and stored at -14° C until analyzed

Tissue samples were analyzed for their riboflavin and nicotinic acid content. Tissue extracts were prepared by acid hydrolysis according to the method described by the Association of Vitamin Chemists (1) which involved the following procedure: A homogenous sample of tissue weighing approximately 1 to 2 grams was blended with 25 ml of water in a Waring Blandor, washed into a 125 ml Erlenmeyer flask and autoclaved with 5 ml of 1 N HCl at 15 lbs. for 15 minutes. The extract was then neutralized with NaOH, transfered to a 100 ml volumeteric flask, made up to volume and filtered into a 125 ml Erlenmyer flask, plugged with cotton and autoclaved. This extract was used for both riboflavin and nicotinic acid. The procedure of Snell and Strong (17) with a modification of the basal media was used for the microbiological assay of riboflavin. The modified basal media contained per liter:

Sodium hydroxide treated peptone	10 g.
l-cystine	200 mg.
l-tryptophan	200 mg.
Yeast Supplement	2 g.
Glucose	40 g.
Adenine, quanine, uracil	20 mg.
P-amino benzoic acid	800 mcg.
Pridoxine	200 mcg.
Nicotinic Acid	200 mcg.
Calcium d-pantothenate	200 mcg.
Salt solution A	10 ml.
Salt solution B	10 ml.

The procedure of Snell and Wright (18) with the following modification of the basal media was used for the microbiological determination of nicotinic acid. The modified basal nicotinic acid media contained per liter:

Acid-hydrolyzed casein	10 g.
l-cystine	400 mg.
l-tryptophane	200 mg.
Glucose	40 g.
Sodium Acetate	40 g.
Biotin	0.4 mcg.
P-amino benzoic acid	200 mcg.
Thiamin	200 mcg.
Calcium d-pantothenate	200 mcg.
Pyridoxine	200 mcg.
Riboflavin	400 mcg.
Adenine, quanine, Uracil	20 mg.
Salt solution A	10 ml.
Salt Solution B	10 ml.

Experiment II was carried out in a similar manner with two modifications. First, the rats in the control and experimental groups were "paired" with respect to sex and body weight and each rat of a pair fed an equivalent amount of the basal and hyperthyroid-inducing ration instead of having the food supplied ad libitum. Second, in addition to riboflavin and nicotinic acid determinations, the tissue samples were also analyzed for their pantothenic acid content.

The 30 male and female rats weighing approximately 200 grams used in this experiment were placed on the basal ration (Table I) for a 14-day equilibrum period and then paired according to sex and weight. The pairs were separated into control and experimental groups. The same rations were used as in experiment I. Water was supplied ad libitum. Each rat of a pair consumed the same amount of ration so as to keep the vitamin intake constant. This was carried out by weighing equal portions of basal and experiment ration for the control and experimental rat respectively, the amount determined by the food intake of the animal consuming the lesser. The rats were kept in individual cages and supplied

with water ad libitum. The average daily food consumption of the respective pairs is shown in Table II.

As in Experiment I, the experimental rats had lost approximately 12 per cent of their body weight 10 days after they had been put on the experimental ration, while the control rats had gained somewhat (Figure 2). All the rats were sacrificed. The liver, kidneys, heart, brain and a section comprised chiefly of the gastrocnemius muscle were removed from each rat. The samples were frozen and stored at -14°C until analyzed.

Tissue analysis for riboflavin and nicotinic acid was carried out as described for Experiment I. The pantothenic acid analysis was done upon an enzymatically hydroyzed extract as recommended by Buskirk (3). The following procedure was employed: A homogenous sample of tissue weighing approximately 1 to 2 grams was blended with 25 ml. water in a Waring Blender and washed into a 100 ml volumeteric flask. A 20 ml aliquot was treated with 0.4 ml of glacial acetic acid, 2 ml of 1 N NaOH and 0.4 grams of Myalase P (Wallerstein Laboratories, New York, N. Y.) and incubated for 2½ hours at 50°C. The extract was transferred to a 100 ml volumeteric flask and made to volume, filtered into a 125 ml Erlenmeyer flask, plugged with cotton and autoclaved. This extract was neutralized before analysis. The microbiological assay procedure described by Pennington and Snell (16) with a modification of the basal media was followed for the pantothenic acid determinations. The basal media used contained per liter:

Sodium hydroxide-treated peptane	10 g.
l-cystine	200 mg.

