Autoimmune haemolytic anaemia complicating methyldopa therapy

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Summary

Ten out of fifty-seven patients on methyldopa therapy were found to have a positive direct Coombs' test, the antibody being in all cases of the IgG type. Four of these had an autoimmune haemolytic anaemia and in spite of associated diseases all four showed rapid haematological improvement when the drug was stopped.

The incidence of autoimmune haemolytic anaemia due to methyldopa may be higher than has previously been suggested, but since the haemolytic process is reversible it seems reasonable at present, in view of its effectiveness as a hypotensive drug, to continue its use under adequate haematological control.

Introduction

Patients receiving methyldopa therapy for hypertension have recently been observed to develop a positive direct anti-human-globulin (Coombs') test which in a number of cases has been associated with an autoimmune haemolytic anaemia (Cahal, 1966; Worlledge, Carstairs & Dacie, 1966; Wurzel & Silverman, 1966; Cantor & Barnett, 1967; Hope & Provan, 1967). In the present investigation a direct Coombs' test was carried out on the red cells of fifty-seven consecutive hypertensive patients receiving methyldopa from one out-patient clinic. Those who gave a positive result were subjected to further haematological investigations.

Methods

Two anti-human-globulin reagents supplied by the National Blood Transfusion Service and by Ortho Laboratories were employed in the direct Coombs' test. Cells giving a positive reaction were further investigated using purified anti-IgG

and anti-IgM (Hyland Laboratories) antisera and also by means of a γ -globulin neutralization test.

Red-cell survival was measured in one case by means of ^{51}Cr -labelled red cells.

Results

The fifty-seven patients had been treated with methyldopa for periods ranging from 1 month to 6 years. The doses varied from 0.78 to 3.0 g daily. Ten cases were found to have a positive direct Coombs' test. The test was positive in each case with both anti-human-globulin reagents employed and the titres varied from 1/32 to 1/3000. Prozoneing was observed in three of the ten cases. The test was negative in all cases after the standard anti-human-globulin reagents had been absorbed with human γ -globulin. When specific anti-IgG and anti-IgM were used as anti-globulin reagents, strongly positive reactions were observed with anti-IgG but the test was negative, or only weakly positive, with anti-IgM.

Of the ten patients with a positive direct Coombs' test, three were found to be anaemic with haemoglobin levels ranging from 37% (5.4 g) to 59% (8.7 g). During the survey a hypertensive patient (case 4) was admitted for restabilization from the same clinic, when it was realized that she had been admitted 18 months previously suffering from a haemolytic anaemia before its association with methyldopa was appreciated.

Case reports

Case 1

A man aged 61 had suffered from essential hypertension for 5 years, with left ventricular hypertrophy and several episodes of paroxysmal nocturnal dyspnoea. Throughout this time

he was treated with methyl dopa 3 g daily. In March 1962, before treatment, he had a haemoglobin level of 100% (14.8 g). In October 1966 he was admitted in cardiac failure.

Investigations. Hb 59% (8.7 g), RBC 3,480,000/mm³, PCV 29%, MCV 83 μ^3 , MCH concentration 29%, reticulocytes 7%, WBC 3200/mm³, and a normal differential count. Slight iron deficiency was noted in the blood film. Tests for occult blood were negative on several occasions and the blood urea was 50 mg/100 ml. The urinary urobilinogen was increased, the Schumm's test was negative and both the osmotic fragility and ⁵¹Cr red-cell survival time were normal. The direct Coombs' test was positive to a titre of 1/32. A γ -globulin neutralization test showed inhibition to a titre of 1/4000. Aspiration sternal marrow biopsy showed active normoblastic erythropoiesis. Guanethidine was substituted for methyl dopa but no specific haematinics were given and over a period of 3 months the haemoglobin level rose to 88% (13.0 g). The direct Coombs' test remained positive throughout this time.

In January 1967 he complained of colicky abdominal pain without signs of intestinal obstruction. A barium enema showed a carcinoma of the transverse colon. He died in April 1967 from cardiac failure. His last blood count (April 1967) showed: Hb of 77% (11.2 g), WBC 4900/mm³, with polymorphs 72%, lymphocytes 23%, monocytes 4% and eosinophils 1%, ESR 8 mm/hr (Westergren).

Necropsy (Dr F. K. Storrington): left ventricular hypertrophy with extensive atherosclerosis and a recent myocardial infarction. There were bilateral pleural effusions with pulmonary congestion and oedema. Bone marrow: active erythropoiesis as far as the distal third of the right femur. A malignant ulcer approximately 3 cm in diameter was found in the transverse colon and histological examination showed it to be an adenocarcinoma. There were no metastases.

Case 2

A female aged 69 had been receiving 0.75 g of methyl dopa daily for 1 year for her hypertension. She presented in November 1966 with an 8-week history of abdominal pain, obstructive jaundice and anaemia.

