Int J Gynecol Obstet, 1992, 39: 29–34
International Federation of Gynecology and Obstetrics

Haptoglobin as a sensitive marker of hemolysis in HELLP-syndrome

G. Wilke[a], W. Rath[a], E. Schutz[b], V.W. Armstrong[b] and W. Kuhn[a]

[a]*Department of Obstetrics and Gynecology, and* [b]*Department of Clinical Chemistry, University of Göttingen, Göttingen (FRG)*

(Received December 7th, 1991)
(Revised and accepted April 15th, 1992)

Abstract

In a prospective study we measured laboratory variables of hemolysis in 25 patients with HELLP-syndrome. Reduced haptoglobin levels were observed in all 25 patients at diagnosis. Elevated bilirubin and plasma hemoglobin levels were observed in 5/25 patients while an abnormal peripheral blood smear was found in 11/25 patients. Our results suggest that haptoglobin is a sensitive parameter for early detection of moderate hemolysis in HELLP-syndrome and should be included in laboratory screening to aid diagnosis.

Keywords: HELLP-syndrome; Hemolysis; Haptoglobin; Early laboratory screening.

Introduction

An association between intravascular hemolysis, elevated serum transaminases, thrombocytopenia and preeclampsia/eclampsia was first reported by Pritchard et al. [10]. Weinstein [17] introduced the term HELLP-syndrome (H, hemolysis; EL, elevated liver enzymes; LP, low platelets) to describe this condition that presents as a severe life-threatening complication of pre-eclampsia. The incidence of this syndrome has been estimated at 1 case in 150–200 births [11]. As judged from a survey of the available literature, the maternal mortality is around 3.5–5% while the perinatal mortality has been estimated to be between 10 and 60% [11,15]. The prognosis for mother and child very much depends on a correct and early diagnosis of this syndrome and an urgent delivery, where necessary by cesarean section [2,11,12].

The etiology of the HELLP-syndrome is still poorly understood. It appears as a group of clinical and pathological manifestations that result from intravascular platelet activation and microvascular endothelial damage [15]. This is accompanied by a release of thromboxane A_2 and a reduction in prostacyclin formation causing vasospasm and pathologic vascular lesions within multiple organ systems [1,4,5,7,16,18]. The hepatic involvement is reflected in the typical clinical symptoms of epigastric or right upper-quadrant pain sometimes with nausea or vomiting.

The laboratory variables used to aid in the diagnosis of this syndrome include low platelets, increased levels of AST (GOT) and ALT (GPT) and laboratory findings consistent with hemolysis such as increased LDH, an abnormal peripheral blood smear, increased bilirubin or increased plasma (free) hemoglobin. However, a survey of the

0020-7292/92/$05.00

literature reveals that in a significant percentage of the patients with HELLP-syndrome, there was no positive evidence of hemolysis. One laboratory variable that has received little attention as a potential marker for hemolysis in HELLP-syndrome is haptoglobin, the transport protein for hemoglobin. The diagnostic sensitivity of haptoglobin in hemolytic disease has been reported as 83% with a specificity of 96% [6]. A reduced haptoglobin concentration has been reported in a case report on one patient with HELLP-syndrome [9] and in a preliminary communication low haptoglobin levels were noted in 20/23 patients with HELLP-syndrome [13].

The aim of this present investigation was to assess the importance of those laboratory variables that are associated with hemolysis in the diagnosis of HELLP-syndrome with special reference to haptoglobin as a possible early marker of hemolysis.

Materials and methods

In a prospective study over 2.5 years (1989–1991) plasma bilirubin, LDH, free hemoglobin and serum haptoglobin were determined in 25 pregnant women with the full clinical presentation of the HELLP-syndrome. A peripheral blood smear was also obtained. Measurements were made on admission to the hospital and, depending on the clinical course, at short intervals thereafter. The laboratory screening included platelet count, hematocrit, hemoglobin, leucocytes, hemostatic variables (PTT, thromboplastin time, fibrinogen, AT-III), LDH, AST, ALT, uric acid, creatinine, total protein and plasma electrolytes. All laboratory parameters were measured using standard methods in the central laboratory of the University of Göttingen. Plasma enzyme activities were determined at 25°C using optimized standard procedures recommended by the German Society for Clinical Chemistry. Serum haptoglobin was determined by rate nephelometry using the Beckmann Array® immunochemistry system. The relative distributions of the five LDH isoenzymes were also quantified in five patients by agarose gel electrophoresis using the Beckmann Paragon System.

