PAROXYSMAL NOCTURNAL HEMOGLOBINURIA

A Report of Three Cases

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Paroxysmal nocturnal hemoglobinuria (PNH) is an acquired chronic hemolytic disease characterized by continuous intravascular hemolysis and punctuated by episodes of hemoglobinuria which occur after sleep. The increased hemolysis is due to a defect in the patient's erythrocytes (an intracorpuscular defect) which render them more susceptible to lysis by normal plasma factors in vivo and by increased serum acidity in vitro. This susceptibility of PNH red cells to lysis by acidified serum is the basis of the test described by T. H. Ham (1). A positive Ham's test is considered to be diagnostic of PNH.

PNH is probably not as rare as has been supposed. In January, 1950, Crosby collected 123 cases from the world literature (2). By November, 1953, 28 more cases had appeared in print (3). The reported incidence is 1 out of every 500,000 people. In June and July, 1961, three Filipino patients were seen in the Philippine General Hospital who had chronic hemolytic anemia and a positive hemolysis test with acidified serum (Ham's Test.)

CASE REPORTS

Case 1. C. M., a 51-year old Filipino male baker, born and residing in Lipa City, Batangas, was admitted for the first time to the Philippine General Hospital on June 9, 1954 because of dizziness and headache for one year and pallor, easy fatigability, exertional dyspnea and weakness of two weeks duration. The past diseases were non-contributory except for a history of an accidental gun-shot wound in the abdomen for which he was operated on in 1944. Foreign bodies, probably

bullets, were still visible on X-ray of the abdomen in 1961. The family, personal and social history were non-contributory. Vital signs on admission were normal except for a temperature of 37.6 C. Physical examination showed a well-developed and well-nourished man with marked pallor of the skin and mucous membranes, subicteric sclera, no organ enlargement and no evidences of a hemorrhagic tendency. Hematological examination showed pancytopenia (Table 1), a macrocytic anemia and absent reticulocytosis. The first bone marrow examination showed normal cellularity and normal myeloid-erythroid ratio. A second bone marrow examination was almost completely acellular. Because of these findings a diagnosis of idiopathic hypoplastic anemia was made. Pernicious anemia was ruled out by the presence of free hydrochloric acid in the gastric juice and by the absence of megaloblasts in the bone marrow. Stool examination was negative for hookworm ova. Blood chemistry studies were normal. He was discharged improved after several blood transfusions. From 1955 to 1960 he was admitted innumerable times into other hospitals for symptoms secondary to severe anemia. He received more than 100 blood transfusions during this interval. In 1960 he was again admitted to the Philippine General Hospital several times because of weakness, easy fatigability and pallor. During these admissions, severe macrocytic anemia, leucopenia and relative lymphocytosis persisted. However, three remarkable changes had occurred. There were normal platelets, well marked reticulocytosis, and a hypercellular bone marrow with erythroid hyperplasia. In December, 1960, after receiving a blood transfusion without any signs of a reaction, he passed out almost black urine the following morning. This was interpreted as hemoglobinuria due to a hemolytic transfusion reaction (in spite of the absence of chills, fever, back pains, etc.). Anti-E agglutinins were identified in his serum. Tests for increased hemolysis were done. The patient's erythrocytes tagged with Cr-51 showed a half survival time of 16 days. Direct Coomb's test, osmotic fragility, and hemoglobin electrophoresis were normal. His urine, on two occasions when he had not received blood transfusions for at least 3 weeks, was noted to be dark brown. These were positive for hemoglobin and the sediment was loaded with hemosiderin. The complete hemolysis test with acidified serum as described by Dr. T. H.

