IDIOPATHIC PAROXYSMAL MYOGLOBINURIA

Şinasi Özsoylu, M. D.* and Suat Akgün, M. D.**

Dark red urine is seen in cases of porphyrinuria, hemoglobinuria, hematuria, bilirubinuria, alkaptonuria, myoglobinuria and following the ingestion of certain foods [1] (containing coloring material such as rhodomine B or beets) and drugs (antipyrin and pyridum etc.). Of these only hemoglobinuria, hematuria and myoglobinuria give positive results when the benzidine or guiaic test is given. If erythrocytes are not seen in a dark red, benzidine positive urine then myoglobinuria or hemoglobinuria should be considered. For gross differentiation of these two conditions, the color of the patient's serum would be helpful. Hemoglobin before appearing in the glomular filtrate must first have saturated the available haptoglobin. Thus its concentration must be more than 100 to 150 mg per cent which could be detected by the naked eye in the serum if hemoglobinuria is present. However myoglobin will appear in the urine at serum concentrations of 15 to 20 mg per cent. In this concentration myoglobin can not discolor the serum so that it will be clear while the urine is pigmented.

Myoglobinuria may sometimes be detected following ischemic peripheral vascular disease [2] (such as arterial occlusion, a beating,[3] crushing injuries,[4] burns with destruction of the big muscle mass, [3] convulsive attacks [5] (or electro convulsive therapy), barbiturate or alcohol intoxication,[6] or severe physical exercise.[7] It may also be observed in the course of Haff's disease,[4] McArdle's syndrome [8] myositic states,[9] muscular dystrophy [10] and idiopathic paroxysmal myoglobinuria.[11 - 20]

The last condition is a rare disorder of unknown etiology, characterized by episodes of pain, weakness of the muscle, and occurrence of myoglobin in the urine. The precipitating factors in patients with this syndrome are often relatively mild exercise and or an infection.[11] Starvation and even a low carbohydrate diet may also trigger off an attack.[13, 21 ***]

From Ankara University Hacettepe Medical Faculty and Hacettepe Children's Hospital.
* Hematologist and Assistant Professor of Pediatrics.
** Associate in Internal Medicine.
*** Quoted in reference 13.

Although more and more cases have been reported in the past decade (in all approximately 80 cases), this is, to our knowledge, the first case report from Turkey.

TABLE 1

THE RESULTS OF URINARY EXAMINATION BEFORE AND AFTER ATTACKS

	First Examination		Second Examination	
	Pre	Post	Pre	Post
Color	yellow	reddish-brown	yellow	reddish-brown
Spec. grav.	1012	1022	1022	1027
Protein	neg	+++(480 mg %)	neg	+++
Sugar	neg	neg	neg	neg
Acetone	neg	neg	neg	neg
pH	5.7	6.95	5.6	6.2
Benzidine	neg	positive	neg	positive
Microscopy	few epitel rare WBC, RBC	few epitel and RBC	rare WBC	rare WBC and epitel

Case Report

G. G. (HMD 65/10664), a 21 year old Turkish male was referred to Hacettepe Medical Center by another hospital for confirmation of their tentative diagnosis, of paroxysmal myoglobinuria. In his case history, as far as he could remember, he was well until nine years of age at which time, following a 200 meter run with his playmate, he complained of severe muscle pain and tenderness, with weakness in the calves and arms. All complaints disappeared within the hour after rest, but he noticed that his urine became reddish-brown, unclear and cloudy. Since then after any strenuous exertion these complaints were repeated, becoming often more severe, and continuing sometimes for more than ten hours. During the attacks he became completely anorexic, almost paralyzed, with firmness, tenderness and swelling of the leg muscles. Although after each attack his urine was discolored, oliguria, vomiting, palor and jaundice were not observed. After learning his limitations, he decreased his physical activities markedly and, as far as he can remember, had not had an attack in the last three years. Recently he has occasionally had soreness, tenderness and tiredness of his chewing muscles at mealtimes. His parents were not related, and no comparable history was obtained either for his sister, her three children, or among other close relatives.

Physical Examination : At the time of admission he was a well developed, well nourished, robust young man. Temperature 37°C, pulse rate 90 and blood pressure 120/80. Besides relative hypertrophy of the leg muscles, physical examination was within normal limits.