Glucose	40 g.
Sodium acetate	28 g.
Acid hydrolyzed casein	4 g.
Adenine, guanine, uracil	20 mg.
Nicotinic Acid	400 mcg.
Biotin	0.8 mcg.
Riboflavin	400 mcg.
Pyridoxine	200 mcg.
p-aminobenzoic acid	200 mcg.
Salt solution A	10 ml.
Salt solution B	10 ml.

RESULTS AND DISCUSSION

The results of the riboflavin determinations of the normal and hyperthyroid rat tissues from Experiment I are presented in Table III. The average vitamin content is compared with the values reported by Mitchell and Isbell and the deviation of the vitamin content of the experimental from the control is also shown. The riboflavin, nicotinic acid and pantothenic acid content of tissues from animals in Experiment II are presented in Tables IV,V, and VI respectively.

Examination of the data on the riboflavin content of tissues from the animals of Experiment I shows that the rats receiving the thyroid-active material had a slightly lower content of riboflavin in the liver; all other tissues showed a slight increase. This general increase in riboflavin content of tissues during thyrotoxicosis is not consistent with the findings of Sure and Ford (19) who administered the hormone by injection. It will be noted from the weight changes (Figure 1) that all animals in the thyroid-fed group lost weight while the control animals gained slightly. Thus, although the controls ate somewhat more food than the experimental animals the intake of riboflavin per unit of body weight was greater in the experimental than in the control group. Further, as will be discussed in greater detail later, the increased metabolic rate occasioned by the addition of thryoxine-active materials would seem to demand a higher level of the various co-enzymes which are involved in the oxidation of foodstuffs. This might

conceivably produce a mobilization of the vitamin reserves from the liver and cause a concommitant decrease in that organ with a gain in the extra-hepatic tissues.

Due to procedural difficulties, only a limited number of the samples were analyzed for nicotinic acid. The average values for those which were examined, however, are presented. The results were as follows: liver, control 182 and experimental 210 micrograms of nicotinic acid per gram of fresh tissue; kidney, 150, 211; heart, 116, 143; brain, 65,89; muscle, 91,98. It will be seen that in every tissue, including liver, the experimental animals had a higher nicotinic acid content than the controls. This is consistent with the findings concerning the riboflavin content of most of these tissues. Examination of the literature has not revealed any previous reports of a comparison of the nicotinic acid content of normal and hyperthyroid rats or other animals.

It was obvious from the results of the first experiment, that one of the complicating factors was the differential food consumption, and hence vitamin consumption, of the normal and thyroid-fed animals. A controlled-feeding experiment was therefore, employed to decrease the effect of variation in food consumption. That this goal was achieved is shown by the figure in Table II; only minor variation in average food consumption during the ten day period was permitted between control and thyroid-fed animals. Again, it was observed that the experimental group was the one which would have consumed the least food on an ad libitum basis.

Examination of the data in Table IV shows that in this controlled-feeding-type study, the same elevation in riboflavin

content of the hyperthyroid animals was found. This was true even in the case of liver, which increased from an average of 16.7 micrograms of riboflavin per gram in the control to 20.8 in the livers of the experimental group. In comparing the data on riboflavin from Experiments I and II, it will be seen that the liver and kidney levels in the first experiment were less than in the second; this applied to both experimental and control animals. No obvious explanation for this deviation can be advanced, since the animals in both experiments were of the same source and were handled in an identical manner except of the control of food intake in Experiment II. It does not seem likely that the reduced food intake would cause an increase in the vitamin content; in view of the fact that the values obtained in the second experiment represent a larger number of animals, these are considered to be the more reliable. Whether the controlled intake of food is likewise responsible for the differences observed in the response of the liver to thyroid administration is likewise not known.

