XLVI. THE PART PLAYED BY IRON AND FAT IN THE RECOVERY OF RATS FROM CHRONIC EXPERIMENTAL ANAEMIA.

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IRON.

THE problem of the degree to which inorganic iron was absorbed from the alimentary canal was one which attracted a great deal of attention in the years between 1885–1900, a fact which was largely due to the work of Bunge. Bunge's view was that inorganic iron was not absorbed, though the administration of such might indirectly aid absorption of organic iron by uniting with the sulphuretted hydrogen in the alimentary canal, thus screening the organic iron. In later years Bunge seemed to give up this view.

Nevertheless there still seemed to be some point in considering in the light of new knowledge the factors which influence the absorption of iron, and other experiments which were recently performed and which have already been described in this Journal [Scott, 1923; Scott and Barcroft, 1924] gave me the opportunity of doing so and lead up rather naturally to it.

It has been stated by Whipple [1918] that in his experiments administration of inorganic iron salts had no effect upon blood regeneration in simple anaemia in dogs. This conclusion is at variance with clinical experience, with earlier experimental work summarised by Sherman [1907], and with the recent experimental results of Schmidt [1912] on mice, Brinchmann [1921] on guinea-pigs, and Hirasawa [1923] on rabbits. In the case of rats made anaemic by breeding on diets of bread and milk [Scott, 1923], the results in Table I demonstrate that inorganic iron has the effect of hastening the cure of the anaemia. When, from the age of 8 weeks, ferrous chloride is given at the rate of 20 mg. per rat per day a cure is reached by the age of 16 weeks. The result shown has been obtained repeatedly and without exception, and the contrast between the blood of the treated animals and the controls is very striking even to the naked eye. The cure is slightly less rapid than that which occurs when green leaves are added to the bread and milk but anticipates the cure which occurs naturally at the age of about six months when the rats are kept on bread and milk alone [Scott, 1923]. Thus the results of the last three workers cited above are confirmed in rats but one has yet to bring the

results of Whipple's experiments into the general scheme. They appear to indicate the contrary conclusion; but a little consideration will show (1) that administration of inorganic iron cannot be expected to have much effect unless the diet is already poor in iron and (2) that the methods adopted by Whipple for producing anaemia may not have been sufficiently drastic seriously to deplete the reserve stores of iron in the tissues; Whipple's animals always had these reserves to draw upon and consequently the administration of iron to them was a work of supererogation.

Table I. *Showing effect of inorganic iron on blood regeneration in anaemic rats at 16 weeks of age. Diet—white bread and whole milk; 20 mg. ferrous chloride added per rat per day.*

Haemoglobin %	Red blood count in millions	Colour index (human = 1)
103	9.98	.52
101	9.46	.53
95	8.80	.54
98	9.12	.54
107	10.02	.53
98	9.62	.51
98	8.74	.56
96	8.15	.59
94	9.47	.50
Average 99	9.26	.53
Average of controls at same age on bread and milk without iron		
55	10.03	.28

FAT.

There seems to have long been an idea that fat is connected in some way with blood formation. References are given by Stockman [1895]. It has been shown by Geiling and Green [1921] and Jencks [1922] that in rats regeneration of red blood corpuscles after a single or double haemorrhage, especially the latter, occurs more rapidly if the animals be on a diet rich in vitamin A. This observation has been confirmed in the case of experimental anaemia in rats, for green leaves and (unpublished experiments) bird seed accelerate the natural cure which occurs on a bread and milk diet at about six months [Scott, 1923]. Hamburger [1921, 1922] has brought forward presumptive evidence that fat and lecithin are necessary for blood regeneration in rabbits. The lipaemia that follows on haemorrhage in these animals, possibly a prelude or accompaniment of blood regeneration, is of course well known [Bloor and Macpherson, 1917; Horiuchi, 1920]. Hamburger's experiments however lend themselves equally well to the interpretation that a supply of vitamin A is essential for blood regeneration. Further evidence has been brought forward that in rats the supply of fat in the diet is comparatively unimportant, the important thing being the supply of fat-soluble A [Drummond, 1920, 1921; Osborne and Mendel, 1920]. Geiling and Green [1921] find that in rats on a fat-deficient diet regeneration after double haemorrhage occurs in normal time. The same