TABLE 2

THE RESULTS OF BIOCHEMICAL DETERMINATIONS IN THE SERUM BEFORE AND AFTER ATTACKS

	First Examination		Second Examination	
	Pre	Post	Pre	Post
NPN	40 mg %	——	40 mg %	52 mg %
Bilirubin	7.0 mg % (D : 0.3)	0.6 mg % (0.2) (D : 0.2)	0.9 mg % (D : 0.4)	0.7 mg % (D : 0.3)
Creatin	0.33 mg %	0.22 mg %	0.1 mg %	0.2 mg %
Creatinin	0.8 mg %	1 mg %	1 mg %	1.1 mg %
Potassium	4.2 mEq/L	3.9 mEq/L	3.6 mEq/L	4 mEq/L
CO_2	26.5 mEq/L	23.5 mEq/L	——	——
Chloride	109 mEq/L	109 mEq/L	——	——
SGOT	30 u	282 u	31 u	200 u
SGPT	24 u	46 u	24 u	27 u
Phosphorus	4.2 mg %	——	——	——
Protein	6.8 gm % (Alb : 4.6)	——	7.9 gm % (Alb : 5.3)	——
Thymol	1.3 u	——	——	——
CCF	++++	——	+++	+++
Lactic acid	——	——	15.5 mg %	24.1 mg %
Cholesterol	182 mg %	——	——	——
Calcium	10.2 mg %	——	12 mg %	——

Laboratory Examination : hemoglobin 13.05 gm per 100 ml, a white blood cell count of 6000 cmm with 76 per cent neutrophils and 24 per cent lymphocytes. Platelets were adequate on the smear and a sedimentation rate (Wintrobe) of 10 mm in the first hour was observed. Direct and indirect Coombs' test and VDRL were negative.

Since the discharge of this patient from active duty depended on our report, he was given the exercise tolerance test twice, each time until an attack was noted. The results of pre and post attack urine examination, and serum chemical changes are listed in Tables 1 and 2.

His hemoglobin starch gel pH : 8.6 (Tris- citric acid) and agar gel pH : 6.45 (citrate-phosphate) electrophoresis did not reveal any abnormal component. Haptoglobin (type 1 - 2) was present before and after attack. Chest x-ray and IVP were normal. Serum electrophoresis and ECG did not show any difference after attack, nor did the serum color.

When 1 cc urine was added to 3 cc sulfosalicylic acid (3 %) reddish brown precipitation was observed only with post attack urine. Eighty per cent ammonium sulfate failed to precipitate the pigment in the post attack urine (2.8 gm ammonium sulfate was added to 5 ml urine). Starch gel electrophoresis pH : 8.6 stained with benzidine and amino-black showed the myoglobin band clearly distinguishable from that of added hemoglobin (Figure 1).