More extensive nicotinic acid analyses were performed on the tissues from the animals in Experiment II and they followed the same pattern as did the limited observations in Experiment I. Variations from one experiment to the other in vitamin content in a single tissue were noted but in general even the specific values agreed fairly well--this latter in contrast to the findings on the riboflavin content of liver and kidney in the two experiments.

Pantothenic acid was determined only on the tissues of the last experiment. Again, the addition of hormone produced a consistant elevation in concentration of this vitamin in the various tissues examined. In no instance did a pair fail to reveal this change in every tissue, although, as might be expected in the case of changes of relatively small magnitude, there was an occasional instance of "overlapping." The values for the content of pantothenic acid agreed fairly well with those reported in the literature (15) except in the case of liver. In view of the storage function of this organ, and the fact that vitamins were supplied at levels of one half optimal, such a decline might well have been expected.

The increased vitamin content of the rat tissues in experimental hyperthyroidism, as demonstrated by the results of Experiments I and II, are not inconsistant with the findings summarized in the Introduction indicating that an enhanced requirement for several members of the B-complex exists during the hyperthyroid state. Presumably, much of this is due to increased metabolic rate with more rapid oxidation of carbohydrate and fat and catabolism of body protein. The requirement for the various coenzymes functional in carbohydrate and protein metabolism would be greater and the concentration in the tissues might be increased if adequate supplies of the specific vitamin was available. If this were the case, it would explain the increased tissue content of riboflavin (component of the flavin-adenine dinucleotide) and nicotinic acid (di- and triphosphopyridine nucleotide) observed

in these studies. Although the enzymatic function of pantothenic acid is less well understood, it appears that derivatives of this vitamin may be involved in both fatty acid and amino acid breakdown.

This observation of an enhanced vitamin level of the tissues during thyrotoxicosis is not consistant with the reported observations of Sure and Ford (19). In their experiment the animals were rendered hyperthyroid by injection of 0.59 mg of synthetic thyroxine per day. They reported that these hyperthyroid animals consumed more food than their corresponding controls. Analyses of liver, kidney, heart, brain, and muscle showed a consistant decline in the riboflavin content. When similar amount of thyroxine was administered orally, however, they did not find any appreciable change in the tissue concentration of either thiamin or riboflavin. They do not give any reason for this anomolous observation, since the animals showed other symptoms of thyroid toxicity.

The fact that we have made similar observations with respect to two other vitamin studies suggests to us that under the particular conditions employed in this experiment an increase rather than a decrease is secured upon the feeding of Protamone. Sure and Ford (19) presented data bearing only on the effect of injected thyroxine. No specific data concerning the number of animals or tissue levels found is contained in that portion of their communication describing the results following oral administration. Both their statement and our findings, however, suggested that

difference in method of administration may be responsible for the differences in results obtained in the two laboratories.

In both experiments the rats exhibited a decreasing food intake as they became more hyperthyrogenic. This was in agreement with the results obtained by Drill and Shaffer (7) who found that the caloric intake of hyperthyroid animals dropped below normal. It does not, however, agree with the statement of Sure and Ford (19) that the hyperthyroid animals consumed more food than the corresponding control animals.

The male rats were found to have a slightly higher food intake than the female rats. The male rats of the control group gained slightly more weight than the females. The male rats of the thyroid-fed group lost slightly more weight than the females. Drill (6) demonstrated a greater resistance toward the thyroid-containing diet by female rats judged by the loss of weight. Our observations corroborate these results with respect to sex differences. Drill (8) noted that 500 mg of thiamin would counteract the loss in weight in female rats induced by as much as 100 mg per day of orally administered dessiccated thyroid gland. In male rate, however, only 50 mg per day of thyroid gland was the maximum level that could be so counteracted.

