CCIII. THE VITAMINS PRESENT IN OX-SERUM. I.

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IN the investigation described below the presence of vitamins A and B in ox-serum was sought for by means of biological tests.

Sherman and Boynton [1925] have already shown that rat-blood contains vitamin A.

Its presence in serum is a matter of inference, rather than of direct evidence, from the experiments of Euler and Virgow [1932], who showed that the carotene isolated from serum possesses vitamin A activity.

Vitamin B₁ was found absent from blood [Kollath, 1930] while vitamin B₂ was found to be present [Hoagland, 1923].

TECHNIQUE.

The experiments were carried out with white rats, all being pure bred Wistar stock and reared at the laboratory. The young were put on to the experimental diets when 3-4 weeks old and weighing between 30 and 40 g.

Four diets were used in the experiments having the following composition: the quantities given are the daily rations per rat in g. (or cc. in the case of liquids).

	Diet 1	Diet 2	Diet 3	Diet 4
Caseinogen	3	2½	3	2½
Hardened cottonseed oil	2	2	2	2
Rice starch	10	10	10	10
Lemon juice	1	1	1	1
Salt mixture	1	1	1	1
Dried yeast	1	1	1	0
Irradiated cholesterol	1/200	1/200	1/200	1/200
Cod-liver oil	1/20	0	0	1/20
Ox-serum	0	10	0	10
Vitamin content of diet	ABCD	BCD + serum- vitamins	BCD	ACD + serum- vitamins

The salt mixture was based on a formula by McCollum and Davis and suggested by Pryde [1928]. It has the following composition:

	%
NaCl	4.7
MgSO ₄ .7H ₂ O	7.1
NaH ₂ PO ₄ .H ₂ O	9.4
K ₂ HPO ₄	25.8
CaH ₄ (PO ₄).H ₂ O	14.6
Ca lactate	35.2
Fe citrate	3.2
KI	Trace (0.003 %)
	100

The cholesterol used was activated by dissolving 2 g. in 100 cc. warm light petroleum and exposing to rays from a mercury vapour quartz lamp at a distance of 12 ins. for 30 minutes. 40 cc. liquid paraffin were then added, and the ether

Table I.

No. of rat	Diet	Vitamins in diet	Initial weight	Gain in weight	Period of observation (days)	Fate of rat
22	1	A, B, C, D	41	159	56	Survived, healthy
23	1	"	43	142	56	"
24	1	"	43	172	56	"
47	2	B, C, D and serum	29	105	55	"
48	2	"	32	136	55	"
49	2	"	30	141	55	"
57	2	"	38	147	56	"
58	2	"	36	109	56	"
59	2	"	36	179	56	"
60	3	B, C, D	39	39	52	Ataxic
61	3	"	32	28	57	Died
62	3	"	37	51	54	"
50	4	A, C, D and serum	32	—	24	"
51	4	"	32	3	25	"
52	4	"	33	5	25	"

Fig. 1. Chart showing that serum contains vitamin A but not the vitamin B complex.

was evaporated off on a steam-bath. The resulting solution contained 5 mg. irradiated cholesterol in 0.1 cc. oil. This amount was administered daily to the rats by means of a graduated pipette.

In the case of rats receiving cod-liver oil in addition to irradiated cholesterol, a mixture was prepared, 0.1 cc. of which contained 5 mg. of cholesterol and

0.05 cc. cod-liver oil. This was obtained by adding 20 cc. liquid paraffin and 20 cc. cod-liver oil to the irradiated solution of 2 g. cholesterol in 100 cc. light petroleum and evaporating off the ether.

The diets were so prepared that the addition of serum did not increase the calorific value of the diet. Thus when a daily ration of 10 cc. serum was allowed, the corresponding amount of protein was deducted from the caseinogen ration. As the serum used had a protein content of 7.5 % (as determined by the Kjeldahl method), a 10 cc. ration represented 0.75 g. protein: rats receiving serum were therefore given 2.25 g. caseinogen instead of 3 g. daily.

In the latter part of the experiment lemon juice was not added to the diets. This omission, however, was not made until it had been definitely established by other experiments that lack of vitamin C did not affect the rats in weight or in any other way over a period of several months longer than the duration of the present experiment (two months).

15 rats were used in the investigation: 3 were put on diet 1 (complete in vitamins); 6 on diet 2 (containing serum instead of vitamin A); 3 on diet 3 (containing no vitamin A); 3 on diet 4 (containing serum instead of vitamin B). The rats were weighed at least twice a week.

The results are indicated in Table I and Fig. 1.

RESULTS.

Vitamin A. From Table I and the accompanying chart it is clear that serum contains vitamin A, since when serum is added to a diet deficient in vitamin A growth proceeds normally.

Since the average rate of growth of the rats was 2.8 g. per day on diet 1 and 2.5 g. per day on diet 2, the vitamin A content of 10 cc. serum is approximately 90 % of that of 0.05 cc. cod-liver oil; *i.e.* the vitamin potency of the serum used was 1/200 (0.45 %) that of the sample of cod-liver oil used.

Vitamin B. As the animals on diet 4 died in 3–4 weeks with scarcely any gain in weight, it is clear that the vitamin B complex is not present in serum. The possibility exists, however, of one or other of the components of vitamin B being present in serum.

CONCLUSION.

Ox-serum contains vitamin A but does not contain the vitamin B complex.

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