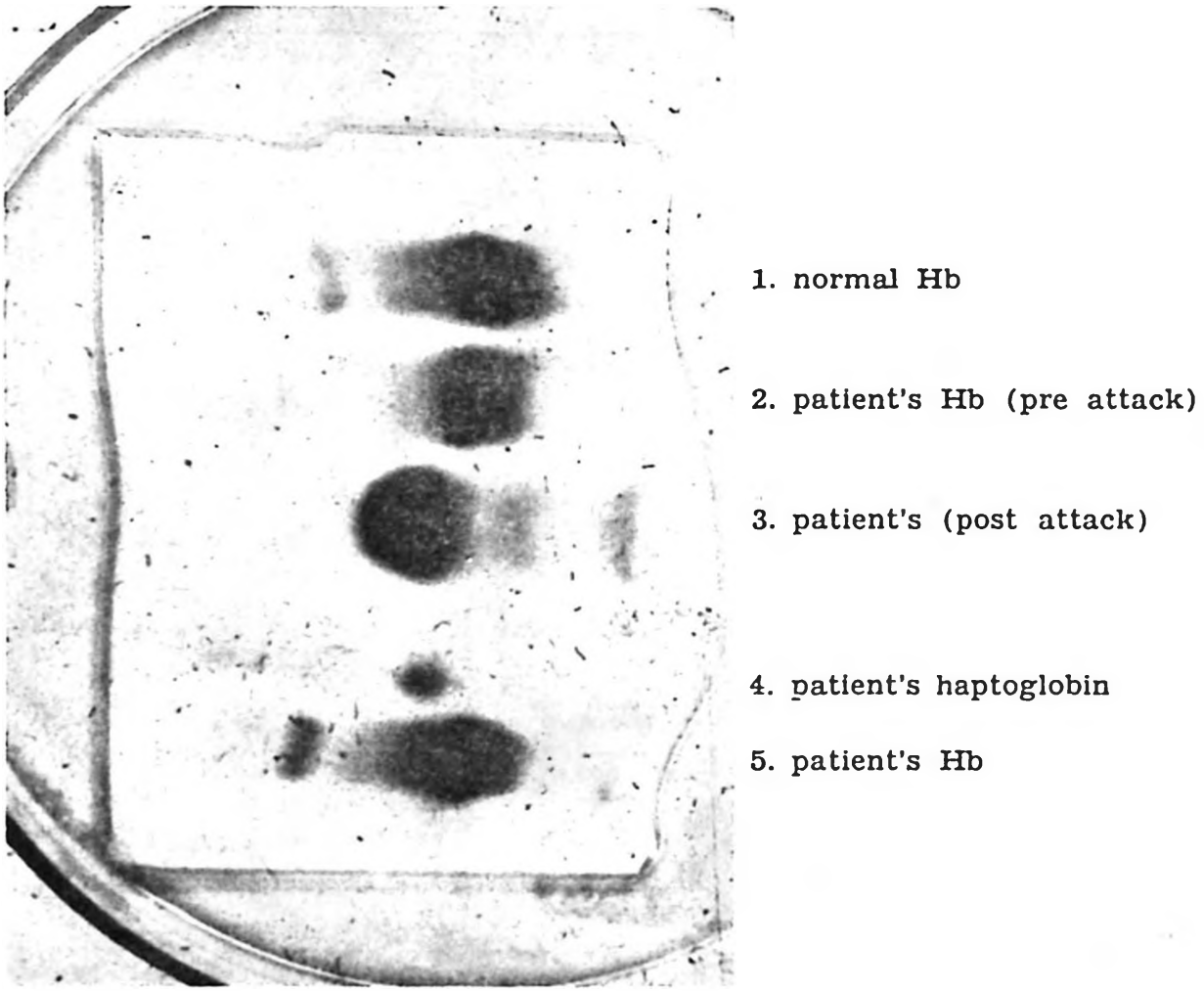

Figure 1. Berezidin stain of the starch gel electrophoresis of patient's attack urine (pH: 8.6 Tris-citric acid).

The results of urine spectroscopic examination are shown in Figure 2. As is seen, absorption curve characteristically showed oxymyoglobin, with an alpha peak at 582 and beta peak at 542 and was clearly distinguished from the hemoglobin absorption curve.

Following each attack, his leg muscles became tender, weak with increased firmness, and the patella deep tendon reflex could not be obtained and he was not able to move his legs or toes for eight to ten hours. In the post exercise period high blood pressure was noticed twice (160/120). Biopsy was obtained twice from the gastrocnemius muscle. The first biopsy

was obtained three days after the attack and microscopic examination revealed necrotic tissue. With Best-Carmen staining no glycogen was seen. Second muscle biopsy showed swelling vacuolization, extensive regeneration with little inflammatory reaction and necrosis. The muscle glycogen level,* phosphorylase (active and inactive), phosphoglucomutase activities were non-existent, and the biopsy material which was taken for biochemical determinations later proved to be necrotic tissue.