Despite differences in food intake and rate of weight change, no apparent sex differences in vitamin content of tissues was found. It may be that if the experimental period had extended over a longer period the greater resistance that female animals apparently have toward toxic levels of thyroid-agents would have

manifested itself in differences in the tissue concentrations of riboflavin, nicotinic acid, and pantohenic acid. On the other hand, it may be that the endocrine relationships which permit greater tolerance by the female are unrelated to the state of nutrition with respect to the B-complex.

TABLE I

COMPOSITION OF RATIONS

Ingredient*	Basal	Experimental
Casein, vitamin	22%	22%
Surcrose	69%	69%
Corn oil	5%	5%
Mineral Mix (10)	4%	4%
Protomone		2.5 g/Kg

*Each kilogram of ration was supplemented with the following vitamins: thiamin, 2 mg; riboflavin, 3 mg; pyridoxine, 1.5 mg; calcium pantothenate, 10 mg; nicotinic acid, 10 mg; pteroylglutamic acid, 0.5 mg; inositol, 10 mg; para-aminobenzoic acid, 10 mg; choline, 0.5 gm. In addition vitamins A and D and alpha tocopherol were administered by dropper twice a week.

TABLE II

AVERAGE DAILY FOOD CONSUMPTION OF RATS

EXPERIMENT II

Day of Experiment	Food Consumed, Grams			
	Male		Female	
	Control	Experimental	Control	Experimental
1	15.0	15.0	15.0	15.0
2	16.0	16.0	15.0	14.6
3	17.0	17.0	14.6	14.4
4	18.0	17.8	14.4	13.2
5	17.8	17.5	13.2	13.2
6	17.5	17.5	13.2	12.8
7	17.5	17.2	12.8	12.4
8	17.2	17.0	12.4	12.4
9	17.0	16.8	12.4	12.0
10	16.8	16.5	12.0	11.7
Average	17.0	16.8	13.5	13.2

TABLE III

RIBOVLAVIN CONTENT OF RAT TISSUES

EXPERIMENT I

Values in mcg per gram moist tissue

Rat Number	Liver		Kidney		Heart		Brain		Muscle	
	C	E	C	E	C	E	C	E	C	E
1	Dead	10.6	Dead	9.14	Dead	21.2	Dead	4.44	Dead	2.98
2	11.3	9.17	6.93	8.62	15.7	17.5	4.07	4.42	1.61	3.22
3	11.2	6.72	6.46	8.30	12.8	21.2	3.68	3.82		3.36
4	11.1	6.66	5.30	15.0	11.9	21.2	3.75	5.17	3.37	3.51
5	14.6	10.0	6.51	9.38	13.1	18.1	3.40	5.35	2.93	3.50
6	12.1	7.3	6.10	13.5	15.2	25.0	3.64	4.72	3.16	3.03
7	11.6	10.1		10.2	16.8	18.2	3.54	4.03	3.03	3.68
8	12.2	6.77	7.9		16.4	17.1	4.32	3.40	3.50	3.72
Average	12.0	8.42	6.53	10.6	14.6	20.0	3.77	4.42	2.93	3.38
Deviation, %		-30.0		+62.8		+37.0		+17.2		+15.4
Literature	30		29		14		2.9		1.7	

