

confirmed. From the data in Table I, it appears that: 1. Castration did not significantly alter muscle glycogen concentration. 2. Neither testosterone nor desoxycorticosterone, in the dosages used, affected the concentration of glycogen in the gastrocnemius muscle of the intact or the castrated male rat. 3. Neither testosterone nor desoxycorticosterone restored to normal the low muscle glycogen levels found in adrenal insufficiency.

This finding, with regard to testosterone, though contrary to that of Schumann, is in agreement with the report of Gaunt *et al.*⁸ 4. None of the experimental procedures led to any marked change in the maximum tension developed by the muscle.

⁸ Gaunt, R., Remington, J. W., and Edelmann, A., *PROC. SOC. EXP. BIOL. AND MED.*, 1939, **41**, 429.

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Effects of Different Intakes of B-Complex Vitamins upon Neuromuscular Regeneration.*

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The frequency with which reports have appeared concerning the occurrence of peripheral nerve degeneration in various vitamin B deficiencies prompted us to investigate the role of vitamin B-complex in neuromuscular regeneration. This was done by making a comparative study of the rates of regeneration in animals subsisting on different levels of vitamin B-complex.

Experimental. The experiments were carried out on the gastrocnemius muscles and tibial nerves of albino rats. The animals were matched as to age, initial body weight and sex. Complete denervation of the gastrocnemius muscle of one limb was produced by crushing the tibial nerve. The non-denervated muscle and nerve of the contralateral limb served as a control. At specified times after operation measurements were made concerning the strength and weight of the regenerating and control muscles. The muscle strength was determined by measuring the maximum isometric tension which developed in response to volleys of adequate stimuli applied directly to the muscle and to its motor nerve. The

technics that were employed for denervation and strength measurements have been described in detail elsewhere.¹ The quantity of functional reinnervation was estimated from the ratios of tension responses to direct muscle stimulation and to motor nerve activation. The tension responses of control non-denervated muscles to nerve stimulation were of the same magnitude as those elicited by direct muscle activation; while in the case of muscles undergoing reinnervation and regeneration the former was only a fraction of the latter.

In our studies the following basal ration has been used: sucrose 74%, purified casein 18%, salt mixture 3% and corn oil 5%. Each animal received in addition 2 mg alpha-tocopherol and 2 drops of halibut liver oil per week. The "control" group received the basal diet supplemented with 8% yeast. The "B-deficient" group received the basal ration without yeast or other B supplements. The diet designated as "B-excess" was prepared by adding to the basal diet 8% yeast and sufficient choline, calcium pantothenate, thiamine, pyridoxine, riboflavin, and rice bran concentrate (Vitab) to provide a ten-fold increase in the calculated vitamin B complex content of the diet. In view of the fact that

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¹ Hines, H. M., Thomson, J. D., and Lazere, B., *Am. J. Physiol.*, 1942, **137**, 527.

TABLE I.
A Summary of Average Values Together with Standard Errors for Normal and Regenerating Gastrocnemius Muscles of Rats Subsisting on Different Levels of B-Complex Vitamins.

Dietary condition	No. of rats	% change in body wt after lesion	Gms tension per g muscle when activated through				Relative strength of denervated musclelet when activated through				Relative weight of regenerating musclelet
			Nerve		Muscle		Nerve	Muscle			
			Exp.*	Control	Exp.*	Control		Control	Control		
Control	26	+14	21	722 ± 80	1847 ± 14	1425 ± 40	1905 ± 47	27.1 ± 2.2	50.1 ± 1.7	67.3 ± 1.3	
B-deficient	20	-26	21	479 ± 46	1496 ± 64	1057 ± 47	1559 ± 59	22.3 ± 3.1	42.2 ± 1.7	64.1 ± 1.0	
B-excess	12	+15	21	724 ± 60	1988 ± 74	1395 ± 36	2069 ± 31	27.5 ± 2.9	44.2 ± 1.2	65.3 ± 0.9	
Fast	11	-33	21	463 ± 25	1208 ± 103	809 ± 30	1277 ± 116	32.8 ± 3.2	48.9 ± 2.8	65.1 ± 2.4	
Fast + B	13	-26	21	528 ± 38	1289 ± 64	925 ± 43	1387 ± 60	29.4 ± 2.8	46.4 ± 2.0	69.2 ± 2.6	
1% yeast	9	+6	21	583 ± 40	1306 ± 78	889 ± 64	1703 ± 88	25.6 ± 3.4	39.2 ± 4.2	62.3 ± 1.0	
Control	15	+20	42	1148 ± 78	1608 ± 30	1295 ± 44	1691 ± 26	66.3 ± 2.6	70.5 ± 1.8	79.7 ± 1.1	
B-excess	9	+19	42	1207 ± 48	1578 ± 41	1396 ± 26	1614 ± 40	69.2 ± 2.6	75.2 ± 1.5	80.5 ± 1.6	

* Exp. refers to muscles previously subjected to denervation and control to contralateral non-denervated muscles.
† Expressed as % of that found in contralateral control.

animals on vitamin B-deficient diets lose considerable amounts of body weight, it seemed desirable to conduct control experiments in which comparable weight losses were brought about by total food withdrawal and by food withdrawal with provisions for supplying the required B vitamins. Accordingly one group of animals was subjected to the standard denervation operation and deprived of all food for the last 11 days of the period allowed for regeneration. Another group designated as "fast + B" was treated as above except that care was taken to provide the animals with

twice their usual daily amount of B-group vitamins. An additional group was placed on the basal diet plus 1% yeast. They were allowed to subsist on this low vitamin B diet for 2 weeks before operation and then for an additional period of 3 weeks before tests were made as to the state of regeneration. Because of anorexia and the resulting loss of body weight, it was deemed advisable to terminate the regeneration studies with B-free diets after 21 days.

Results. The extent of muscle reinnervation and regeneration, as estimated from comparisons of the strength and weight of regenerating muscles to that of their contralateral controls, was but little affected by the levels of B-complex intakes (Table I). This was true for the response to motor nerve stimulation as well as to direct muscle activation. The strength per unit weight of control and regenerating muscles in animals subsisting on B-deficient diets was somewhat inferior to that of comparable muscles in animals on adequate vitamin intakes. However, the fact that even lower values were found for the strength of regenerating and non-denervated muscles of animals subjected to acute inanition and to food withdrawal with provisions for vitamin B intakes suggests that the neuromuscular weakness was due more to the effects of acute inanition than to the absence of any specific vitamin effect. It is not intended to imply that the B-vitamins are non-essential for optimum neuromuscular regeneration because it is probable that some amounts were available to our animals from bacterial and storage sources. It is apparent, however, that animals can be subjected to an acute vitamin B-complex withdrawal for a considerable period of time and show signs of B deficiency without exhibiting slower rates of regeneration.

Conclusions. Neuromuscular regeneration was not appreciably affected by acute vitamin B-complex withdrawal. Excess intakes of these vitamins failed to enhance reinnervation and regeneration. The neuromuscular weakness of regenerating and non-denervated muscles of animals on B-deficient diets was of the order of that found in animals subjected to either acute inanition or food withdrawal and B supplements.