

4548

Inorganic Elements of Spinach in Treatment of Nutritional Anemia.

HELEN S. MITCHELL AND LILA MILLER. (Introduced by H. B. Lewis.)

From the Nutrition Laboratory, Battle Creek Sanitarium, Battle Creek, Mich.

The factors necessary for the synthesis of hemoglobin in the animal body have been attracting increasing interest since the announcement by Hart and Steenbock¹ that copper seemed to play a catalytic rôle in this connection. In attempting to determine whether other inorganic factors played a similar rôle, it occurred to us to try once more the combination of minerals naturally present in a food generally conceded to be a good blood builder. A concentrated water extract of spinach was fed in quantities sufficient to supply 0.5 mg. Fe per day as supplement to the milk diet. It had previously been determined that this extract contained copper in appreciable amounts, Fe:Cu::37:1. It was to be expected that hemoglobin synthesis on spinach extract would be rapid, but to our surprise the response was almost twice as rapid and the ultimate level of hemoglobin reached and maintained was 25% higher than the response on the iron and copper combination.

This led to the question whether there might be an organic factor involved as has been so often suggested. The extract was dried and ashed and the ash fed to rats in equivalent quantities, *i. e.*, 0.5 mg. Fe per day. The ash was not completely soluble, therefore daily feedings were weighed out in the dry state and mixed with a few cc. of milk which was fed to the rats when hungry. No more milk was fed until the dishes were licked clean. In spite of this precaution to make sure that the animals received their full amount of ash, they failed to show as rapid hemoglobin synthesis as the rats on the original spinach extract. This observation made us question again the possibility of an organic factor. Previous experience² suggested that the insoluble nature of this ash might have some bearing on the problem. Consequently another group of animals were fed the same amount of ash which had been dissolved in a minimum of concentrated HCl which solution was then diluted to such an extent as to make it suitable for feeding. When the ash solution was used as supplement to the milk ration, a prompt and rapid response in hemoglobin was observed, equal in nearly every case both in rate and ultimate height to that obtained on the original extract.

¹ Hart, Steenbock, *et al.*, *J. Biol. Chem.*, 1928, lxxvii, 797.

² Mitchell and Vaughn, *J. Biol. Chem.*, 1927, lxxv, 123.

These results were further confirmed by feeding the extract and its ash in half the previous doses, *i. e.*, 0.25 mg. Fe per day under which circumstances differences in efficiency would be accentuated because the amount fed is well below the optimum requirement. The results, however, were entirely similar to the first series.

It is concluded that the inorganic combination naturally present in spinach is better than any combination of inorganic salts we have been able to find, *i. e.*, iron and copper, iron and copper manganese, etc. Furthermore, it would seem that the solubility of at least some of the factors present may have a real significance. A complete qualitative and further analysis of this spinach extract ash are the next steps in the investigation and will be carried out in the near future.

4549

Studies on Stimulation in *Nitella*.

L. R. BLINKS, E. S. HARRIS AND W. J. V. OSTERHOUT.

From the Laboratories of The Rockefeller Institute for Medical Research.

Waves of negativity occurring spontaneously in *Nitella*, sometimes in regular rhythm, have been previously described.¹ Similar negative variations, passing down the cell, may be induced in a variety of ways, by touching, bending, by application of solutions or by small direct² potentials.³ They resemble the action currents of muscle and nerve in form and magnitude but last longer and are propagated more slowly.

Nitella appears to follow in general the laws of stimulation of muscle and nerve but there are some interesting exceptions. For example, it is far from obeying the all or none law completely since the P.D. (normally positive⁴ when the cell is in contact with dilute

¹ Osterhout, W. J. V., and Harris, E. S., *PROC. SOC. EXP. BIOL. AND MED.*, 1928-29, xxvi, 124.

² The production of negative waves by induction shocks in *Nitella* was apparently first reported by Hörmann (Hörmann, G., *Studien über die Protoplasmaströmung bei den Characeen*, Jena, 1898).

³ These are ordinarily effective at cathodal closing and anodal opening but may be induced at anodal opening by higher voltages.

⁴ The protoplasm is regarded as positive when the positive current is outward, *i. e.*, when the positive current tends to flow from the inner surface through the protoplasm to the outer surface or from the outer surface through the electrometer and thence through an (imaginary) capillary passing through the protoplasm into the sap.