TABLE IV

RIBOFLAVIN CONTENT OF RAT TISSUES

EXPERIMENT II

Values in mcg per gram moist tissue

Pair Number	Sex	Liver		Kidney		Heart		Brain		Muscle	
		C	E	C	E	C	E	C	E	C	E
1	Male	13.5	21.6	22.0	23.4	13.7	15.6	1.89	2.34	1.83	2.56
2	Male	17.8	19.7	19.6	24.2	14.2	15.3	1.88	2.39	2.11	2.42
3	Male	16.1	18.6	21.8	24.9	13.6	14.9	1.97	2.29	2.24	3.09
4	Male	15.9	18.7	20.2	24.0	12.6	14.2	1.90	2.48	2.36	3.07
5	Male	16.8	23.7	21.2	27.4	14.4	15.2	1.86	2.53	2.27	2.92
6	Male	19.4	25.1	20.3	22.8	13.6	14.8	1.86	2.63	2.34	2.93
7	Male	15.5	26.7	20.0	23.1	14.0	16.0	1.93	2.44	2.19	2.83
8	Female	12.1	19.9	21.6	22.8	12.1	14.0	2.12	2.69	2.19	3.04
9	Female	17.0	20.2	19.9	22.4	12.9	14.1	2.02	2.81	2.29	2.93
10	Female	16.4	20.4	20.5	22.1	13.1	14.2	2.04	2.81	2.31	2.71
11	Female	16.9	19.0	20.5	22.4	12.3	15.6	2.02	2.39	2.56	2.80
12	Female	17.4	16.9	20.1	21.4	13.3	16.0	1.92	2.31	2.50	2.96
13	Female	19.4	22.2	19.7	22.2	13.1	15.8	2.01	2.65	2.42	2.55
14	Female	17.9	19.1	21.8	24.8	12.6	14.0	1.85	2.49	2.64	2.91
15	Female	18.6	19.9	21.5	22.6	11.0	13.9	2.00	2.61	2.43	2.96
Average, Total		16.7	20.8	20.7	23.3	13.1	14.9	1.95	2.52	2.31	2.85
Male		16.4	22.0	20.7	24.2	13.7	15.1	1.90	2.44	2.19	2.83
Female		17.0	19.7	20.7	22.6	12.6	14.7	1.99	2.59	2.42	2.86
Deviation, Total %			+24.6		+12.6		+13.7		+29.2		+23.4
Male, %			+34.1		+16.9		+10.2		+28.4		+29.2
Female, %			+15.9		+ 9.2		+16.7		+30.2		+18.2
Literature											
Male		30		29		14		2.9		1.7	
Female		25		26		12		3.2		1.9	

TABLE V

NICOTINIC ACID CONTENT OF RAT TISSUES

EXPERIMENT II

Values in mcg per gram moist tissue

Pair Number	Sex	Liver		Kidney		Heart		Brain		Muscle	
		C	E	C	E	C	E	C	E	C	E
1	Male	191	210	90.3	120	90.3	101	45.5	49.7	64.2	83.0
2	Male	187	221	94.6	115	85.6	103	45.8	51.5	59.2	70.1
3	Male	175	236	95.0		94.2	112	46.3	47.1	70.1	73.3
4	Male	160	218	84.5		87.6	114	42.9	48.2	62.0	68.0
5	Male	172	199	99.7		82.7	106	43.8	47.7	63.5	70.6
6	Male	187	198		123	95.0	107	45.6	46.6	56.8	71.9
7	Male	193	206		132	96.6	100	46.3	47.6	66.2	76.3
8	Female	184	205		140	97.1	119	45.3	50.4	86.7	99.2
9	Female	179	236	122	130	86.0	112	47.2	50.3	67.1	78.3
10	Female	168	231		143	87.2	108	45.7	51.6	72.3	89.9
11	Female	172	201	145	154	91.0	109	48.6	51.0	86.4	88.3
12	Female	180	195	124	145	98.9	120	49.6	52.5	88.0	89.3
13	Female	186	211		144	99.5	125	45.3	50.4	89.3	92.0
14	Female	190	214		135	97.0	120	47.5	53.1	90.1	94.3
15	Female	174	213		136	95.1	117	47.9	50.4	92.3	92.3
Average, Total		180	213	107	135	92.3	112	46.2	49.9	74.3	82.5
Male		181	212	92.8	125	90.3	106	45.2	48.3	63.1	73.3
Female		179	213	130	141	93.9	116	47.2	51.2	84.0	90.5
Deviation, Total %			+18.3		+26.1		+21.7		+8.0		+11.0
Male, %			+17.1		+34.4		+17.7		+6.9		+16.2
Female, %			+19.0		+ 8.5		+23.4		+8.5		+ 7.7
Literature											
Male		190		110		120		64		64	
Female		170		120		130		64		97	

