CCLXXXII. THE TRANSMISSION OF VITAMIN A FROM PARENTS TO YOUNG IN MAMMALS.

IV. EFFECT OF THE LIVER RESERVES OF THE MOTHER ON THE TRANSMISSION OF VITAMIN A TO THE FOETAL AND SUCKLING RAT.

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IN an earlier paper [Dann, 1932] a preliminary account was published of the influence of several factors on the amount of vitamin A stored by the young rat up to the time of weaning. One of the conclusions reached at that time was stated as follows: "The amount of vitamin A in the liver of the rat at birth is small and subject to little variation. It cannot be increased by giving large amounts of carotene to the mother before or during pregnancy." Further, "It is probable that the vitamin A reserve of the mother at parturition... exerts an effect on the vitamin A content of the milk but it is... not large enough to show up unequivocally in the work described." In the experiments from which these conclusions were drawn carotene had been used for feeding to the does, since vitamin A concentrates were not available at the time in sufficient quantity. The highest liver reserve of vitamin A accumulated by one of the experimental does under such conditions was 22,000 B.U. In later work [Dann, 1934] does were given vitamin A concentrate in the diet over considerable periods, in place of the carotene; their reserve stores then rose as high as 300,000 B.U. of vitamin A. As a result it was found that when the doe's liver contained such large amounts of the vitamin, the vitamin A in the liver of the foetus at birth was increased to an amount of about 30 B.U. per foetus. No observations were made to show the effect of the vitamin A reserves of the mother on the amount of vitamin A transmitted to the suckling in the milk.

The present paper deals with the effects of the vitamin A reserves of the mother on the amount of vitamin A which can be transmitted to and retained by the young rat up to the time of weaning. The transmission during the periods of gestation and suckling are not considered separately; the reserves stored by the young rat during the whole period from conception to weaning have been examined in their relation to the mother's reserve.

EXPERIMENTAL.

The work to be described comprises a study of 38 piebald does and their litters and was carried out over a period of a year. The does were divided into two groups, which received different dietary treatment in order that they should

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have widely different reserve stores of vitamin A. All does of group A received after weaning for the remainder of their lives a stock diet consisting of bread, boiled wheat, mixed corn, meat and carrots, cabbage or lettuce when available. Roughly skimmed milk was the only fluid provided. The does of group B received

Table I. *The transmission of vitamin A to young rats before weaning.*

Data obtained by the examination of the livers of young rats at weaning, and of their mothers. 38 litters were examined and all figures are included without selection; arranged in order of increasing reserves of the mother.

Doe	Total B.U. in liver	Wt. liver	B.U./g. liver	No. of young reared	Total B.U. in livers of litter	Av. B.U. per member of litter	Total B.U. stored by litter as % of doe's reserve	Av. B.U. of young as % of doe's reserve
B 14	560	8.7	64	6	15	2.5	2.7	0.45
B 15	600	8.1	74	7	10	1.5	1.6	0.23
A 21	750	7.75	97	4	50	12.5	6.6	1.6
A 23	875	10.2	86	4	20	5	2.3	0.56
A 24	1,000	7.9	127	6	125	21	12.5	2.1
A 27	1,250	11.2	112	12	200	17	16	1.3
A 22	1,500	10.5	143	3	30	9	2	0.66
A 25	1,500	7.5	200	9	100	11	6.6	0.73
A 26	1,875	10.9	172	6	50	8	3	0.5
A 11	2,000	—	—	3	20	6	1	0.33
A 31	2,750	9.3	296	5	15	3	0.5	0.1
A 44	5,000	7.5	660	8	60	7.5	1	0.13
A 32	5,500	8.5	647	8	135	17	2.5	0.3
A 43	5,625	5.0	1125	7	70	10	1.3	0.2
B 42	6,250	8.75	730	4	125	31	2	0.5
A 49	6,875	—	—	6	150	25	2.1	0.35
A 45	7,500	9.0	833	9	18	2	0.25	0.03
A 48	10,000	11.75	850	11	120	11	1.2	0.1
A 47	11,250	11.75	960	4	75	19	0.6	0.15
B 46	11,250	13.25	850	9	450	50	4	0.5
B 45	12,500	10.25	1220	9	225	25	2	0.2
B 47	12,500	10.25	1220	7	190	28	1.8	0.25
B 23	12,500	11.4	1090	7	150	21	1	0.13
B 13	12,500	8.5	1470	4	10	2.5	0.08	0.02
A 41	12,750	10.5	1210	8	60	7.5	0.5	0.06
B 22	15,625	12.2	1280	8	350	44	2.2	0.27
A 46	18,750	9.9	1900	6	150	33	0.9	0.15
B 21	22,500	7.5	3000	1	20	20	0.1	0.1
B 32	24,750	9.7	2550	7	190	27	0.6	0.1
B 11	25,000	11.8	2100	8	250	31	1	0.12
B 12	30,000	11.3	2660	9	100	11	0.3	0.04
B 43	31,250	8.0	3905	3	125	42	0.2	0.06
B 48	31,250	9.5	3280	4	125	31	0.2	0.05
B 34	36,750	8.5	4325	6	290	48	0.4	0.06
B 35	52,500	7.5	7000	6	320	53	0.6	0.1
B 31	55,000	8.8	6250	7	235	33	0.4	0.05
B 16	62,500	11.6	5400	9	600	66	0.9	0.1
B 33	73,500	10.1	7280	6	250	41	0.3	0.05

