Relationships of Hormones *to the Utilization of* Essential Nutrients *in* Erythropoiesis

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CARTWRIGHT[1] divided the known factors concerned in erythropoiesis into vitamins, amino acids, and minerals. In his survey he reported that the following vitamins played a role in this process in at least one animal species, although not necessarily in the human being: riboflavin, nicotinic acid, pyridoxine, *Lactobacillus casei* group, extrinsic factor (vitamin B_{12}), ascorbic acid, pantothenic acid, and biotin. The globin fraction of the hemoglobin molecule is known to contain many of the so-called essential amino acids and many of those called nonessential. Of the amino acids, tryptophane, lysine, phenylalanine, isoleucine, and glycine have received greatest attention and anemias have been described as resulting from deficiencies in these substances. Three minerals —iron, copper, and cobalt—have been shown to be essential for normal erythropoiesis.

Except for the well-known role the endocrines play in general metabolism, a discussion of which is not the purpose of this paper, there is very little known about the specific role of the hormones in the metabolism of these essential nutrients in erythropoiesis. On the other hand, many of these substances have been used in a therapeutic manner in an attempt to prevent or alleviate anemias which have been induced by the removal of the endocrine glands. This paper, therefore, will discuss the problem from this point of view.

The anemia induced by the removal of the hypophysis has received greatest attention. This anemia, in rats, is characterized by a hypoplasia of the bone marrow with a decrease in erythroid elements, a 30 per cent decrease in erythrocyte count, a 33 per cent decrease in hematocrit, a 31 per cent decrease in hemoglobin, a slight decrease in mean corpuscular volume, a slight decrease in mean corpuscular hemoglobin, and a normal mean corpuscular hemoglobin concentration.[2] If the hemoconcentration which occurs after hypophysectomy is taken into account, the anemia is actually more severe.[3] This anemia has been found in every experimental animal studied, and human beings with panhypopituitarism exhibit a similar blood picture.[4]

Of the known causes for such an anemia, inadequate iron intake, faulty iron metabolism, and hypothyroidism seemed the only logical ones to investigate.

Daily injections of 0.5 mg. of ferrous sulfate into hypophysectomized rats had beneficial effects, but did not prevent the anemia.[5] Daily injections of 0.025 mg. of cupric sulfate in addition to the iron induced results similar to those obtained with iron alone.[5] Further investigations showed that in spite of the fact that there was a decrease in gastric acidity[6] and a decrease in serum iron[7] after hypophysectomy, there was a normal or elevated amount of iron in storage.[7] Therefore, iron is available in the hypophysectomized animal for hemoglobin formation.

Daily injections of 0.01 mg. of thyroxine into hypophysectomized rats prevented the decrease in erythrocyte number but did not prevent the decrease in hemoglobin.[5] A combination of thyroxine, iron, and copper induced slightly better but similar results.[5]

Androgen injections have been found to induce an increased erythropoiesis in rats,[8-11] in fowl,[12-15] in hamsters,[16] and in the human.[17,18] Daily injections of 2.0 mg. of testosterone propionate into hypophysectomized rats prevented the decrease in erythrocytes

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but did not prevent the decrease in hemoglobin,[19] thus producing results simliar to those after thyroxine therapy.

Because of the difficulty in maintaining a normal hemoglobin level in the hypophysectomized rats, and because protein metabolism is not normal in such animals, it was thought that the globin factor of hemoglobin might be involved. Accordingly, hypophysectomized rats were fed a high protein diet and given 0.005 mg. of thyroxine and 1.0 mg. of testosterone propionate daily to aid in the utilization of that diet.[20] With this combination a normal hemoglobin level as well as a normal erythrocyte count was maintained.

Growth hormone, on the other hand, will not prevent posthypophysectomy anemia even in doses which are adequate to maintain growth in hypophysectomized rats.[21] Similar negative results have been reported by others.[22,23] Because of these negative results, it was thought that the good results obtained with the combined thyroxine, androgen, high protein diet therapy might have been due to the thyroxine and androgen only. Further experimentation, however, showed that although thyroxine and androgen would prevent posthypophysectomy anemia, better results were obtained if a high protein diet was added.[24] These results have led some to conclude that faulty protein metabolism is the cause for posthypophysectomy anemia.

Previous work[25] had shown that thyroidectomy induced an anemia which was not as severe as that after hypophysectomy, and that adrenalectomy induced a temporary anemia. This has been confirmed by others.[26,27] Combined thyroidectomy and adrenalectomy, however, induced an anemia which was similar to that found after hypophysectomy in Wistar rats.[28] In addition, it was found that thyroxine and cortisone therapy would prevent posthypophysectomy anemia.[29] Thus, it was concluded that posthypophysectomy anemia might be due to a combined hypothyroidism and hypoadrenocorticalism. This work has been challenged by Van Dyke *et al.*[30] because they were able to repeat the results obtained by a combined thyroidectomy and adrenalectomy, but found that the anemia after hypophysectomy was more severe. This work was performed on younger rats and of the Long-Evans strain. Work is now in progress on a comparison of the effects of hypophysectomy and of combined thyroidectomy and adrenalectomy on erythropoiesis in Wistar, Long-Evans, and Sprague Dawley rats.[31]

Before leaving this aspect of the problem, it should be mentioned that the idea of an erythropoietic hormone being released by the pituitary has been introduced. This idea was first published in 1938 by Flaks, Himmel and Zlotnik.[32] They made extracts of sheep pituitaries which were free of other pituitary factors, and were able to prevent posthypophysectomy anemia by oral administration of this extract. This work was repeated by Contopoulos *et al.*,[33] and although they could not confirm these observations with sheep extracts, they did obtain similar results with extracts of cattle pituitaries. Because of the many objections raised as to the effectiveness of orally administered pituitary hormones, the latter workers have prevented posthypophysectomy anemia with this same extract given intramuscularly.[34] Thus, we have the very interesting contradiction in the literature of data which indicate that the pituitary is responsible for posthypophysectomy anemia in a direct manner (erythropoietic hormone) and other data which implicate the thyroid and adrenal glands.

An increased erythrocyte destruction would not seem to be the cause for posthypophysectomy anemia, in spite of the fact that there is a great increase in the concentration of splenic iron after hypophysectomy. This increase seems to be due to the marked decrease in size of the spleen in these animals. The total amount of iron is normal. In addition, splenectomized-hypophysectomized rats exhibit the same anemia as rats hypophysectomized only, and there is no increase in serum bilirubin after hypophysectomy.[35]

Amazing results have been obtained with cobalt in hypophysectomized rats.[36,37] This mineral not only prevented posthypophysectomy anemia but actually increased the erythrocyte count from a normal of 8.00 mil-

lion cells per cu. mm. to almost 12 million cells; the hemoglobin from 15.8 Gm. per 100 cc. to 21.0 Gm. Because of the finding that vitamin B_{12} contained cobalt,[38–40] hypophysectomized rats were injected with this vitamin and others with liver extract.[36] Neither of these substances had any effect on posthypophysectomy anemia.

In conclusion, there is no doubt that the endocrines have a secondary influence on the essential nutrients for erythropoiesis through their influence on general metabolism. Whether they have a direct influence on these nutrients is still problematical.

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