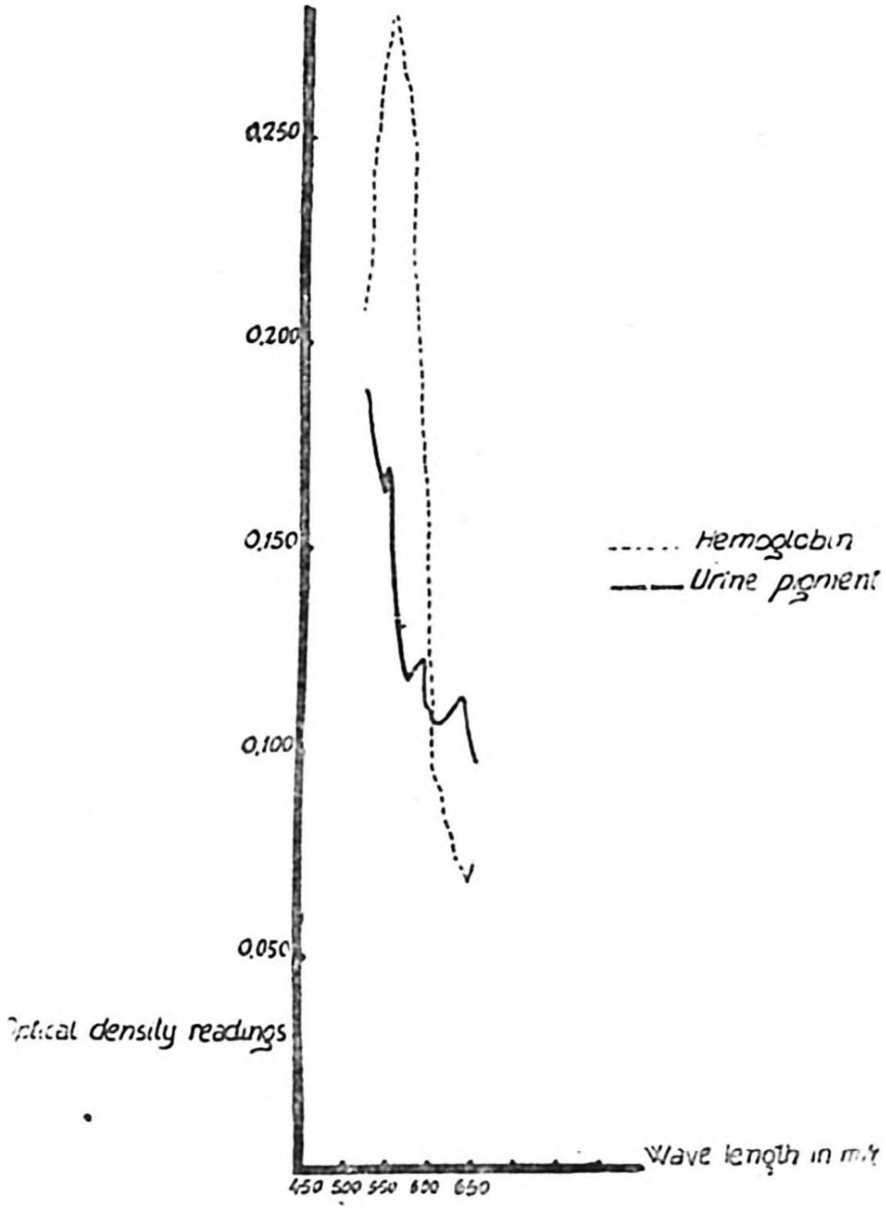

Figure 2. Spectroscopic examination of the urine pgiment is completely different from Hb absorption line. The Hb absorption band was determined after the addition of patient's Hb to the pre attack urine in which the pH adjustment was 6.9 (the pH of the post attack urine) with the addition of 1/10 N NaOH.

* Dr. Pınar Özand kindly performed these determinations.

Electromyography on gastrocnemius and solaris revealed normal voluntary action potential with low amplitude. These findings were reported as compatible with an acute myopathic process.

Discussion

This disease could easily be differentiated from secondary myoglobinuria cases. It has been reported to occur in persons of from 14 months to 57 years of age and more often in males than in females.[3, 11, 23] A history of severe exercise preceeding the attack had been reported in more than 50 per cent of the cases. The attacks of most of the remainder of the patients were found to follow acute febrile illness. In a few cases, myoglobinuria was precipitated by barbiturates, excessive alcohol ingestion or fasting. Infrequently no precipitating factor can be discovered.[20] Familial incidence of the disease has been reported,[11, 21] and Scarpelli et al [16] claimed that a sister of their patient with idiopathic myoglobinuria had elevated serum enzyme levels despite the absence of clinical evidence of the disease No other tests, that might reveal the presence of an abnormal gene in a heterozygote carrier, have been suggested. The muscles most often affected, those in the legs, back and arms, contain the highest amounts of myoglobin. In severe cases, the muscles of respiration or deglutition may also be involved. Other symptoms, besides muscular involvement and benzidine positive dark urine, may include fever, chills and anorexia.

In our case, the history and physical examination during attacks were characteristic; swelling and tenderness of the involved muscle groups, associated with hyporeflexia of the knee.

The precipitating factor in this patient was muscular exertion. Following attacks, SGOT, SGPT, NPN, lactic acid levels increased in the serum as expected.[12, 13, 18, 23] Although creatinuria [12, 15] is reported as a constant finding, possibly due to the inability of the regenerating muscle to assimilate synthesized creatine; it was not observed in our case.