criticism may be directed against a too rigid interpretation of this work as was levelled above in the case of Whipple's work; but the evidence cited so far would lead one to suppose that it is not fat but vitamin A that is essential in blood regeneration. This constitutes the evidence one has to build up and it is somewhat contradictory.

Large numbers of rats with a sub-chronic anaemia could be obtained and it seemed that the following inquiries might aid in further elucidating the matter:

1. Does the fat of milk play a part in the recovery of rats from the sub-chronic experimental anaemia described?
2. Can equally good recovery be obtained if a hydrogenated fat be substituted for milk fat?

Anaemic rats which had been fed on bread and whole milk were placed at 12 weeks of age on a diet of bread and skimmed milk plus 20 mg. of ferrous chloride and 2 g. palm kernel oil per rat per day. Table II demonstrates the findings at 20 weeks of age.

Table II. *Showing effect of bread and skimmed milk plus 20 mg. ferrous chloride and 2 g. palm kernel oil per rat per day on blood regeneration in anaemic rats at 20 weeks of age.*

Haemoglobin %	Red blood count in millions	Colour index (human = 1)
61	10.03	.30
61	9.67	.31
58	9.43	.31
68	10.32	.33
58	9.24	.31
61	11.21	.27
63	9.82	.32
51	8.96	.28
Average 60	9.84	.30
Average of controls on bread and whole milk and ferrous chloride		
98	8.68	.56

CONCLUSION.

Iron. The diet on which this anaemia develops in rats is a comparatively iron-poor diet; on this diet natural cure occurs slowly as the animals get older. Since this is so and since the sole addition of iron to the diet results in a hastening of the natural process of cure it may be concluded that the anaemia is due to deficiency of iron in the diet. It is therefore another deficiency disease.

Fat. In the case of rats fed on bread and milk with or without the addition of iron the milk fat or something associated with it is necessary for blood regeneration, but the evidence is insufficient to decide whether its absence or the diminished supply of vitamin A leads to a state of chronic under-nutrition, whether its function lies in facilitating absorption of iron [Macallum, 1894] or whether it is necessary for the functioning of the haemopoietic tissues or

directly for the construction of the haemoglobin molecule. One must admit the possibility of the first explanation for the reasons that on this diet (1) they show signs of under-nutrition in their rough coats, and (2) they show dilated stomachs, presumably because they have to take a large bulk of food to secure a sufficiency of calories. One must therefore be cautious not to claim too much from these experiments, but they do demonstrate the broad (and self-evident) fact that iron will not effect a cure if the diet be otherwise inadequate.

It is of interest that this result should be obtained, for it provides an experimental analogy to the clinical experience that anaemias of chlorotic type are sometimes met with which are refractory to treatment with iron alone [Stockman, 1895]. If one may reason from the analogy of the rats a possible explanation is that administration of iron is without avail where the diet is otherwise inadequate, particularly in the supply of vitamin A. It has been shown above that administration of inorganic iron, though efficacious, is in no way superior to feeding natural foods containing iron, and the latter have the great advantage of also containing vitamin A, which would appear to play a part in blood regeneration. The use of suitable natural foods is thus preferable to the administration of iron in the treatment of anaemia.

SUMMARY.

1. The anaemia discussed is a deficiency disease and is due to lack of iron in organic or inorganic form.
2. Administration of iron is without avail for its cure where the diet is otherwise inadequate, particularly in respect of the only natural fat which has been tested—namely that of milk. Vitamin-free fat (palm kernel oil) does not supply the deficiency.

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