TABLE 1
Summary of Laboratory Findings

	CASE I (C.M.)		CASE II (C.A.)		CASE III (M.D.)
	1954	1961	1957	1961	1961
BLOOD					
RBC (Millions)	1.3	1.9	1.5	0.7	1.57
Hb Gms %	6.0	4.0	4:6	2.8	5.2
Hct %		16	16	8	19.0
Size	MACRO	MACRO	NORMAL	NORMAL	NORMAL
Color			NORMAL	HYPOCH	HYPOCH
WBC	2200	3300	3000	3000	1300
Platelets	35,000	312,000	68,000	49,000	119,000
Reticulocytes %	1 — 2	8 — 22	4.3	8.4	2 — 6
BONE MARROW					
Cellularity	0 — 2 +	4 +	4+	3+	4+
M :E ratio	3 — 7 : 1	1 : 1	1 : 2	3 : 2	1 : 3
Iron				Neg.	Neg.
HEMOLYSIS TESTS					
B I mgs %	trace	1.5	1.05	0.8	1.4
B II mgs %	0	0.82—1.64	1.64	3.2	1.64
URINE					
Urobilinogen		Neg.	+	1 : 16	1:128
Hemoglobin		++++		Neg.	Neg.
Hemosiderin		++++		++	++++
Osmotic Fragility		NORMAL		NORMAL	NORMAL
Direct Coombs		Neg.		Neg.	Neg.
Indirect Coombs		+		Neg.	Neg.
RBC Survival T/2		16 Days		19 Days	11 Days
Hb Electrophoresis		Normal		Normal	Normal
Ham's Test		+		+	+

Ham was positive. This established the diagnosis of paroxysmal nocturnal hemoglobinuria.

Case 2. C. A., a 32-year old female Filipino school teacher was admitted to the Philippine General Hospital on March 31, 1957 because of blurring of vision, weakness, easy fatigability and low back pains of four weeks duration. Two years previously she had similar complaints and was told that her hemoglobin was 40%. Various hematinics were prescribed but her symptoms progressively became worse. A year later she became jaundiced. She was admitted in the Mary Johnston Hospital, Manila where 1,100 cc of blood was transfused. She was discharged symptom free and remained so for the next eleven months. Past diseases, family, personal and social history were non-contributory. On admission to the Philippine General Hospital, the vital signs were normal except for a temperature of 37.8 C. Physical examination showed a well-developed, well-nourished woman with marked pallor, subicteric sclerae, a small old submucosal hemorrhage in the gums, a hemic murmur and no organ enlargement. Hematological examination (Table 1) showed pancytopenia, slight reticulocytosis and an extremely hypercellular bone marrow with erythroid hyperplasia. The serum bilirubin was 2.69 mgs %; 1.64 mgs % was direct reacting. The liver function tests were normal. A diagnosis of chronic hemolytic anemia was made and several blood transfusions were given. She remained asymptomatic for the next four years until one month before her second admission to the Philippine General Hospital on May 21, 1961, when she began to have easy fatigability, pallor, jaundice, epistaxis and bleeding of the gums after brushing her teeth. The vital signs were normal except for a temperature of 37.7 C. The findings on physical examination were essentially similar to those on the first admission. Hematological examination showed pancytopenia, moderate reticulocytosis, hypercellular bone marrow, no iron deposits in the bone marrow, hemosiderin in the urinary sediment, rbc survival of 19 days (T/2, Cr-51 tagged patient's rbc) and a positive Ham's test. During her three months' stay in the hospital she never had hemoglobinuria.

Case 3. N. D., a 44-year old male jeepney driver, born in Palo, Leyte, but a resident of Manila for the last 26 years, was first seen in the out-patient department of the Philippine General Hospital on November, 1960, because of pallor and jaundice. Pallor and weakness were first noted eight years previously, followed two years later by spontaneous bleeding of the gums. Jaundice was noted four years before consultation. Inspite of the above complaints he was able to work. After several visits, it was established that he had chronic hemolytic anemia and was admitted to the Philippine General Hospital on July 19, 1961. The vital signs on admission were normal. Physical examination showed a well-developed and well-nourished man with marked pallor, icteric sclerae, no organ enlargement and no evidence of a hemorrhagic tendency. Hematological examination (Table 1) showed severe anemia and leucopenia and slight thrombocytopenia and reticulocytosis. The bone marrow was hypercellular due to erythroid (normoblastic) hyperplasia. Hemosiderin deposits were not seen in the bone marrow. Serum bilirubin was 3.04 mgs %, 1.64 mgs % being of the direct reacting type. The urine was negative for hemoglobin but positive for urobilinogen up to a 1:128 dilution The urine sediment showed heavy deposits of hemosiderin. The patient's erythrocytes tagged with chromium 51 had a half survival time of eleven days. Coomb's test, osmotic fragility, hemoglobin electrophoresis, and serum electrophoresis were normal. Blood smears were negative for malaria parasites. Stool examination was negative for hookworm and schistosome ova. Rectal biopsy was positive for schistosome ova. The Donath-Landsteiner test was negative. Ham's test was positive.

