

duction by using about 0.004% NaHCO_3 in the Ringer; with ordinary concentrations (about 0.015%) it is about 0.40 and readily seen with phenol red or bromthymol blue as indicator. The temporary increase in coronary flow may be as great as 100%, or more, and may be very constantly obtained by using an arbitrary 6 minute interruption of perfusion. The perfusate is best collected at one minute intervals, and the pH determined at once. Brown's³ method is convenient.

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Iron Deficiency in the White Rat and the White Mouse.

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This work is a development of the view that pellagra may be an iron-deficiency disease.¹ We have studied the physiological effects of iron restriction in white rats and white mice. The diets low in iron have been of 2 types: (a) A synthetic diet of casein, sucrose, butter fat, lard, a salt mixture which is iron-poor but assumed to be otherwise balanced,² vitamin B concentrate (Harris) and viosterol. (b) A diet of natural foodstuffs such as banana, orange juice, sucrose, freshly separated egg white, cream, and a salt mixture very low in iron but assumed to be balanced for the other elements, viosterol, vitamin B concentrate (Harris) and agar. These diets furnish protein of good quality in adequate amount, sufficient fat and carbohydrate and an abundance of vitamins A, B, D and G. All of our rats grew well.

Six series of from 6 to 8 rats each, and one series of 10 mice, raised in the laboratory and appearing vigorous and healthy at weaning age—about 21 days—were used. The young rats weighed at least 35 gm.

The results obtained have been entirely uniform, all rats and mice kept on these iron-poor diets developed the following symptoms:

If the young rats have not had access to rich sources of iron between the time that they open their eyes and the time of starting the experiment, the first symptoms of falling hair may appear as

³ Brown, J. Howard, *J. Lab. and Clin. Med.*, 1924, **9**, 239.

¹ Bliss, S., *Science*, 1930, **72**, 577.

² Cowgill, G. R., *J. Biol. Chem.*, 1923, **56**, 725.

early as the 15th day of the experiment, or as late as 2 months.

The loss of hair is always strikingly bi-laterally symmetrical. The hair is not chewed off by the other rats, necessarily, because (a) when rats are kept alone the hair loss occurs in regions that are inaccessible to the rat itself, and (b) the intact hair with the bulbous enlargement, where it had been attached in the follicle, is found on the screen below the floor of the cage. Coprophagy is an important consideration in experiments of this type, and our attempts to prevent it have met with varying degrees of success.

When the daily collection of feces and cleaning of the cage reveals a layer of hair on the screen below, one observes symmetrical areas of thinning of the coat. This often begins just above the shoulders but may also occur as a streak in the exact middle of the back—or it may take the form of longitudinal areas along both sides of the rat, leaving the area over the middle of the back entirely unaffected. When the hair is lost along both sides of the rat these areas frequently extend out upon the face. Occasionally a baldness of the head develops and may spread down over the face.

The condition in rats (unlike that in another species to be described) is usually non-fatal. The denudation may become very extensive, and a dermatitis develops in the denuded areas. This involvement of the skin itself assumes a buff-colored appearance and goes through a process of scaling off. Scaling occurs at the corners of the mouth as well.

Analyses of food and feces of rats on a low-iron diet show that they are in a negative balance. The addition of ferric citrate to the drinking water or the addition of solid ferric citrate to the food has no demonstrable effect upon the symptoms over a period of months. When excess ferric citrate is eaten, diarrhea and death supervene without any alteration or alleviation of the skin or hair condition. Prompt growth of hair and return of well-being in the rat is accomplished by feeding either cooked or dried liver, dried whole dog blood or crystalline hemin prepared from dog blood or Armour's dried hemoglobin. These substances are fed in such quantities as to furnish equivalent amounts of iron, the amount given being calculated from the state of the negative balance. The kind of iron given is obviously of much importance.

This relationship between diets deficient in iron and the loss of hair is particularly interesting in connection with analyses reported for beef hair in which it was shown that gram for gram, hair contains more iron than does any other tissue, including liver.³

³ Elvehjem, C. A., and Peterson, W. H., *J. Biol. Chem.*, 1927, **74**, 433.

In the series of white mice we have tested prophylaxis with crystalline hemin. Ten white mice from 3 litters were divided into 2 groups of 5 each—weighing 42 gm. per group at weaning age—21 days. Both groups were fed diet B. One group received a solution of crystalline hemin as an addition to the drinking water. Within 19 days all 5 mice on the iron-poor diet had developed symptoms as described above for rats, while the group which was protected by the addition of hemin was absolutely untouched by any of the symptoms at the time of writing (30th day).

Since our rats receive an abundance of the complete vitamin B group of substances, yet show much of the symptomatology of so-called G-avitaminosis or "rat-pellagra", the question arises as to whether or not some of the confusion in the vitamin G issue may not be due to the inclusion of multiple variations of which the one here described may be more or less important.

We are pursuing these studies in detail because of the appreciation that so many of the chemical and physiological properties heretofore attributed to "Vitamin G" fit in excellently with the hypothesis that the substance itself is iron. We are actively working on this problem in our laboratory from a number of angles including work with dogs, monkeys and guinea pigs.