Case Reports

DI GUGLIELMO'S SYNDROME WITH LOW SERUM VITAMIN B₁₂

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DI GUGLIELMO'S syndrome is a rare myeloproliferative disorder characterized by malignant erythroid hyperplasia with megaloblastoid features. Di Guglielmo, who first described it, regarded it as a pure disorder of the erythropoietic tissue. Further work, notably by Dameshek and his colleagues, has shown this concept to be too narrow (Baldini and Dameshek, 1958; Baldini, Fudenberg, Dameshek and Fukutake, 1959; Dameshek, 1958; Dameshek and Gunz, 1958). The syndrome is now regarded as one that passes through three phases. In the first phase there is excessive erythroid proliferation of the marrow, the erythraemic myelosis of Di Guglielmo. The second phase is one of erythroleukæmia involving both red and white cell series, which finally passes into the third phase, an acute myeloblastic leukæmia. The disease may present at any stage, the second being the most frequent, and may run a total course of a few weeks to several years, being fatal at any stage. Anerythraemic forms may occur in which few or no nucleated red cells are found in the peripheral blood, and in these cases diagnosis depends on the appearances of the marrow, which also differentiate leukæmia presenting with some nucleated red cells in the blood. In this paper another case of Di Guglielmo's syndrome is presented, with features of unusual interest.

Case Report

History. A man of 66 was admitted to the Victoria Infirmary on November 1, 1960, with a history of progressive loss of energy, dyspnoea, angina of effort and ankle oedema of one month's duration. He had been diabetic for seven years, well controlled on diet alone. Three months before admission he was passed as fully fit at medical examination before going to Canada, where he was in excellent health for two months until the onset of the above symptoms. One week after these started there was recurrence of glycosuria and a general practitioner prescribed tolbutamide and vitamin B complex tablets, with apparent improvement of his diabetic control. However, his general condition deteriorated so rapidly that within another week he was too weak to dress and undress himself. There was no history of exposure to ionizing radiation or to chemicals, and no family history of blood dyscrasia. There had been no obvious blood loss or purpura.

Examination. On admission the patient was extremely pale. There was ankle and sacral oedema and crepitations were detected at both lung bases. The liver, spleen and lymph glands were not enlarged. Purpura was not present but the capillary fragility test was positive. Retinal examination showed bilateral haemorrhages and exudates. The other systems were normal.

Investigations. Hb. 4.5 g./100 ml., r.b.c. 1.83 mill./cu. mm., PCV 18%, MCV 100 cu.µ, MCHC 25%. The initial 'white cell count' was 103,400/cu.mm., but examination of a stained film showed no mature white cells. The nucleated cells were in fact almost all red cell precursors. Differential count: Undifferentiated cells 8% (including a few myeloblasts and others which could not be identified), nucleated red cells 92% (basophilic 9%, polychromatic 41%, orthochromatic 42%). Many of these were megaloblastic. The red cells were hypochromic with moderate anisocytosis, slight poikilocytosis, and marked polychromasia. Reticulocyte count 3.2%, platelet count 59,000/cu. mm.

Sternal puncture revealed an intensely cellular marrow, containing many very primitive stem cells. There was extreme erythroid hyperplasia, with many pro-erythroblasts and early basophilic forms. At the later stages both normoblasts and megaloblasts were present. Many cells in mitosis could be seen, including some large polyploid forms. In the granular series there were a few myeloblasts and promyelocytes but no more mature forms, and no Auer rods were present. No megakaryocytes and no myeloma or other tumour cells could be seen. The periodic acid Schiff (PAS) reaction (Hayhoe, Quaglino and Flemans, 1960) was strongly positive in the erythroid precursors. Paraffin sections of the marrow confirmed the hypercellularity and the absence of megakaryocytes and tumour cells. Perls' reaction showed free iron to be reduced.

Direct and indirect Coomb's tests, Donath-Landsteiner, and Ham's acid serum tests were negative. Paper electrophoresis and alkali denaturation of hæmoglobin gave normal results. The blood group was A, Rh. positive. Bleeding time by Duke's method was 7 min., and clotting time by Lee and White's method 7 min. Hess's capillary fragility test was strongly positive. Prothrombin time by Quick's one-stage method was 16 sec., control being 14 sec. Serum vitamin B₁₂ level was 20 µg./ml. on two occasions. An augmented histamine test meal (Kay, 1953) showed hyperchlorhydria. The direct serum bilirubin reaction was negative, but the indirect serum bilirubin was 2.4 mg./100 ml. Other liver function tests and plasma protein levels were normal. Urobilinogen, urobilin,

and bilirubin were absent from the urine. Faecal urobilinogen level was 43.2 mg./100 g. wet faeces, and repeated examinations for occult blood were negative. Microscopic examination of the faeces showed no abnormality. Serum electrolytes normal, blood urea 49 mg./100 ml., ESR 52 mm./hr. (Westergren). Chest X-ray was normal. ECG showed left ventricular strain.

Clinical Course and Treatment. Tolbutamide was discontinued, the diabetes was well controlled on soluble insulin for ten days, and thereafter with 28 units lente insulin per day. Transfusions of packed cells and whole blood were given. Crystalline penicillin, 1 million units twice daily, was administered for nine days. This was followed by oral penicillin until 9.12.60, when tetracycline, 1 g./day, was started. A good diuretic response was obtained to mersalyl but improvement was transient. In view of the low serum vitamin B₁₂ level he was given 1,000 µg. cyanocobalamin daily for ten days and on 28.11.60, three weeks after admission, the serum B₁₂ level was over 1,000 µg./ml. Reticulocyte counts repeated during the course of the illness never rose to more than 4%. The anaemia progressed and on 7.11.60, after transfusion of 1½ pints of packed cells, total nucleated cell count had fallen to 8,400/cu. mm. in the peripheral blood, of which 1,386 were lymphocytes. The remaining cells were nucleated red and blast cells. This picture remained fundamentally unchanged until the patient died. Epistaxis and further retinal haemorrhages occurred and petechiae became evident. Prednisolone, 30 mg./day, was started four weeks after the patient's admission but did not effect any improvement. Pyrexia continued and blood culture revealed a *B. coli* septicaemia. The patient died on 9.12.60.