DISCUSSION

These three patients illustrate many of the clinical features of PNH. It occurs equally in males and females and in any age group although the peak incidence is between twenty to thirty years of age. There is no seasonal geographic or racial predilection and no familial coincidence. It is insidious in onset and is compatible with a long life. The course is accompanied by periods of remissions and exacerbations. Our second patient was symptom free during the four years be-

tween the two admissions. The anemia is accompanied by leucopenia and thrombocytopenia. It has been postulated that the leucopenia and thrombocytopenia may be manifestations of the same defect that causes the erythrocytes to be more susceptible to lysis by normal plasma factors. There is reticulocytosis; however, the bone marrow response is less than optinum. This has been attributed to intramarrow hemolysis. Nussey and Dawson (4) found that the bone marrow cells showed greater sensitivity to lysis. Bone marow aspirates, like the circulating erythrocytes, had decreased acetyl cholinesterase (5). Occasionally, instead of erythroid hyperplasia there may be a hypoactive bone marrow, as in our first patient and in several other reported cases, so that refractory or aplastic anemia is diagnosed.

Hemoglobinuria is an inconstant finding, so much so that some investigators believe that paroxysmal nocturnal hemoglobinemia might be more appropriate. Only one of our three patients had hemoglobinuria. This variability of hemoglobin excretion in the urine is due to fluctuating serum haptoglobin levels. Serum haptoglobins complex with hemoglobin up to plasma hemoglobin concentations of 125 mgs. per 100 ml. This hemoglobin-haptoglobin complex is too large to pass the glomerular filter so that hemoglobin will not appear in the urine unless its concentration exceeds the haptoglobin binding capacity. Hemosiderinuria is a more frequent finding. It was present in our three patients. Leonardi and Ruol (6) performed renal function tests and needle biopsy of the kidney in two patients with PNH. They found hemosiderin deposits in the renal tubular cells but no functional impairment. Hemosiderinuria may be so heavy as to cause iron deficiency (7).

Venous thrombosis is the most dangerous manifestation of this disease (8). Of 53 deaths in the literature, 24 were due to thrombosis usually of the brain or portal system. Our first patient complained of severe headache unrelieved by analgesics. These headaches disappeared during a short course of Dicumarol therapy which was started because of continuous 24-hour hemoglobinuria.

Numerous studies are being made to elucidate the pathogenesis of PNH. It is agreed that the basic defect is in the

erythrocyte itself. A decrease in various enzymes such as acetylcholinesterase and alkaline phosphatase have been reported. Lipid stroma abnormalities have been noted by chemical and electron microscope studies (9).

The normal serum factors supporting lysis of these defective erythrocytes have also been investigated. Certain facts are known (8). (1) Optimal hemolysis occurs at pH 6.8 to 7.2. (2) The ability of normal serum to hemolyze PNH cells is destroyed by heating to 56 C. for 30 minutes, reminiscent of many complement dependent systems. (3) Removal of any single component of complement and properdin inhibits hemolysis. (4) The PNH hemolytic system is dependent upon the presence of Mg^{++} ions. (5) The addition of thrombin to serum greatly enhances lysis of PNH cells.

The use of washed erythrocytes for transfusion and of dicumarol during periods of increased hemolysis have been recommended and generally accepted. Our three patients received only these two forms of therapy. Iron is given in the presence of iron deficiency. Steroids, splenectomy and alkalinizing salts have their proponents.

SUMMARY

Three cases of paroxysmal nocturnal hemoglobnuria seen in the Philippine General Hospital are reported. The current explorations for the many unusual clinical and laboratory manifestations of this disease are given. A brief review of studies on the pathogenesis of this disease is included.

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