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Effect of Wheat Germ Oil Upon E-Deficient Muscular Dystrophy.

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It has been shown that 5-months-old rats reared from birth on a vitamin E-deficient diet show dystrophic changes in the skeletal muscle.[1] This report is concerned with attempts to promote recovery in such animals by the administration of wheat germ oil or its vitamin E-containing concentrate.

E-deficient rats were prepared by placing prospective mothers on an E-deficient diet (Olcott[2]) when about 18 days pregnant. The mothers were continued on this diet throughout the suckling period. The young rats were weaned at 22 days of age and continued on the E-deficient diet to 5 months of age. At this time some were used for study, and the remainder divided into 3 groups as follows: (1) continued on the E-deficient diet, (2) E-deficient diet plus 10% of wheat germ oil, (3) E-deficient diet plus a weekly dose of 30 mg of a concentrate of wheat germ oil. Thirty mg of this concentrate was proven adequate to carry an established E-deficient female through the gestation and suckling period. A part of the animals of each of these 3 groups were studied at an age of 7 months, the remainder at an age of 8 months.

The gastrocnemii of the above animals were studied with regard to histologic changes, water and chloride concentration and maximum power. The maximum power was taken as the tension developed against an isometric lever when the intact muscle was stimulated with condensor discharge shocks at the rate of 50 per second. This same muscle was then removed from the animal, weighed and fixed in Zenker-formol. Sections were stained with hematoxylin and eosin.

The male rats were apparently more susceptible to the E-deficient regime than were the females. Therefore, the average results for the 2 sexes are shown separately (Tables I and II). Average values from normal rats reared on a stock diet are included for comparison.

Morphologically, the changes observed in the untreated E-deficient

[1] Knowlton, G. C., and Hines, H. M., PROC. SOC. EXP. BIOL. AND MED., 1938, **38**, 665.

[2] Olcott, H. S., *J. Nutrition*, 1938, **15**, 221.

TABLE I.
Males.

Group	Normal	E-deficient diet			WGO* supplement		Concentrate† supplement	
Age, mo	6	5	7	8	7	8	7	8
Muscle tension in g/g	1763	1177	1168	1106	1166	995	1149	1112
No. of animals	12	4	2	2	3	3	2	4
Water, %	75.61	75.87	76.11	76.48	75.82	75.83	75.54	76.11
Cl, mg/100 g muscle	45.5	53.1	53.1	66.7	48.5	48.3	51.6	47.4

TABLE II.
Females.

Group	Normal	E-deficient diet			WGO* supplement		Concentrate† supplement	
Age, mo	6	5	7	8	7	8	7	8
Muscle tension in g/g	1842	1489	1397	1339	1426	1269	1379	1297
No. of animals	13	4	3	3	4	2	2	4
Water, %	75.51	75.56	75.81	75.98	75.08	75.19	75.42	75.53
Cl, mg/100 g muscle	46.1	49.8	45.2	53.7	44.8	45.2	46.7	46.2

* Wheat germ oil.
† Concentrate of wheat germ oil.

rats were similar to those reported by Evans, Emerson and Telford,[3] but are less extensive than those observed by Olcott,[2] Pappenheimer[4] and Goettsch and Ritzmann[5] in suckling rats of E-deficient mothers. In our series lesions were fully developed when the rats were first examined at the age of 5 months, and there was little, if any, change in the histological picture during the following 3 months. In no case were degenerative lesions present in more than a few fibers in any one section. The degenerative changes consisted of hyaline necrosis involving the entire fiber or merely a segment of the latter. The necrotic zone was usually infiltrated with large mononuclear cells and occasionally with a few neutrophilic and eosinophilic polymorphonuclear leukocytes. In some of the affected areas the necrotic tissue had been resorbed and the fibers were collapsed, but the sarcolemma was still distinct and numbers of large mononuclears and proliferating muscle nuclei were present. No inflammation of the interstitial tissues was seen, as in the more severe lesions described by Olcott[2] and by Pappenheimer.[4]

Along with these degenerative changes were to be seen evidences of regeneration of two types:

(1) An "early" form in which the muscle fibers were small, with a basophilic sarcoplasm, and with numerous nuclei, commonly centrally placed within the fiber. The nuclei were oval and vesicular, which implied active proliferation. Few myofibrils were seen in these fibers.

(2) A type in which the fibers were of nearly normal size with both longitudinal and cross striations, and with more slender nuclei. The nuclei occurred singly, in small groups, or in chains of 6 to 8, and were situated both in the subsarcolemmal region and centrally in the fiber. These changes were interpreted as representing "late" stages of regeneration. Neither in this group nor in the treated group described below was there any replacement fibrosis or adiposity within the muscles.

All treated animals, with one exception, showed complete absence of necrosis, and even this one case showed merely one necrotic fiber in 2 sections. In 4 other cases an occasional collapsed fiber was seen. It is possible that in these particular cases the original necrosis occurred before the supplement was added to the diet. In no case were regenerative lesions of the "early" form described above to be

[3] Evans, H. M., Emerson, G. A., and Telford, I. R., PROC. SOC. EXP. BIOL. AND MED., 1938, **38**, 625.

[4] Pappenheimer, A. M., *Am. J. Path.*, 1939, **15**, 179.

[5] Goettsch, M., and Ritzmann, J., *J. Nutrition*, 1939, **17**, 371.

found. Regenerative lesions of the "late" type were fairly common, approximately as common as in the untreated animals.

It is apparent from data given in the tables that the chloride and water concentrations, which were elevated in the E-deficient animals, returned to the normal range after treating the rats for 2 to 3 months with wheat germ oil or with vitamin E concentrate.

In marked contrast, the muscular power was definitely impaired by the E-deficient diet. This loss of power, as the tables show, was approximately 30%, and yet it was not so great as to handicap the animal in its normal cage activity. The muscular power, however, showed no improvement after feeding the vitamin supplements. This is of particular interest in view of the fact that muscles of the treated animals had normal concentrations of chloride and water, and showed no necrosis or "early" regenerative lesions. It is possible that all of the muscle fibers were weak, or that only those fibers showing "late" regeneration were weak. Evidently the physiological damage is something rather subtle which cannot easily be evaluated by histological or chemical methods.

Summary. Muscular dystrophy was produced in rats by feeding a diet deficient in vitamin E, beginning at birth. At the age of 5 months muscle necrosis and muscle regeneration was evident histologically and chemical studies showed elevation of the chloride and water concentrations of the muscles.

Subsequent addition of wheat germ oil or vitamin E concentrate to the diet for 2 to 3 months resulted in complete recovery with respect to the concentration of chloride and water in the muscle. Histologically the muscles showed practically no evidence of necrosis. Despite the chemical and histological evidence of recovery the muscles still lacked the ability to develop the normal amount of tension when tested by quantitative methods.