Delivery was by cesarean section in 24 patients and in 1 case of a patient presenting with a ripe cervix a spontaneous delivery occurred.

Results

The mean age of the patients was 28 ± 5 years (range 21–41 years) and the mean gestational age at diagnosis was 35.4 ± 3.3 weeks (range 28–40 weeks). There were 2 cases of twins. Twenty-two patients complained of upper right quadrant pain on admission. Two patients had diabetes mellitus. The mean systolic blood pressure was 153 mmHg (range 100–220 mmHg) and the mean diastolic blood pressure was 98 mmHg (range 60–130 mmHg). Five patients were normotone. The mean time between admission and delivery was 3.3 ± 3.6 h (range 1–14.5 h). No maternal or perinatal death occurred. The average weight of the children at birth was 2124 ± 890 g (range 700–3490 g). In one infant there was a severe retardation of over 7 weeks (weight at birth: 700 g in the 35th gestational week). An Apgar score below 7 was observed in 3 children. An arterial umbilical cord pH below 7.2 was found in 3 children.

On admission to the hospital all patients had platelet levels below the lower reference limit of 150 000/mm^3. A further decrease in these levels was generally observed during the close laboratory monitoring of the patients such that all patients had levels below 100 000/mm^3 at the time of diagnosis. The liver enzymes AST and ALT were at least twofold elevated above the upper reference limits of 15 U/l and 22 U/l respectively (Table 1) and plasma LDH concentrations were also increased above the upper reference limit of 200 U/l in all patients (Table 2). It should be noted that the enzyme activities were determined at 25°C using optimized standard procedures recommended by the German Society for Clinical Chemistry. The reference inter-

Table 1. Laboratory findings at the time of admission to the hospital.

Variable	Reference interval	Mean ± SD	Median	Range	Sensitivity (%)
Platelets	> 150 000/mm^3	80 700 ± 33 000	83 000	16 000–149 000	100
AST	< 150/l	85 ± 35	86	35–148	100
ALT	< 220/l	88 ± 37	83	34–203	100

vals are therefore lower than those observed in laboratories that measured enzyme activities at 37°C.

The relative distributions of the 5 LDH isoenzymes were determined in 5 patients (Table 3). In 4 out of the 5 cases there was a relative increase in LDH5, the hepatic isoenzyme, compared with the other 4 isoenzymes. In the remaining case the distribution of the 5 LDH isoenzymes was similar to the reference values indicating a relative increase in all five isoenzymes.

Total plasma bilirubin concentrations averaged 1.0 mg/dl; values above the upper reference limit of 1.2 mg/dl were only detected in 5/25 cases while elevated plasma hemoglobin levels were found in only 5/25 cases (Table 2). Abnormal peripheral blood smears were seen in 11/25(44%) cases.

At the time of diagnosis serum haptoglobin concentrations below the lower reference limit of 0.7 mg/ml were observed in all patients (Table 2). In 3 patients the serum haptoglobin concentrations from the first blood sample taken immediately on admission to the hospital were still in the lower normal range (0.7–1.0 mg/ml). However, in all 3 cases a sharp drop in haptoglobin levels occurred during the next 10 h (Fig. 1). In three individuals in whom haptoglobin levels were measured postpartum, normal levels had been reattained within 24–48 h while a trend to normalization was observed in a further two patients.

As the disease progressed clinically, both haptoglobin levels and thrombozyte counts decreased. The decreases in haptoglobin levels were, however, much more dramatic (–92% of initial value on admission) than the drop in thrombocyte levels (–24%).

Discussion

The 3 laboratory criteria suggested by Weinstein [17,18] to be indicative of the HELLP-syndrome are low platelets (< 100 000/mm^3), elevated liver enzymes and hemolysis. The criteria for hemolysis were defined by Sibai et al. [14] as an abnormal peripheral blood smear, increased bilirubin (> 1.2 mg/l) and increased LDH. Elevated LDH concentrations appear to be an almost universal finding in HELLP-syndrome and all patients in this present investigation displayed

Table 2. Laboratory variables of hemolysis[a] at the time of diagnosis.