Permission for autopsy was refused but further sternal marrow tissue obtained post mortem confirmed the findings from two marrow aspirations made during life.

Discussion

Dameshek and his colleagues have proposed certain criteria in the diagnosis of this disease. The marrow should show erythroid hyperplasia with maturation arrest at a primitive level, megaloblastoid features, increased mitoses with frequent abnormal figures, degenerative features, and a varying degree of myeloblastic proliferation. The blood picture is that of a macrocytic anaemia, with aniso-

cytosis, poikilocytosis and erythroblastæmia, normal, decreased or slightly increased white cell count and occasional to frequent myeloblasts. The disease is invariably fatal and, although supportive therapy may prolong life, no specific treatment is effective.

In addition, Quaglini and Hayhoe (1960) have pointed out the importance of the periodic acid Schiff test for glycogen and related mucopolysaccharides in the diagnosis of this disease. They found that the erythroblasts were strongly positive. A similar reaction was found in iron-deficiency anaemia and thalassaemia major, but in no other megaloblastic anaemia apart from Di Guglielmo's disease. In the case presented all the features of the blood and bone marrow were as described by Dameshek and the periodic acid Schiff reaction was strongly positive.

In the pathogenesis of this disease toxicity or sensitivity to drugs did not seem possible, since tolbutamide was the only drug which he had been given. No blood dyscrasias have been reported with this and symptoms had actually started before therapy was commenced.

It is interesting to compare this marrow picture with that found in Addisonian pernicious anaemia in which there is also erythroid hyperplasia and megaloblastosis, and without treatment this disease also has a fatal outcome. It is, of course, known that the marrow changes in pernicious anaemia are due to defective absorption of vitamin B₁₂ with consequent low serum vitamin B₁₂ levels.

In six cases of Di Guglielmo's syndrome serum vitamin B₁₂ levels have been normal or high. The levels recorded were 1,100 µg./ml., 1,180 µg./ml., 2,544 µg./ml. (Baldini and others, 1959); 1,800 µg./ml., 510 µg./ml. (Spray and Wits, 1958); 150 µg./ml. (Adams and Seaton, 1960). Spray and Wits used microbiological assay by *Lactobacillus leichmannii*, as in this case, while *Euglena gracilis* was the organism used by the others. Although the results given by the two methods are not strictly comparable, they are of the same order (Girdwood, 1960). The unusual feature in this case was the extremely low vitamin B₁₂ level, which was repeated with identical result. The possibility of inhibition of the microbiological assay by penicillin was considered, since the patient had received one million units twice daily for three days before the first estimation was made. Bruce (1961) has shown that penicillin may affect the estimation, but only appreciably when serum B₁₂ levels are normal or high, when active growth of the organism is possible and penicillin can exert its bactericidal action. In the method used to estimate the serum B₁₂ the penicillin was diluted beyond its effective concentration. The very low levels found in this case could not be accounted for by the antibiotic.

Di Guglielmo's disease is a megaloblastic anaemia, which is known not to respond to vitamin B₁₂. The high serum vitamin B₁₂ levels found in other cases have led to the suggestion that there is a defect of utilization at the cellular level (Dameshek, 1958). This case throws some doubt on this concept, since the problem posed here was that of a

FIG. 1.—Showing megaloblasts and normoblasts in peripheral blood. $\times 860$.

megaloblastic anaemia, with low serum B₁₂, unresponsive to parenteral cyanocobalamin therapy. Although the patient's serum vitamin B₁₂ level was raised from 20 to 1,000 µg./ml. by therapy, there was no clinical or haematological response. Further study of vitamin B₁₂ metabolism in this disease is clearly required.

Summary

A case of Di Guglielmo's syndrome has been described, in which megaloblastic anaemia was associated with low serum vitamin B₁₂ level. The case is compared with others previously described. The need for further study of vitamin B₁₂ metabolism is emphasized.

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Addendum

Since this paper was prepared work has been published (Boczarow, 1961) suggesting that penicillin will invalidate B₁₂ assay by the *L. leichmannii* method. Previous work by Dr. L. G. Bruce, in the clinical laboratories of the Victoria Infirmary, did not suggest this and estimations of serum vitamin B₁₂ levels in our patient were done by his methods, which differ slightly from those of Boczarow. The validity of the results was confirmed by further studies, which will shortly be published.

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LIFE FROM A COUVELAIRE UTERUS

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As a result of the use of human fibrinogen in the treatment of the blood coagulation defect associated with abruptio placentæ, for which the work of Weiner and Schneider was mainly responsible, it is now widely appreciated that, until the correction of this coagulation defect, active interference of any sort is fraught with danger. On the subject of the method of delivery, however, there appears to be a great divergence of opinion.

The foetal mortality rate in the more severe degrees of abruptio placentæ is reported by almost

all authors to be 100%. In the presence of a Couvelaire uterus a living infant is even more rare.

Case 1

A gravida three, aged 29, was admitted to St. Paul's Hospital, Hemel Hempstead, as an emergency case of ante-partum hæmorrhage, on July 3, 1961. On admission a history of a sudden painless blood loss of approximately 10 oz. was obtained. The pregnancy had been uneventful. The estimated date of delivery was June 9, 1961.

The previous pregnancies had been 43 weeks and