TABLE VI

PANTOTHENIC ACID CONTENT OF RAT TISSUES

EXPERIMENT II

Values in mcg per gram moist tissue

Pair Number		Liver		Kidney		Heart		Brain		Muscle	
		C	E	C	E	C	E	C	E	C	E
1	Male	32.3	40.7	31.0	36.4	25.2	30.6	8.02	12.5	5.46	5.55
2	Male	31.2	44.1	25.2	35.9	19.0	29.0	10.7	15.2	4.28	4.80
3	Male	37.1	44.1	27.5	36.3	22.0	27.0	9.01	17.1	4.39	4.72
4	Male	29.9	40.7	32.2	36.7	23.0	30.6	9.50	13.0	3.90	3.92
5	Male	30.1	54.3	28.5	35.3	25.7	32.7	8.78	17.2	5.20	6.00
6	Male	30.9	54.4	33.8	37.1	28.0	35.0	10.0	17.5	5.53	5.70
7	Male	27.3	54.7	32.2	37.7	17.0	29.0	6.50	13.5	4.90	4.87
8	Female	26.0	38.6	30.0	34.1	19.6	29.5	9.75	13.7	3.80	4.00
9	Female	30.8	59.3	29.1	33.3	19.9	33.0	8.50	14.4	3.99	4.10
10	Female	35.2	38.2	34.6	36.8	20.6	36.0	7.50	13.2	4.90	4.98
11	Female	26.9	45.8	36.4	39.2	27.0	38.0	10.5	14.6	3.27	4.50
12	Female	29.3	43.9	36.7	38.1	29.0	40.0	10.6	15.4	3.17	4.73
13	Female	31.7	43.7	32.4	32.6	26.8	31.0	11.0	15.2	3.71	4.06
14	Female	31.8	47.9	29.0	33.3	21.0	29.7	10.1	13.8	3.46	4.01
15	Female	32.5	55.8	33.7	42.0	22.0	34.4	10.9	17.7	3.14	3.75
Average, Total		30.8	47.1	31.5	36.3	23.1	32.4	9.42	14.9	4.22	4.65
Male		31.3	47.7	30.1	36.5	22.9	30.6	8.93	15.1	4.81	5.08
Female		30.5	46.7	32.7	36.2	23.2	33.9	9.86	14.5	3.62	4.27
Deviation, Total %			+52.9		+15.2		+40.2		+58.4		+10.2
Male, %			+52.4		+21.3		+33.6		+69.4		+ 5.6
Female, %			+53.1		+10.7		+46.1		+47.0		+18.0
Literature											
Male	150			32		30		12		5.5	
Female	73			34		40		12		4.5	