the same stock diet up to the age of 10 weeks, and then for 4 weeks they were given a diet made up fresh daily of 13 parts basal diet *plus* 2 parts vitamin A solution and milk to drink. The basal diet contained maize meal 1060, whole milk powder 400, whole grain flour 200, dried yeast 200, wheat germ 60, calcium phosphate 60 and salt mixture 20 parts. The vitamin A solution was prepared by dissolving 1 g. of vitamin A concentrate (B.D.H.) in 200 g. of arachis oil. Thus the diet prepared daily was very rich in vitamin A, containing 0.066 % of the concentrate. At 14 weeks of age the group B does were put back on the stock diet again and remained on that diet until the end of the experiment.

Table II. *The transmission of vitamin A to young rats before weaning.*

In this table the litters dealt with singly in Table I are arranged in six groups and the mean figures for each group set out (amounts of vitamin A expressed in B.U.).

Group	Range of liver reserves of does in group	No. of does in group	Mean liver reserves of does	Range of av. liver reserves of litters	Mean of liver reserves of all young in group
1	0- 1,000	5	757	1.5-21	8
2	1,001- 5,000	7	2,296	3 -17	10
3	5,001-10,000	6	6,960	2 -31	12
4	10,001-20,000	9	13,290	2.5-50	28
5	20,001-40,000	7	28,800	11 -48	29
6	Over 40,000	4	60,900	33 -66	50

At 14 weeks the does were mated and when seen to be pregnant were segregated to await the birth of the litters. The young were allowed to grow until the eighteenth day when the young and their mother were killed and the liver reserves determined in the manner previously described [Dann, 1932]. Generally the livers of all the animals in one litter were pooled and the total vitamin A content determined. Some litters on the other hand were treated differently—a number of the young were examined individually for vitamin A reserves, and the livers of the remaining animals were pooled for examination. It will be noticed that the young animals were killed for examination about 3 days earlier than the usual weaning time. This procedure is adopted as a precaution against any of the young eating food provided for the doe, and so obtaining vitamin A from a source other than the mother's milk.

Results.

The results are summarised in Tables I, II and III. Table I gives the average vitamin A reserve of the young in each litter irrespective of whether all the livers were pooled; the figures are arranged according to the vitamin A reserve of the doe when she was killed after suckling her litter for 18 days. The liver reserves of the does examined varied from 560 to 73,000 B.U. of vitamin A. The average liver reserves of the members of one litter varied from 1.5 to 66 B.U. In general, the higher the reserves of the doe, the higher the reserves of the young, but the results are very scattered. If however the does are divided into groups according to the size of their liver reserves, and mean figures for the reserves of all the offspring in each group are taken, the figures set out in Table II are obtained. Table III includes all the data obtained by the examination of individual members of the litters.

Table III. *The variation in vitamin A reserves among rats from the same litter at 18 days (B.U.).*

Doe	Liver reserve of 1st pup	Liver reserve of 2nd pup	Liver reserve of 3rd pup	Re-maining young in litter	Total liver reserves of group	Av. liver reserve of each member of group	Av. liver reserve of each member of the whole litter
B 31	20	20	20	4	175	44	33
B 32	20	25	20	4	125	31	27
B 33	25	35	60	3	125	42	41
B 34	60	20	60	3	150	50	48
B 35	30	80	60	3	150	50	53

DISCUSSION.