Muscle biopsies, in this condition, show rather nonspecific histologic changes including necrosis and regeneration of muscle with little inflammatory reaction.[12, 15, 23]

For the indentification of myoglobin in the urine, a test described by Blondheim et al,[22] is fairly simple and reliable in most cases. The test depends on the fact that hemoglobin will precipitate at 80 per

cent ammonium sulfate concentration but that myoglobin will not. There have been at least two reports in which Blondheim's test failed to support a diagnosis of myoglobinuria which was subsequently proved by more sensitive techniques such as spectrophotometry, electrophoresis and ultracentrifugation.[17, 20] In our case using Blondheim's technique, the urinary pigment was easily proven to be myoglobin. The starch gel technique seems to be more sensitive, and with appropriate stains for heme pigments (benzidine stain), is a convenient and satisfactory method for confirming diagnoses and differentiating between the types of myoglobin present in the urine.[12, 24] Although spectrophotometry is a suitable method for the diagnosis of myoglobinuria it does not differentiate between fetal myoglobin and adult myoglobin, however the starch gel technique does.[24]

In our patient, using starch gel electrophoresis, at least three benzidine positive bands were separated. The largest component was slower than hemoglobin, the other bands faster. In the case of the patient reported by Kossmann et al, four bands were demonstrated in the urine using starch gel electrophoresis.[12] The urinary pigment of our patient showed an absorption curve characteristic of oxymyoglobin with an alpha peak at 582 and a beta peak at 542 mμ.[12, 20] Normal muscle phosphorylase activity is enough to differentiate this case from McArdle's disease.

As stated above, the patient's serum stayed colorless during or after attacks. The bindings of myoglobin by haptoglobins has also been shown by Lathem.[25] Neither hemoglobin nor myoglobin will appear in the urine until the plasma binding capacity is exceeded ding capacity of haptoglobin is less than 25 mg per 100 ml and the and unbound pigment is present in the plasma. The myoglobin binding capacity of haptoglobin is less than 25 mg per 100 ml and the pigment will not color the plasma up to the concentration of 40 mg per 100 ml. For that reason myoglobin will appear in the urine before it is visible to the naked eye in the plasma.

The etiologic factors in paroxysmal myoglobinuria have not been defined. Recently, several authors have given some data to support the hypothesis that the disease in at least some of the cases is due to an abnormality in the myoglobin molecule.[9, 12, 17]

Fetal myoglobin exists normally during the first six months of life and is replaced by the adult form shortly thereafter just as fetal hemoglobin is replaced by adult hemoglobin.[26] Perkoff found

that, in primary myoglobinuria, the F_3 fraction, similar to normal fetal myoglobin, is relatively increased.[9] Electrophoretically, urinary myoglobin, in Benoit and colleagues' case was proved to be fetal myoglobin. However it is not a general observation in this condition.

Since attacks generally followed an exercise or a fasting period in patients with primary paroxysmal myoglobinuria a defect in the muscle's energy metabolism is a target in the explanation of its etiology. Kontes and associates have shown that the low exercise tolerance of their patient could be increased by elevation of the blood glucose level either by the intravenous glucose and insulin or by injection of glucagon.[27] These authors believed that the «uncoupling of oxidative phosphorylation» could best explain their results. More recently Hinz and his associates [18] in a study of three patients with primary myoglobinuria, could not detect any specific defects which would indicate enzymatic abnormality related to the availability of glycogen or to the utilization of carbohydrate via the Embden-Meyerhof pathway. Their findings suggest that if there is a primary defect in muscle carbohydrate metabolism abnormalities may exist in the metabolic reaction of the Krebs cycle.

Since the etiology is still unknown, the treatment is only supportive during acute attacks. For long-term care, essential points are: excessive physical activity should be prohibited, and infections should be prevented as far as possible. Death has occurred principally from acute renal failure. In other instances, death has been ascribed to respiratory failure, and rarely, to hyperkalemia. During acute attacks, to decrease the myoglobin cast formation, the urinary output should be measured and an attempt should be made to alkalinize the urine.

Summary

A case of idiopathic paroxysmal myoglobinuria is reported. The first attack was observed at nine years of age following physical activity. Its diagnosis was made with Blondheim et al's method and confirmed by spectroscopic and electrophoretic studies. With starch gel electrophoresis, three benzidine positive bands were demonstrated in the urine of this patient. Haptoglobin was present in the serum, and muscle glycogen and phosphorylase activity could not be determined.

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