AVERAGE CHANGE IN WEIGHT OF RATS

Experiment I

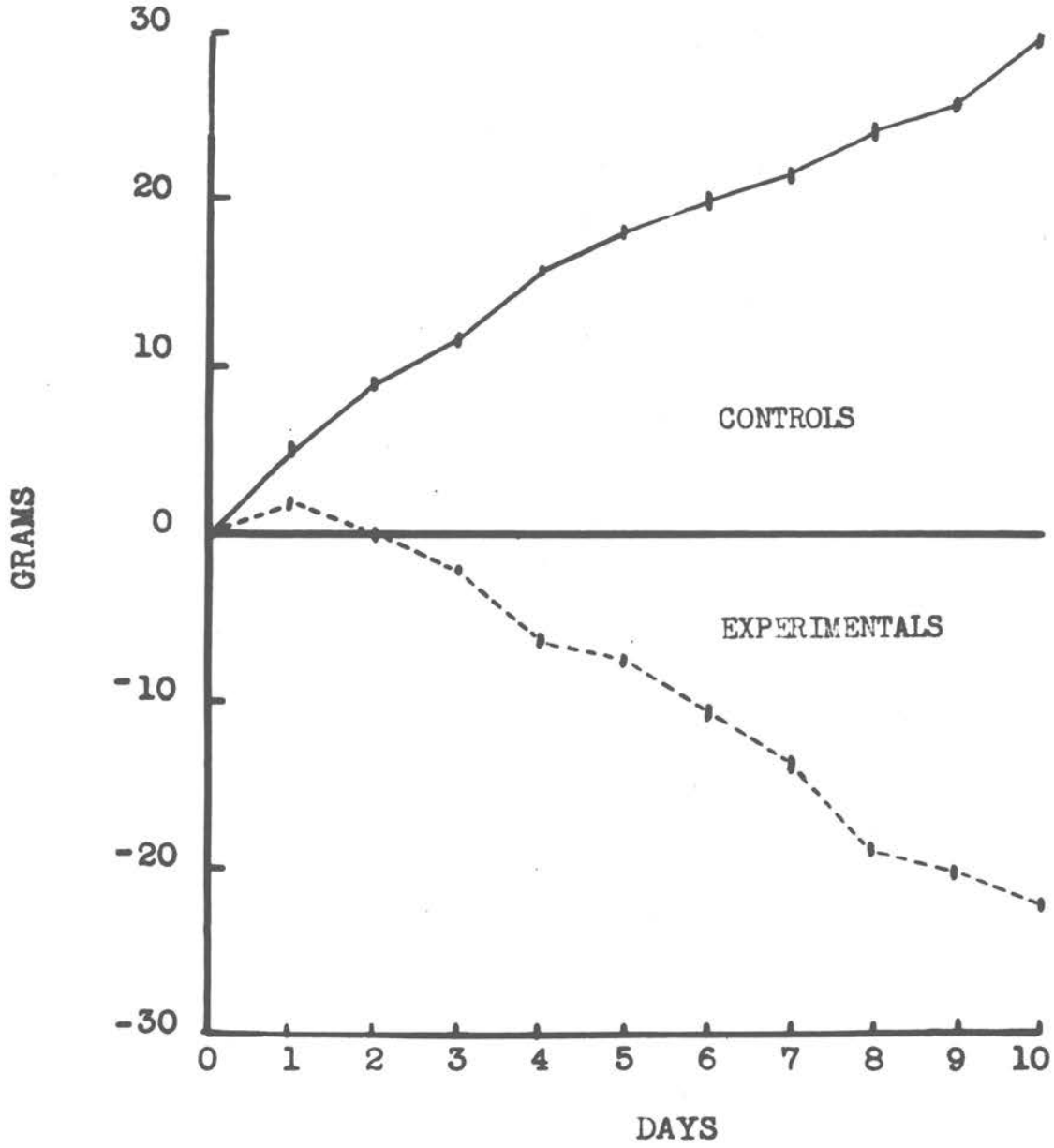

Figure 1

AVERAGE CHANGE IN WEIGHT OF RATS

Experiment II

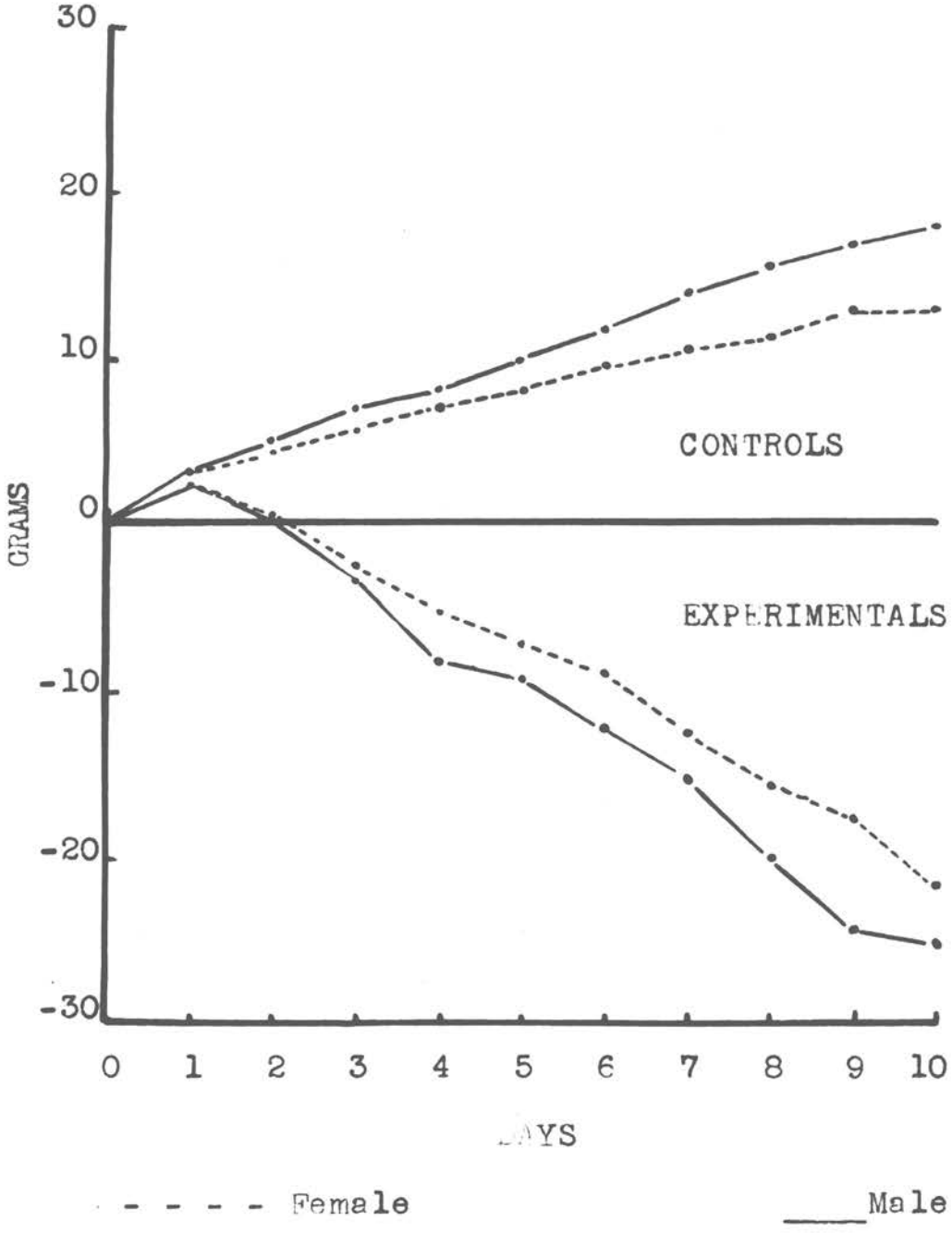

Figure 2

SUMMARY

The effect of experimentally induced hyperthyroidism in rats upon the riboflavin nicotinic acid and pantothenic acid content of various tissues has been studied. Animals were brought into a state of thyroid toxicity by the oral administration of iodinated casein for a period of ten days, at which time the symptoms of toxicity were apparent. Equal intake of food and vitamins was assured by pair-feeding selected pairs of animals to the same level of consumption. The vitamin intake during a previous adjustment period and during the actual experimental period was one half the usually accepted optimal level.

In all tissues examined, the administration of the thyroid-active material resulted in an increase, usually in the range of from 10 to 50 per cent in the vitamin content of the tissue. Pantothenic acid was increased most and nicotinic acid least. Results obtained in liver, kidney, heart, brain and muscle were all consistent when a controlled-feeding technique was employed. In a preliminary ad libitum feeding experiment, similar though not identical results were obtained.

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