XLI. THE PHOSPHOLIPIN OF THE BLOOD AND LIVER IN EXPERIMENTAL RICKETS IN DOGS.

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IN a paper in which he advances a theory of the metabolism in rickets, Prof. Noël Paton [1922] points out that recent work seems to indicate that the supply of phosphorus rather than of calcium may be the limiting factor in ossification and develops the idea that a disturbance in the metabolism of the phospholipins may be involved.

Since the phospholipins and especially lecithin play so important a part in the metabolism of phosphorus an attempt has been made to discover whether there is any modification in the amount in the blood and liver in rickets.

Unfortunately, the material available has not been of such a nature as to enable a conclusive answer to be given, for in each of the experiments in which the blood and liver were preserved for analysis, either all the pups remained free of rickets or all developed the condition. The results obtained are however sufficient to justify their being recorded and to allow a tentative conclusion to be drawn.

A considerable amount of work has been recorded upon the phospholipin of the blood but little of it has a bearing upon the present investigation. Bloor [1915] showed that in feeding with fat the lecithin content of the blood is raised and [1918] that this is due to an increase of phospholipin in the corpuscles.

METHOD.

The blood and the livers were taken at the time of death by chloroform and bleeding, from experiments 5, 6, 7 and 8 of the series described by Noël Paton and Watson [1921]. The details of experiment 12 have not yet been published, but all the pups remained free from rickets. Unfortunately the non-phospholipin phosphoric acid was determined only in lots 7 and 8.

METHODS OF ANALYSIS.

Livers. The livers were preserved in formalin. A weighed amount was repeatedly extracted with alcohol-ether (10-1) by rubbing up in a mortar. The extract was dried in a steam-bath and in a vacuum desiccator and weighed and from this total fats and alcohol-ether soluble P_2O_5 were determined. The alcohol-ether-insoluble P_2O_5 was estimated in the residue. Each was ignited

NORMAL.

I. Pups of VII killed at 16 weeks old, those of VIII at 18 weeks old.

Experiments	VII. B 7	VIII. B 5	VIII. B 4	VII. D 4	VIII. D 2	VII. D 5
Diet per diem	...	Separated milk (dried) 22.5 g. Bread 40 g.	Separated milk (dried) 22.5 g. Bread 30 g. Lard 4 g.	...	...	Full cream milk (dried) 22.5 g. Bread 30 g. 366
Gain in weight %	...	620	280	240	600	300
Total fats %	...	Blood 2.94 Liver 2.19	Blood 1.60 Liver 1.2	Blood 1.25 Liver 2.45	Blood 2.12 Liver 3.4	Blood 2.04 Liver 2.45
Total P ₂ O ₅ %	...	0.140	0.098	0.085	0.085	0.113
P ₂ O ₅ in phospholipins %	...	0.049	0.098	0.027	0.161	0.086
P ₂ O ₅ insoluble in alcohol %	...	0.091	0.063	0.213	0.058	0.038
P ₂ O ₅ in phospholipins as % of total fats	...	1.7	2.1	2.1	1.2	0.075
						1.9
						4.4
						1.8
						3.9

II. Pups killed at 15 weeks old.

	XII				
	1	2	3	4	5
Diet per diem	...	Full cream milk (dried) 22.5 g. increased to 34 g. Bread 30 g. increased to 50 g.	...	...	...
Gain in weight %	...	200	120	200	50
Total fats %	...	Blood 1.86 Liver 5.5	Blood 1.32 Liver 5.8	Blood 2.15 Liver 2.15	Blood 1.90 Liver 1.90
P ₂ O ₅ in phospholipins	...	0.052	0.209	0.320	0.042
P ₂ O ₅ in phospholipins as % of total fats	...	2.8	3.2	3.6	2.2

RICKETS.

Pups of V killed at 17 weeks old, those of VI at 13 weeks old.

Experiments	VI. B 5	V. B 1	V. B 2	VI. D 2	VI. B 6	V. D 5	VI. D 3
Diet per diem	...	Whole milk 250 cc. Oatmeal 45 g. after 5th week	Separated milk 250 cc. after 5th week	Whole milk 250 cc. Oatmeal reduced to 150 cc. after 5th week	Whole milk 250 cc. Oatmeal 45 g. after 5th week	Whole milk (dried) 30 g. Oatmeal 45 g. after 5th week	Separated milk 250 cc. reduced to 150 cc. Oatmeal 45 g. after 5th week
Gain in weight %	...	156	22	116	161	135	65
Condition	...	Slight rickets	Marked rickets	Rickets	Rickets	Slight rickets	Rickets
Total fats %	...	Blood 0.97 Liver 3.17	Blood 1.12 Liver 1.90	Blood 1.11 Liver 2.83	Blood 1.15 Liver 2.76	Blood 0.96 Liver 2.94	Blood 0.80 Liver 2.84
P ₂ O ₅ in phospholipin %	...	0.022	0.076	0.024	0.026	0.024	0.019
P ₂ O ₅ in phospholipin as % of total fats	...	2.2	2.4	2.1	2.3	2.5	2.3
							3.3

with an excess of phosphorus-free calcium carbonate, the residue dissolved in hydrochloric acid with the addition of a little nitric acid and the phosphorus estimated by the molybdate method in the usual way.

Blood. The blood was drawn into a measured quantity of methylated spirit, well shaken and allowed to stand several days. This was filtered into a weighed flask and the residue extracted with alcohol-ether (10-1) many times, the washings being added to the filtrate. This was evaporated and the total fat and the P_2O_5 determined as in the liver.

SUMMARY OF TABLES.

P_2O_5 in Phospholipin.

	Non-rachitic			Rachitic		
	Maximum	Average	Minimum	Maximum	Average	Minimum
Blood	0.059	0.043	0.026	0.033	0.025	0.019
Liver	0.320	0.162	0.096	0.100	0.082	0.068

CONCLUSIONS.

The tables show:

1. That the percentage of phospholipin in the blood and in the liver bears no relation to the gain in weight, to the amount of fat in the diet, to the total fat in blood and liver or to the age of the animal.
2. That in the rachitic pups the phospholipins of the blood and of the liver are lower than in the non-rachitic.
3. That the amount of phospholipins of the liver is smaller in proportion to that of the blood in rachitic than in non-rachitic animals.

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