

to interfere with bloodstain grouping. Furthermore, the cutting of a garment may not be desirable. Extracts of the bloodstain can be made either by soaking a piece of bloodstained fabric in water or by the application of water to the uncut garment and scraping with a scalpel. Minimum quantities of water should be used (6-12 drops). Similar treatment to a control area as near as possible to the stain is essential. Saline extracts are not satisfactory due to the formation of salt crystals on drying. The procedure is as follows: (1) Prepare the slides by placing one or two drops of extract in each of the three cavities. With a glass rod, cover the whole of the cavity with extract. Control A, B and O stains may be made from 1/10 solutions of A, B and O cells in water. Extracts from control areas of fabrics will not adhere to the slide unless protein is added. For this purpose, to fabric controls, add one drop of 1/10 O serum in water to each cavity. (2) Dry in air or on a hot plate at 40-45° C. (3) Fix in McIlvaine buffer at 99° C. for 1 min. (For large-scale grouping the use of stainless steel racks is recommended). (4) Blot dry. (5) Dispense one or two drops of anti-A, anti-B and anti-H antisera to the three cavities, ensuring that all the stain is covered. (6) Place in a moist chamber and leave for 3 hr. to absorb. (A plastic refrigerator box approx. 9 in. x 4 in. is a suitable chamber. Three or four layers of filter paper should be moistened with water and forced into the bottom of the box. Invert the box, and use the lid as a slide tray.) (7) Place in refrigerator at 4° C. for ½ hr. to increase agglutination intensities. (8) Wash off excess antibody with cold saline. Place slides in stainless steel rack and wash by alternate raising and lowering in each of two beakers of cold saline, ten or fifteen times in each beaker. (9) Blot dry. (10) Dispense one or two drops of A, B and O cells 2 per cent v/v to the relevant cavities. (11) Elute at 50°-55° C. for 10 min., in a moist chamber. (12) Remove from oven, rotate, leave for 15 min., read.

All bloodstain grouping tests in this laboratory are performed in duplicate. Known A, B and O controls are run at the same time, together with a 1/10 O serum blank. Slides may be marked for purposes of identification with a lead pencil if one end of the slide is finely ground into an opaque surface. Sensitivity tests have shown that the minimum quantities of dried whole blood necessary to produce good macroscopic results are approximately 10 µgm. when testing for A or B antigens and 200 µgm. when testing for H antigen.

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PHARMACOLOGY

Hæmatological Effect of a Derivative of the Monocarboxylic Acid of Vitamin B₁₂ in Primary Polycythæmia

AN increasing number of derivatives of the monocarboxylic acid of vitamin B₁₂ is known to antagonize the growth-promoting effect of vitamin B₁₂ in various microbiological cultures (*Escherichia coli*, *Euglena*, *Ochromonas*, etc.) in the chick embryo and the rat¹⁻³.

Very few papers have, however, appeared dealing with the clinical indications of the B₁₂ antagonists. Beard⁴ reports that if administered in combination with 5,000γ of anti-vitamin B₁₂, 5γ of vitamin B₁₂ will not be followed by a reticulocyte crisis in patients with pernicious anæmia. Smith⁵ found that in patients suffering from chronic myeloid leukaemia, anti-vitamin B₁₂ failed to improve general condition or depress the high level of serum vitamin B₁₂.

The present communication describes clinical experience gained with one of the B₁₂ antagonists in primary polycythæmia. It is true that the number of patients involved is very low, and 6 months only have passed since the trials were begun, yet the results already observable are thought remarkable enough to merit preliminary communication.

The B₁₂ antagonist used was the homogeneous monocarboxylic acid of vitamin B₁₂ prepared by Kelemen⁶. It was administered to four patients in intramuscular daily doses of 1-3 mgm. Fig. 1 demonstrates the substantial reduction in the number of red cells in circulation after a total dosage of 20-30 mgm. in three cases, and of 90 mgm. in the fourth. The white cell count and the qualitative blood picture remained in all patients practically unchanged, nor was there any deterioration in the platelet count. Marrow puncture showed the erythroid-myeloid ratio to have come within the normal range in one case, and to have improved markedly in two, and moderately in one, patient. The hæmatocrit decreased and the sedimentation-rate increased. The spleen remained unchanged in size.

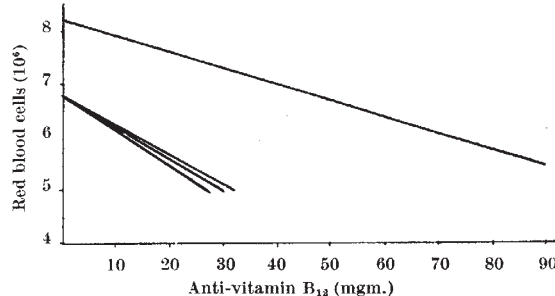


Fig. 1. Effect of anti-vitamin B₁₂ on red blood cells of polycythæmic patients

All four patients showed alleviation in subjective symptoms. In three cases this was followed by lasting remission. In one patient who had been given previous treatments with radioactive phosphorus, the slow remissive response required 93 mgm. of the drug. This was followed by a relapse, and renewal of the treatment proved of no avail.

Extended investigations are in progress to see if the reduction in the red blood cell count following anti-vitamin B₁₂ in polycythæmia vera is statistically significant, and to understand the mechanism involved.

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