Variable	Reference interval	Mean ± SD	Median	Range	Sensitivity (%)
Bilirubin	< 1.2 mg/dl	1.0 ± 0.8	0.8	0.2–4.1	20
Plasma hemoglobin	< 40 mg/dl	32 ± 13	28	20–70	20
LDH	< 200 U/ml	447 ± 214	400	221–1225	100
Haptoglobin	0.7–3.2 mg/dl	0.28 ± 0.22	0.16	< 0.1–0.68	100

[a]Abnormal periphal blood smears seen in 11/25 cases.

Table 3. Distribution of LDH isoenzymes in 5 patients with HELLP-syndrome.

Variable	Reference interval	K.K.	Sc.A.	S.K.	Su.A.	R.H.
Total LDH	<200 U/l	733	491	591	1225	400
LDH 1	15–32 rel.%	23	11	22	14	17
LDH 2	33–50 rel.%	44	18	31	33	30
LDH 3	13–29 rel.%	17	9	14	15	14
LDH 4	4–9 rel.%	4	8	10	12	10
LDH 5	3–15 rel.%	13	54	23	36	29

increased levels of this cytoplasmic enzyme. However, LDH is ubiquitous to many organs including the liver and the erythrocytes. The isoenzymes LDH1 and LDH2 are the predominant forms in heart muscle and erythrocytes while LDH5 is the major isoenzyme present in the liver. In 4 out of the 5 patients in whom we performed an electrophoretic separation of the LDH isoenzymes, the elevated plasma LDH concentrations were associated with a relative increase in LDH5, the hepatic enzyme. Increased levels of LDH do not therefore necessarily indicate the presence of hemolysis in cases of suspected HELLP-syndrome.

In one of the largest retrospective studies so far Sibai et al. [14] reported their observations on 112 cases with 'true' HELLP-syndrome. All patients had an abnormal peripheral blood smear as well as elevated bilirubin and a platelet level below 100 000/mm^3. These laboratory findings were associated with poor maternal-perinatal outcome, the perinatal mortality rate being 36.7%. Weinstein reported 86% of his 56 patients with HELLP-syndrome as having an abnormal peripheral blood smear while 62% had raised bilirubin. The perinatal mortality rate was 7.9%. Rath [12] reported 100% of his 50 patients with HELLP-syndrome as having elevated LDH, while 33 of the 50 patients demonstrated