The results obtained in the experiment described show that there is some proportionality between the vitamin A stores of young rats at the end of the suckling period and the vitamin A reserves of their mothers. This rough proportionality is clearly shown by the mean figures set out in Table III, which are plotted in Fig. 1, but two points need attention. First, the graph suggests that the mean value of the vitamin A store in the livers of young rats 18 days old, if born of and suckled by does with no vitamin A reserves would be about 7 B.U. Secondly, the individual figures from which the means are calculated are a good

Fig. 1. Relationship between vitamin A stored by the young rat before weaning and the mother's reserves of the vitamin. Each point plotted represents a mean for all animals in one of six groups arranged according to the amount of the doe's reserve.

deal scattered. It seems probable that both of these circumstances may be attributed in part to the particular conditions of the present work. The object of the work was to determine the effect of the doe's liver reserves on transmission of vitamin A to the young by healthy parents subsisting on a normal diet. The basal diet given contained small but normal quantities of vitamin A and carotene, and it seems likely that some of the vitamin A ingested by the doe should enter the circulation and be transmitted *via* the placenta and the mammary glands to the young irrespective of the reserve store. Further, since the does were given abundant rations, they would be in a position to exercise choice between the various articles of diet provided, and it is possible that some would ingest much more carotene and vitamin A than others. These considerations might account in part at least for the scatter of the results obtained, and would also allow for the possibility of vitamin A being transmitted to their young by does with no vitamin A reserves. This explanation can only be regarded as tentative for the present, but it can be tested by a repetition of the work in which the parents are given a vitamin A-free diet during gestation and lactation. This is now being done.

Individual variation in ability to store vitamin A. The extent to which this scattering of the results obtained can be accounted for by the suggested explanation must await further experimental observations. There are however indications in the results already obtained that with does having equal liver reserves and the same daily intake of vitamin A, the amounts of vitamin A

transmitted to the young would still vary considerably from animal to animal. It is well that this should be clearly stated, as the assumption that animals having equal opportunities will amass equal reserves of vitamin A appears to be made by a number of workers. Thus Richards and Simpson [1934] make the implicit assumption that rats "with exactly similar opportunities for storing vitamin A" will store equal amounts of the vitamin. Such an assumption takes no account of individual idiosyncrasy, the existence of which can be seen from the present work. Thus, all the group A does had "similar opportunities" to store vitamin A, but when examined their vitamin A reserves varied from 750 to 18,750 B.U. The same applies to the does of group B, whose reserves varied from 560 to 73,500 B.U. In this group two of the does had reserves far lower than any of the remainder, but even if they are rejected on grounds of possible abnormality the range of variation among the others is from 6250 to 73,500 B.U.

This individual variation among animals similarly fed under the same conditions is most strikingly demonstrated by comparing young animals from the same litter. Some figures are given in Table III for 5 of the litters of group B. The liver stores of three members of each litter were measured singly, and the livers of the remainder were pooled for examination. As much as threefold variation between the liver stores of members of the same litter was found, but further work is needed to determine the usual variation found within the litter, and this is at present in progress.

The drain on vitamin A reserves of the doe by the litter. It is clear from the figures given in Table I that there is no clear proportionality between the size of litter and the average store of individual members of the litter. Thus the members of a large litter appear just as likely as the members of a small litter to have relatively large stores of vitamin A at weaning. Therefore the total amount of vitamin A present in the livers of all the members of a litter at 18 days old shows less clear proportionality to the reserves of the doe than does the average store of the individual young rat. The amount of vitamin A present in all the members of a litter at 18 days old is however generally small in the present experiment, even though, as has been suggested above, part of it may have come direct from the supply in the doe's diet independently of her reserves. Only four litters had each stored an amount of vitamin A greater than 3 % of the doe's reserve. The average store of single members of a litter at 18 days old varies among the litters described from 2.1 % to 0.02 % of the doe's reserve and, in general, when expressed as a fraction of the doe's reserve, decreases as the doe's reserves increase. It is plain that in the rat the rearing of a litter on a normal diet is unlikely to cause a dangerous drain on the mother's reserve of vitamin A.

SUMMARY.

1. When female rats bear and suckle their young on a mixed diet of natural foods, the store of vitamin A amassed by the young rat up to the time of weaning is roughly related to the vitamin A reserves of the mother. The amount of the vitamin stored by the young is very small compared with the doe's reserve; thus under the conditions examined if the doe's reserve were 1000 B.U. each young rat would be likely to store about 10 B.U., and if the doe's reserve were 50,000 B.U. each of her young would store about 40 B.U. of vitamin A.

2. The figures obtained show considerable scatter, which is attributed to (a) variation in amount of vitamin A ingested by the doe during gestation and lactation owing to her exercise of choice among the articles of diet provided, and (b) individual idiosyncrasy.

3. Evidence obtained in the course of the work suggests that the individual idiosyncrasy is an important factor; and that the amount of vitamin A stored by rats having equal opportunities of obtaining it may vary considerably.

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