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Vitamins B_1 and B_2 in Tissues of Normal and Experimental Rats.

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The relative concentration of Vitamins B_1 and B_2 in normal and experimental rats was determined by feeding dried rat tissue as the source of these vitamins. "Normal rat tissue" was prepared from stock rats, 200-300 gm. in weight. The rats were decapitated, the carcasses (except skin, feet, tail, and intestines) were hashed in a meat grinder and the hashed material was dried in a warm air dryer (50°C.). In the same manner "B_1 rat tissue" was prepared from 20 rats after 30 days on the Evans and Burr diet supplemented with cod liver oil and tikitiki. The average starting and final weights of this group were 186 gm. and 172 gm. respectively, indicating cessation of growth and a loss of weight during the 30-day period on the diet deficient in B_2. "B_2 rat tissue" was similarly prepared from 20 rats after 30 days on the Evans and Burr diet supplemented with cod liver oil and autoclaved liver. The average starting and final weights of this group were 184 gm. and 148 gm. respectively, indicating cessation of growth and a loss of weight during the 30-day period on the diet deficient in B_1. These 3 rat tissue preparations were fed as sources of B_1 and B_2 to young 50 gm. male rats fed the Evans and Burr diet supplemented with cod liver oil and either tikitiki or autoclaved liver.

Normal rat tissue was not a good source of either B_1 or B_2 but was definitely richer in the latter. Better growth resulted with 0.3 gm. of this tissue as a source of B_2 (Group 8) than with 0.7 gm. as a source of B_1 (Group 7).

Normal rat tissue contained more B_1 than either the B_1 or the B_2 rat tissue preparations. The rats fed the B_2 rat tissue as a source of B_1 (Group 11) failed to survive unless tikitiki was added, the results resembling those obtained with autoclaved liver alone (Group 1). The feeding of B_1 rat tissue as a source of B_1 (Group 9) permitted survival but there was no growth after the first 10 days. It was evident from these experiments that the bodies of the rats used in the preparation of the B_1 and B_2 rat tissues were practically depleted of vitamin B_1 during the preliminary 30-day period.

These results were in marked contrast to those obtained when the same tissue preparations were used as a source of vitamin B_2.

Normal (Group 8) and B_2 (Group 12) rat tissue were of nearly equal value as sources of B_2. The concentration of this factor in B_1 rat tissue (Group 10) was lower than in the normal rat tissue but enough was present to permit growth. The rats used in the preparation of the B_1 rat tissue showed partial loss of appetite, failure of growth and loss of weight during the 30 day period on the diet containing B_1 and lacking B_2, yet the bodies of these rats still contained appreciable amounts of B_2 at the end of the 30-day period.

It was concluded that the tissues of rats on deficient diets may be readily depleted of B_1 but not of B_2. Whether this residual B_2 represents stored B_2, or B_2 which is conserved by the body and used sparingly is being investigated. The results are in accord with the observation of Osborne and Mendel[1] that livers of rats fed a diet deficient in the vitamin B complex could not supplement a diet deficient in the B complex and indicate, in addition, that the limiting factor in their experiments was vitamin B_1.

The interpretation of experimental results obtained with diets deficient in B_2 should include recognition of this residual B_2. The presence of this factor in the tissues may be partially responsible for the better appetite and for the resulting longer survival of rats fed tikitiki (Groups 2, 3, and 4) as the sole source of the vitamin B complex. Our results confirm the observation of Sherman and Sandels[2] that the appetite and food consumption of rats decrease more rapidly on diets low in B_2 than on diets low in B_1

Our previous experiments[3, 4] have suggested that B_1 and B_2 are both appetite stimulants but only in the presence of each other. The present data introduce the possibility that the sustained although subnormal appetite of rats on B_2 deficient diets may be due to the presence of residual B_2 in the tissues whereas the sharp failure of the appetite of rats on B_1 deficient diets may be due to the fact that the tissues are rapidly depleted of B_1. In the former both factors would be present, in the latter only B_2. If dogs also retain tissue B_2 then the above possibility might apply to the experiments of Burack and Cowgill.[5] On the assumption that both factors, B_1 and B_2, are required for normal appetite, a stimulation of appetite of ani-

[1] Osborne, T. B., and Mendel, L. B., *J. Biol. Chem.*, 1923, **58**, 363.

[2] Sherman, H. C., and Sandels, M. R., *J. Nutrition*, 1931, **3**, 395.

[3] Graham, C. E., and Griffith, W. H., *J. Biol. Chem.*, 1931, **92**, lxiii.

[4] Graham, C. E., and Griffith, W. H., PROC. SOC. EXP. BIOL. AND MED., 1931, **28**, 1086.

[5] Burack, E., and Cowgill, G. R., PROC. SOC. EXP. BIOL. AND MED., 1931, **28**, 750.

TABLE I.

Group	No. of Rats	Gain in Weight in 30 days low-high	Gain in Weight in 30 days mean	Gain in Weight in 50 days low-high	Gain in Weight in 50 days mean	Average Total Calories	Tiki-tiki	Auto. liver	Special Supplements
		gm.	gm.	gm.	gm.	cal.	cc.	gm.	
1	20	(−4)-22	7*	—	—†	—	—	1.0	None
2	12	(−3)-25	11	(−6)-16	9	846	0.25	—	None
3	4	5-32	17	9-35	17	842	0.50	—	None
4	5	12-25	16	11-28	18	895	0.50‡	—	None
5	6	78-142	110	105-212	152	2088	0.25	0.5	None
6	6	90-105	99	122-158	148	2025	0.50	1.0	None
7	8	22-55	43	40-81	55	1107	—	0.5	0.7 gm. normal rat tissue
8	8	35-78	52	46-99	72	1290	0.25	—	0.3 gm. normal rat tissue
9	10	14-33	24	14-32	22	881	—	0.5	0.7 gm. B_1 rat tissue
10	9	25-56	40	22-64	50	1077	0.25	—	0.3 gm. B_1 rat tissue
11	10	(−9)-17	4	—	—	—	—	0.5	0.7 gm. B_2 rat tissue
12	10	32-65	48	51-85	66	1272	0.25	—	0.3 gm. B_2 rat tissue

* 16 survived. † 5 survived. ‡ Changed to 0.25 cc. on 30th day.

mals on a diet lacking the B complex might be expected from the administration of B_1 as long as residual B_2 was present in the tissues whereas negative results would follow the administration of B_2 due to the absence of tissue B_1. Obviously, in such a case it would be necessary to perform similar experiments on animals wholly depleted of residual or tissue B_2. The real significance of the residual B_2 must await further investigation.