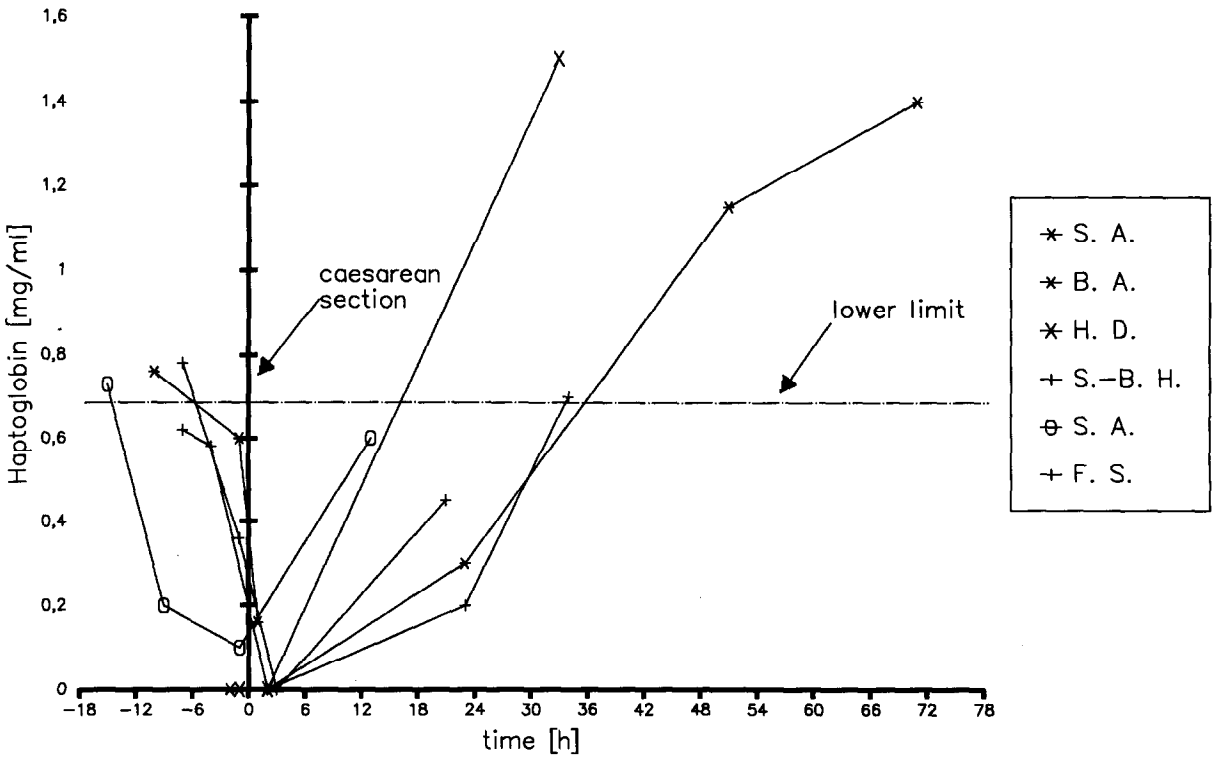

Fig. 1. Course of haptoglobin serum-levels in 6 patients with HELLP syndrome, before and after delivery.

elevated bilirubin. In the 17 patients with normal bilirubin, schistocytes were found. The perinatal mortality rate in this study was 7.8% and no maternal death occurred. Kuhn [3] reported on 71 patients with HELLP-syndrome. A very small perinatal mortality rate of 6.7% was described and no maternal death occurred presumably due to the early diagnosis and treatment of this syndrome.

A platelet count of below 100 000/mm^3 has been cited as one of the diagnostic criteria for HELLP-syndrome [15]. In this present investigation 4/25 patients had platelet levels above 100 000/mm^3 but below our lower reference limit of 150 000/mm^3 at the time of admission to our clinic. However in all cases the levels dropped during the next 4 h such that at the time of diagnosis they were below 100 000/mm^3.

Only 25% of the patients in this study had elevated bilirubin concentrations while 44% had an abnormal peripheral blood smear.

An abnormal peripheral blood smear has been suggested as a criteria for hemolysis and earlier reports [14,17] have found a higher percentage of patients with abnormal smears. However, the maternal and infant mortality was generally higher than in the present investigation indicating a more advanced stage of the disease.

In contrast to these laboratory variables of hemolysis, serum haptoglobin levels were reduced in all 25 patients at the time of diagnosis. Haptoglobin binds specifically and with high affinity to the protein (globin) in hemoglobin. The haptoglobin-hemoglobin complex is then rapidly cleared in a matter of minutes from the circulation by the mononuclear-phagocyte system while free haptoglobin has a relatively long residence time ($t_{1/2}$ = 4 days). A decrease in haptoglobin levels is thus a very sensitive method for detecting significant but moderate levels of hemolysis [8]. In the interpretation of serum haptoglobin levels consideration may have to be made of the fact that haptoglobin synthesis is reduced in hepatocellular disease and increased in the acute phase response. In the present investigation serum haptoglobin levels were either low or absent in all patients at the diagnosis of HELLP-syndrome confirming the hemolytic component of this disease even though bilirubin and plasma hemoglobin concentrations were normal in 75% of cases. Furthermore, in three cases with haptoglobin levels in the lower normal range on admission to the hospital a rapid reduction was observed within a matter of hours (Fig. 1) such that at diagnosis all values were pathological. Such a rapid decrease is not consistent with a reduction in hepatocellular synthesis but must rather be due to an ongoing hemolytic process.

In comparison to the decrease in thrombocytes, the haptoglobin values fell much more dramatically in the patients who were serially monitored. Haptoglobin would therefore appear to be a sensitive parameter for the control of the course of the disease.