Investigations: Hb 56% (8.3 g), RBC 2,300,000/mm³, CI 1.2, PCV 24%, MCHC 34.5%, MCV 104 μ^3 , reticulocytes 11.4%, platelets 253,000/mm³, WBC 4200/mm³, and a normal differential count. Red-cell morphology showed anisocytosis and microspherocytosis. Red-cell fragility was increased; haemolysis commenced at 0.8%

and was complete at 0.5%. There was an increased excretion of urinary urobilinogen. Aspiration sternal marrow biopsy showed an extremely cellular marrow with normoblastic erythropoiesis. The direct Coombs' test was strongly positive against dilutions of anti-human-globulin from 1/2 to 1/3000. Difficulty was experienced in determining her Rh type since her cells were agglutinated by neutral AB serum using albumin techniques. Tests in saline revealed her to be C, D negative, E positive, phenotype ccdE. In the presence of a positive direct Coombs' test, tests for the D^u antigen were impossible to perform. An eluate prepared from the red cells reacted against all cells of common Rh types (including cDE/cDE) both in albumin and in papain but there was a suggestion of anti-e specificity using the anti-human-globulin technique. Her serum reacted against all cells in albumin and in papain, but showed no abnormality using saline and anti-human-globulin techniques. Methyl dopa was withdrawn and blood transfusions were given. In spite of the apparent specificity of the eluate to e antigen Rh negative cde/cde blood was given without difficulty. Haematinics were not administered.

At laparotomy (Mr G. S. Heard) to relieve her obstructive jaundice, she was found to have a slightly enlarged liver and the spleen was approximately three times the normal size. The gall bladder and common bile duct were dilated. A stone was present in the lower end of the common duct. Cholecystectomy and choledocholithotomy were performed, but the spleen was not removed. Although an immediate post-operative cholangiogram was normal, the jaundice took 3 weeks to subside. This may have been partly due to a cholangitis which was demonstrated by hepatic biopsy. Post-operatively her blood urea rose to 200 mg/100 ml and the creatinine clearance fell to 22 ml/min. Three months later (see Fig. 1) her haemoglobin was 91% (13.3 g) and her blood urea, reticulocytes and bilirubin had returned to within normal limits. The direct Coombs' test was still positive to a titre of 1/256. After 9 months the Coombs' test had become negative. The haemoglobin was 105% (15.3 g) with 0.6% reticulocytes and a normal white cell and platelet count.

Case 3

A male, aged 64, in whom hypertension was discovered 6 years earlier, presented with blurring of vision in the left eye and a transient weakness in the left arm and leg. A partial gastrectomy had been performed 14 years previously for a duodenal ulcer.

FIG. 1. Case 2 showing principal laboratory findings.

Investigations (April 1961): Hb 94% (13.9 g), WBC 5400/mm³, platelets 224,000/mm³, and normal chest X-ray, intravenous pyelogram and renal arteriogram. Excretion of urinary 17-ketosteroids was within normal limits and the urinary catecholamines were not increased.

Treatment with guanethidine was abandoned in August 1963, because of troublesome bowel symptoms, and replaced by methyldopa 2.5 g daily, and increased to 3.0 g daily in October 1964. At this time his blood pressure was well controlled. In March 1967 a direct Coombs' test was positive to a titre of 1/512, with haemoglobin 75% (11.1 g), and reticulocytes 2%. Methyldopa therapy was continued at the same dose, but in October 1967 his haemoglobin had fallen to 57% (8.4 g). Methyldopa was discontinued. A fortnight later, a full blood count showed: Hb 68% (10.1 g), RBC 3,490,000/mm³, WBC 5700/mm³, neutrophils 61%, lymphocytes 35%, monocytes 4%, MCHC 31%, MCV 92 μ^3 reticulocytes 3.2% and platelets 254,000/mm³. ESR 32 mm/hr (Westergren). Tests for serum bilirubin, methaemalbumin and haptoglobulins were all normal. Urobilinogen excretion was slightly increased, but the red-cell fragility was normal. The direct Coombs' test was positive to dilutions of anti-human-globulin (supplied by the National Blood Transfusion Service), from 1/4

to 1/1024; and using human anti-IgG to 1/256. His blood group was determined as O Rh negative. The eluate from the red cells did not react by an indirect Coombs' technique and an indirect Coombs' test on the serum was negative.

Other investigations: blood urea 57 mg/100 ml, creatinine clearance 78 ml/min, serum vitamin B₁₂ 185 pg/100 ml. Measurement of immunoglobulins in serum by gel diffusion was IgM 118 mg/100 ml, IgA 170 mg/100 ml and IgG 2.5 g/100 ml. Aspiration marrow biopsy showed a highly cellular marrow, in which the myeloid: erythroid ratio was slightly diminished. Erythropoiesis was entirely normoblastic in type and no abnormal cells were seen. Megakaryocytes were somewhat increased in number and platelets were abundant. The appearance was that of an active marrow with normoblastic hyperplasia. A faecal fat excretion of 4.3 g was measured in a 3-day collection and occult blood tests were negative on six occasions. A barium meal showed no evidence of peptic ulceration or hiatus hernia. The partial gastrectomy was visualized as an end-to-side anastomosis of the stomach to the jejunum. A chest X-ray showed an increased heart size and an intravenous pyelogram showed poor renal function, but no structural abnormalities. An ECG showed the pattern of left ventricular hypertrophy. The haemoglobin rose slowly

without treatment to 93% (13.7 g) with 3.5% reticulocytes and MCHC of 34% during the next 8 weeks.

