

tance when, in truth, the causative factor is transmitted by both sexes and under strict Mendelian rules. The location of the factor within the chromosome is of interest, as the results show it to be at or near the point of this V-shaped chromosome where presumably the spindle fiber attachment takes place. This fact may be significant in view of the evidence accumulating to show that the spindle fiber attachment is frequently associated with abnormalities in chromosome behavior. The frequency of occurrence of these mutant forms points to their random distribution within any female's progeny and, therefore, that the non-disjunction of the chromosomes occurs in the last oögonial divisions or later. The non-disjunction generally involves both the paternal and maternal sex chromosomes and not, as might be the case, only paternal or maternal chromosomes.

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#### REGULATING THE STORAGE OF VITAMIN A IN ANIMALS THAT ARE TO BE USED FOR THE DETERMINATION OF THIS VITAMIN

THE following observations and considerations led to a study of the problem of the storage of vitamin A in rats reared under carefully controlled conditions:

(1) In a previous study of this problem by Steenbock, Sell and Nelson,<sup>1</sup> the authors were cognizant of the possible influence of the consumption of the stock diet by the young on the storage of vitamin A.

(2) It is extremely difficult to have a uniform supply of A in any stock diet. The instability of the vitamin and its consequent variation in natural foods, particularly foods in such a state of comminution as to prevent selection from the diet, is one of the problems to be dealt with.

(3) Keeping animals on a diet which is low enough in vitamin A to insure a low storage in the young may interfere with satisfactory reproduction if the level becomes just a little too low.

(4) Some data obtained in studies in this laboratory on a prophylactic technique of vitamin A determination in which the rats were given a single or limited number of feedings of vitamin-containing material were difficult to interpret because of the variations in the time of onset of symptoms of vitamin A deficiency.

(5) If mother rats are kept on a diet which insures a very generous supply of A, so that the milk at all times contains a maximum amount of this vitamin and the young receive no other vitamin A than that which is contained in the mother's milk

before they are put on a vitamin A free diet, will such young be suitable for vitamin A studies?

Information on the storage of vitamin A in young rats was obtained under the following conditions: Stock rats were bred when kept on a diet that included a generous supply of cod liver oil known to be very potent in vitamin A. Pregnant females were segregated. Litters of six animals were used, and the number of rats in a litter was reduced when necessary. Mothers were put on an A-free diet, some the day the young were born and others when the young were five, ten, fifteen, sixteen, seventeen, eighteen, nineteen and twenty days old. When the young had attained a weight of from forty to forty-five grams they were weaned and put on the A-free diet. If the mother is put on the A-free diet when the young are sixteen to eighteen days of age and weigh not less than twenty-seven or more than thirty grams, and the young are weaned when they weigh from forty to forty-five grams, the onset of symptoms of vitamin A deficiency in the young seems to be independent of the amount of vitamin A given the mother during the entire period of lactation, provided a certain minimum level is not reached. On such a regimen, for the past six months the animals in this laboratory have shown definite symptoms of vitamin A starvation between the fifth and sixth week after weaning. Ophthalmia from vitamin A deficiency seems to occur with the greatest degree of regularity in rapidly growing animals, and our experience indicates that animals do not grow rapidly if the mother is put on the A-free diet before the young are fifteen days old. It is possible that this retardation in the growth of rats can be eliminated by adjusting the diet of the mother, but for the results desired this seems to be unnecessary. Animals that weigh much more than thirty grams will eat the stock diet. All rats which were put on the A-free diet at the age of nineteen days or more showed a distinct delay in the onset of ophthalmia as compared with those which were put on the A-free diet before that age.

It seems very probable that by following the essentials of the technique outlined herein with rats reared in any laboratory the storage of vitamin A will be comparatively uniform. There is the added advantage of maintaining the stock animals on a diet which is known to be adequate in every respect. There has been no indication that the time the females are kept on the A-free diet (from five to eight days) is long enough to deplete her reserves of A to the extent that it becomes apparent.

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<sup>1</sup> Steenbock, H., Sell, M. T., and Nelson, E. M. *J. Biol. Chem.*, Vol. 56, p. 327. 1923.