Conclusions

The present results taken together with previous preliminary reports [9,13] suggest that the measurement of serum haptoglobin concentrations can serve as a sensitive and early marker for the hemolytic component of HELLP-syndrome. Serial measurements of this variable should aid in the accurate diagnosis of this syndrome before the HELLP-process becomes too far advanced and so help reduce maternal and perinatal morbidity and mortality.

References

1 Beller FK, Dame WR, Ebert C: Pregnancy induced hypertension complicated by thrombocytopenia, haemolysis and elevated liver enzymes (HELLP) syndrome. Renal biopsies and outcome. Aust NZ J Obstet Gynaecol *25:* 83, 1985.

2 Kuhn W, Graeff H, Rath W, Loss W: HELLP-syndrome. Arch Gynecol Obstet *245:* 817, 1989.

3 Kuhn W, Rath W, Loss W, Graeff H: Le syndrome Hellp — Resultats cliniques et d'analyses en laboratoire. Revue Franc Gynecol Obstet 1991, in press.

4 Killam AP, Dillard SM, Patton RC, Pedersen PR: Pregnancy induced hypertension complicated by acute liver

disease and disseminated intravascular coagulation. Am J Obstet Gynecol *123:* 823, 1975.
5 Lopez-Llera M, Espinosa M, Leon MD, Linares UR: Abnormal coagulation and fibrinolysis in eclampsia. A clinical and laboratory correlation study. Am J Obstet Gynecol *124:* 681, 1976.
6 Marchand A, Galen RS, Lonte VF: The predictive value of serum haptoglobin in haemolytic disease. J Am Med Assoc *243:* 1909, 1980.
7 McKenna J, Dover NL, Brame RG: Preeclampsia associated with hemolysis, elevated liver enzymes and low plateles: An obstetric emergency? Obstet Gynecol *62:* 751, 1983.
8 Pesce AT: Hematological disorders; laboratory approaches. In: Methods of clinical chemistry 1 (eds. AT Pesce and LA Kaplan). C V Mosby Co., St. Louis, Washington, Toronto. pp 1230, 1987.
9 Poldre PA: Haptoglobin helps diagnose the HELLP-syndrome. Am J Obstet Gynecol *157:* 1267, 1987.
10 Pritchard JA, Weismann R, Ratnoff OD, Vosburgli G: Intravascular hemolysis, thrombocytopenia and other hematologic abnormalities associated with severe toxemia of pregnancy. N Engl J Med *250:* 89, 1954.
11 Rath W, Loos W, Kuhn W, Graeff H: The importance of early laboratory work-up for obstetric procedure in case of severe gestosis and HELLP-syndrome. Geburtsh Frauenheilk *48:* 127, 1988.
12 Rath W, Kuhn W: The importance of early laboratory screening methods for maternal and fetal outcome in cases of HELLP-syndrome. Eur J Obstet Gynecol Reprod Biol *36:* 43, 1990.
13 Schröcksnadel H: Haptoglobin and free haemoglobin in pregnancy-induced hypertension. Lancet *ii:* 1594, 1990.
14 Sibai BM, Taslimi MM, El-Nazar A, Amon E, Mabie BC, Ryan GM: Maternal perinatal outcome associated with the syndrome of hemolysis, elevated liver enzymes and low plateles in severe preeclampsia — eclampsia. Am J Obstet Gynecol *155:* 501, 1986.
15 Sibai BM: The HELLP-syndrome — hemolysis, elevated liver enzymes and low plateles: much ado about nothing? Am J Obstet Gynecol *162:* 311, 1990.
16 Thiagarajah S, Bourgeois FS, Harbet GM, Caudle MR: Thrombocytopenia in preeclampsia: associated abnormalities and management principles. Am J Obstet Gynecol *150:* 1, 1984.
17 Weinstein L: Syndrome of haemolysis, elevated liver enzymes, and low platelet count: a severe consequence of hypertension in pregnancy. Am J Obstet Gynecol *142:* 159, 1982.
18 Weinstein L: Preeclampsia/eclampsia with hemolysis, elevated liver enzymes and thrombocytopenia. Obstet Gynecol *66:* 657, 1985.

Address for reprints:

G. Wilke
Department of Obstetrics and Gynecology
University Hospital
Georg-August-University
Robert-Koch-Strasse 40
3400 Göttingen, FRG