Case 4

A female aged 53, who had suffered from rheumatoid arthritis for 6 years, had hypertension with left ventricular hypertrophy with episodes of paroxysmal dyspnoea and proteinuria. The fundi were normal, blood urea was 40 mg/100 ml and her intravenous pyelogram was normal. She had received phenylbutazone 300 mg daily for 1 year and methyl dopa 0.75 g daily for 10 months. Hydrochlorothiazide-K and pentaerythritol tetranitrate had been taken intermittently. At the commencement of methyl dopa therapy her haemoglobin was 85% (12.7 g). Ten months later she presented with a severe anaemia and a palpable spleen.

Investigations: Hb 37% (5.5 g), RBC 1,380,000/mm³, PCV 15%, MCV 110 μ^3 , WBC 13,400/mm³; a differential count showed both immature neutrophil myelocytes and nucleated red cells, reticulocytes 26%. Platelet count was 282,000/mm³ and ESR 160 mm/hr, (Westergren). The red-cell fragility was increased; haemolysis commenced at 0.8% saline and was complete at 0.4%. Aspiration marrow biopsy showed active normoblastic erythropoiesis. Tests for occult blood in the stools were negative and LE cells were not found on four occasions. Serum bilirubin was 2.5 mg/100 ml and urinary urobilinogen was slightly increased. The direct Coombs' test was strongly positive. All her previous drug therapy was stopped. She was transfused with three pints of whole blood and given a 6-weeks' course of prednisone, starting with a daily dose of 100 mg reducing to 20 mg after 3 weeks. Within 6 weeks her haemoglobin had risen to 93% (13.6 g) and her reticulocyte count had fallen to 2% (see Fig. 2). Although the relationship between methyl dopa and an autoimmune haemolytic anaemia was not known at the time, guanethidine was substituted for methyl dopa in the treatment of her hypertension. Phenylbutazone was again used as an analgesic without further haematological changes developing. Eighteen months later she had maintained a haemoglobin level of 93% (13.6 g) with 4% reticulocytes, normal red-cell fragility and a negative direct Coombs' test.

Discussion

The incidence of a positive direct Coombs' test in the present sample of fifty-seven patients treated with methyl dopa is similar to that previously reported (Carstairs *et al.*, 1966; Cantor &

FIG. 2. Case 4 showing haemoglobin and reticulocyte response to therapy.

Barnett, 1967). A previous estimate of the incidence of autoimmune haemolytic anaemia due to the drug is about 0.02% (Worledge *et al.*, 1966; Breckenridge *et al.*, 1967). In the sixty cases reported by Hope & Provan (1967) only one had an overt haemolytic anaemia. Although the present numbers are small, four cases encountered in fifty-seven consecutive hypertensive patients treated with the drug from one clinic would suggest a higher incidence. The thrombocytopenia due to the drug, reported by ten-Pas, de Leuw & Stacey (1966) was not present in the three anaemic cases in this series in which the platelet count was done. Results obtained from direct Coombs' test and specific anti-IgG and IgM sera confirm that the antibody is of the IgG type as previously reported by Carstairs *et al.* (1966) and lo Buglio & Jandl (1967). The evidence of a haemolytic process in these four cases is based on the presence of anaemia, a raised reticulocyte count, an active marrow showing normoblastic hyperplasia, increased red-cell fragility (in two cases), a positive direct Coombs' test and an increased excretion of urobilinogen. Two of the four patients had diseases—rheumatoid arthritis and carcinoma—which are known to cause haemolytic anaemia. Case 2 had obstructive jaundice due to a gall-stone and Case 3 a previous partial gastrectomy. The aetiological relationship between haemolytic anaemia and methyl dopa is based on the improvement of the anaemia and the maintenance of a relatively high haemoglobin level with a normal or only slightly raised reticulocyte count

after discontinuing the drug. It is just possible, however, that the presence of associated diseases in the four patients may have played some part in the development of haemolytic anaemia.

Methyldopa has proved a highly successful drug in the control of hypertension even in severe cases with papilloedema (Daley & Evans, 1962). A large number of patients are receiving the drug. About 15–20% of such patients develop a positive Coombs' test but fortunately only a few of the patients will develop haemolytic anaemia and this can be corrected when the drug is withdrawn. Thus a positive direct Coombs' test is not in itself sufficient indication to withhold this valuable hypotensive drug. Frequent haematological examination including a direct Coombs' test is essential on all patients receiving this drug.

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