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Successive Generations of Vitamin E-Low Rats.*

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This study was undertaken to determine the effects of continued vitamin E deficiency upon growth, general well being and reproductive behavior in successive generations of vitamin E-low rats.

Thirty rats from the stock colony were placed on the standard vitamin E-low diet 427† at 21 days of age. They were bred for a trial gestation at the first pro-oestrus which occurred after the 60th day of age. All animals in the group underwent a typical resorption gestation. Twenty-five rats of this group were re-bred and were given a single dose of 1.0 g of wheat germ oil. All cast litters but in 3 cases all the young were dead. They were allowed to suckle their young.‡ Throughout the study each litter of more than 6 young was reduced to that number. Fifty-six from a total of 130 young survived 30 or more days; they are designated as second generation E-low rats. All were paralyzed to some extent. The paralyses in some cases were flaccid—in others spastic. The picture other than the occurrence of paralyses included such phenomena as the existence of a square "boxy" body form, puffy face and corneal films. The 56 animals that survived recovered from external signs of the dystrophy and the females were then handled as the first

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† Diet 427:

Casein, commercial, precipitated with HCl	27
Cornstarch (cooked)	35
Salts (McCollum 185)	4
Lard	22
Cod liver oil (Squibb's)	2
Brewer's yeast	10

The mixed diet without the cod liver oil is allowed to stand for 2 weeks at room temperature to permit the rancid substances of the lard to destroy incipient traces of vitamin E. The cod liver oil is added just before feeding.

‡ During the lactation period each female was given in addition to diet 427 1.0 g of brewers' yeast 6 times weekly.

generation animals, *i. e.*, they were allowed to undergo a resorption gestation and were given 1.0 g of wheat germ oil on the first day of their second gestation.

Of a total of 27 second generation E-low females bred, 26 cast litters (2 litters consisted of dead young) and 1 resorbed. Of a total of 134 young, 48 survived 30 or more days; these were the third generation of E-low animals. The clinical pictures presented were the same as for the young of the first generation mothers.

A total of 22 third generation females were bred resulting in 6 failed implantations, 1 resorption and 15 litters, 2 of which had only dead young. Of a total of 70 living young, 30 survived 30 or more days; these are designated as fourth generation E-low animals.

The fourth generation animals ovulated infrequently and only 7 females could be bred for the next generation. Two resorbed and a total of only 20 young were born, 6 of which survived 30 or more days.

One female of the fourth generation was given 0.15 cc of wheat germ oil daily from day 10 and was subsequently bred; 6 out of 6 young survived 30 days. The results are summarized in Table I.

The progressive debility of successive generations of E-low females is shown in Table II. The opening of the vagina and first oestrus appeared at more advanced ages in successive generations of vitamin E-low females. Growth was likewise affected as can be

TABLE I.
Reproduction and Lactation History of Vitamin E-low Female Rats.

Generation	No. of rats bred	No. of litters	No. of young suckled	No. of young surviving 30 days
1st	25	25	130	56
2nd	27	26	134	48
3rd	22	15	70	30
4th	7	5	20	6
4th (received 0.15 cc wheat germ oil daily)	1	1	6	6

TABLE II.
Progressive "Debility" of Successive Generations of E-low Females.

Generation	No. of rats	Avg wt 21 days	Avg wt 60 days	Avg age opening of vagina	Avg age at 1st oestrus
1	30	46.4	188	30*	35
2	30	40.1	159	42	51
3	24	39.3	137	47	57
4	6	32.5	125	55	63

*This is appreciably earlier than is usual in our colony.

seen from the average weight of 188 g at 60 days for the first generation when contrasted with 125 g for the fourth.

Growth and sexual physiology were studied in males of the second, third and fourth generations. The differences were not as marked as in the females but all showed the characteristic plateauing in weight. It is of interest to note, however, that 3 fourth generation E-low males were given 0.15 cc of wheat germ oil from day 10. At the age of 1 year the 3 animals receiving wheat germ oil averaged 110 g more in weight than did 8 untreated males of the same generation.

The males in each generation were offered oestrus females approximately twice monthly from the age of 4 months.

As can be seen from Table III, although the E-low males in all generations were sterile, the prophylactic administration of wheat germ oil prevented such changes when administered to 3 fourth generation males.

Note was made of the appearance of deficiency symptoms, other than sterility, in older males. (Table IV.) The fourth generation animals receiving wheat germ oil did not show these deficiency symptoms; they were also fertile, as has been mentioned. These facts are important because they appear to indicate that the observed progressive "debility" of the generations of vitamin E-low rats herein reported was due solely to inadequate E, adequacy of the diet in other respects being thus established.

TABLE III.
Sexual Physiology of Successive Generations of E-low Males.

Generation	No. of males	No. of offerings of oestrus females	No. of instances of plug	No. of instances of sperm	No. of young sired
2nd	27	384	147	0	0
3rd	15	273	117	0	0
4th	8	101	40	0	0
4th (received 0.15 cc wheat germ oil daily)	3	18	11	18	142

TABLE IV.
Deficiency Symptoms Other Than Sterility in E-low Male Rats.

Generation	No. of animals	Sparse, coarse or yellowish* fur Age in days	Stiff gait noted Age in days
2nd	27	194	277
3rd	15	121	178
4th	8	100	150

*The fur of black rats turned a reddish brown.

Summary. Four generations of vitamin E-low rats were observed as to growth and reproductive behavior. In the female there was a delay in maturity in each successive generation. Growth was also affected.

The males in each generation were sterile but fertility was maintained in the fourth generation by the prophylactic administration of wheat germ oil.

Coarse, sparse, yellowish fur and dystrophy that appear in rats maintained on vitamin E-low diets appeared progressively earlier in each successive generation.

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A Virus from Cases of Influenza-like Upper-respiratory Infection.

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During the past few years at least two groups of investigators[1,2] have reported cases of upper-respiratory infection of unknown etiology, that resembled true "epidemic influenza" in clinical respects but which differed in that the convalescent serums failed to show any increase in capacity to neutralize standard strains of the influenza virus. Two similar cases of influenza-like infection occurred among the workers in this laboratory in February, 1940. The serums obtained from these cases 4 weeks after the infection and those obtained either at the time of or 3 weeks before the onset of illness were tested against the PR8 strain of the virus of epidemic influenza. Neither of the convalescent serums showed any detectable increase in capacity to neutralize the usual 1000 lethal doses of the virus; and neither of them fixed complement in tests against antigens prepared in the usual manner[3] from mouse lung suspensions. This apparent lack of development of antibodies reactive against this standard strain of influenza virus seemed to indicate that the influenza-like infection of neither of the cases had been due to the virus of epidemic influenza.

1 Reimann, H. A., and Stokes, J., Jr., *Tr. Assn. Am. Physn.*, 1939, **54**, 123.

2 Stuart-Harris, C. H., Smith, W., and Andrewes, C. H., *Lancet*, 1940, **1**, 205.

3 Francis, T., Jr., Magill, T. P., Rickard, E. R., and Beck, M. D., *Am. J. Pub. Health*, 1937, **27**, 1141.