ORIGINAL REPORT

Association of Haptoglobin Phenotypes with Clinical Features of Preterm Labor Disease

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Abstract- Preterm birth means the birth before thirty seven week of pregnancy that causes a lot of complications for the baby. Variety factors are suggested to be involved in disease. In this study, we decided to evaluate haptoglobin (Hp) phenotypes association with clinical features of patients suffered from premature delivery to understand better the possible correlation of genetic and clinical features in this disease. This cross sectional analytic descriptive study has been carried out in two groups of 120 women, 60 with preterm and 60 with term labor. Patients were selected with previously diagnosed by a gynecologist with preterm birth in hospital during the study period. After performing diagnostic tests, the frequency of each haptoglobin phenotype in the two groups was analyzed using the X2 test and SPSS software. The maximum serum haptoglobin phenotype frequency in patients with Hp2-2, was 43 (71.7%) whereas in healthy individuals, 35 (58.3%). No statistically significant differences between the two groups was found (P=0.310). But based on some patients clinical features such as their history of preterm delivery, previous history of recurrent abortions and history of preterm delivery in their family, significant association was found with Hp2-2 compared with healthy control (P<0.003). This study showed that Hp2-2 phenotypes levels in the case group was higher than in control but the factors influencing the presence or absence of preterm labor is clinically various.

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Introduction

The term of preterm birth is the birth before the thirty seven week of pregnancy that can cause many complications for babies (1). Investigations have been mentioned the role of low birth weight, amniotic fluid infections, genitourinary anomalies and intrauterine growth as predisposing factors of disease (2-4).

Other factors including age, ethnicity, education level, occupation and economic status (5) living in busy and crowded urban environments, inactivity and obesity, stress and employment at risk for a difficult and hazardous jobs (6) pre eclampsia and lack of in their care prenatal behaviors leading to damage to the fetus early membranes rapture, food intake and certain drugs (7) immunological changes such as the role of CRP (8,9), the role of heat shock proteins (10), level of saliva sterols (11), cytokines and pro inflammatory agents (12-15) without cause, or idiopathic (8) can be named.

According to available reports, 11.6% of different ethnics have this type of delivery, and black women compared with whites, are more susceptible to this kind of parity and in this context do not exist in Iran. Based on WHO reports, underweight newborn infants, showing

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as an index for predicting death in the first 28 days of life has been related to the preterm delivery whereas in the developing countries, have been attributed to intrauterine growth retardation (9).

Despite the increase in neonatal care, preterm birth is still one of the major factors for baby's brain disability and other long-term disability. Considering that it can be difficult to stop preterm labor process but it is suggested that through the identification of predisposing factors and predictors of premature and underweight births, deaths and complications in mother and in fetus will be prevented. A large number of ultrasonographic parameters in predicting preterm delivery and reduced infant mortality and morbidity have been studied (16).

The various methods, including laboratory examination of amniotic fluid, cervical and vaginal secretions, urine, saliva, fluid periodontal and biochemical components of the immune serum, and risk of preterm delivery were studied so far none of them has provided good information in predicting the occurrence of preterm delivery (11,17), so we asses to evaluate haptoglobin phenotypes in patients suffering from premature delivery compared to women who have timely deliveries, with aims of if any relationship is existed, design plan for early detection and treatment is necessary.

Haptoglobin is a serum α2 cyaloglycoprotein that belongs to acute phase biomarkers and synthesis by hepatocyte and cells of the reticuloendothelial system and its level increases in inflammation (18). Its structure is similar to immunoglobulins and has two light chains (α) and two heavy chains (β) and based on molecular weight consists of three phenotypes Hp1-1, Hp2-1 and Hp2-2 (19).

This proteins has important roles, including protecting the body against toxic free radicals (20), bacteriostatic (21) and finally has the role in balance between Th1 and Th2 immune responses (22).

Several reports have shown haptoglobin phenotypes relation with various diseases. For example, TB patients who have had Hp2-2 phenotype, were likely six times more to die compared with those of their Hp1-1phenotypes Hp1-1 (23). Diabetes disease is associated with the Hp2-2phenotype (24), Hp1-1 with premature rupture of membranes (25) and pre eclampsia (26). Phenotype association with asthma has also been reported (27). Concentration of this protein in the serum of pregnant women has been studied, and results showed that 39% of patients had less than normal range (28).

There is no report about the relation of these protein phenotypes with preterm labor in Iran. We aimed to study the possible relation of this proteins as well as demographic factors associated with premature delivery to design control and treatment plan for such patients.

Materials and Methods

This cross sectional analytic descriptive study was conducted on 120 pregnant women who were divided in two groups, 60 cases of preterm and 60 controls with term labor at Ali Ebneh Abitable Hospital of Zahedan city, Zahedan-Iran in 2011. Patients with previously diagnosed by gynecologist were selected based on preterm labor criteria issued from American Children Academy (ACOU) in 1997 (1). Questionnaire forms containing epidemiological information were completed for each group. After obtaining informed consent and filled out the questionnaire, 5 ml of peripheral venous blood was taken from them.

After clotting, serums were separated and haptoglobin phenotypes of each individual were isolated by protein electrophoresis method in polyacrylamide gel and determined using specific peroxidase staining as described in our article (29). The frequency of each phenotype was reported as a percent for each group. The comparison of frequencies of each phenotype in two groups were evaluated using the X^2 statistical test and SPSS software.

Results

In this study, 120 pregnant women aged ranging between 19 to 35 years with a mean of 25.8±4.6 years old. The Job of most of them was housewives and their level education was below high school. Majority of them had a history of preterm delivery, previous history of recurrent abortions and preterm delivery in their family, that significant association was found with these and Hp2-2 compared with healthy control ($P \leq 0.003$). Other demographic factors showed no significant differences.

Maximum serum haptoglobin phenotype frequencies of case group was observed in 71.7% patients with Hp2-2. The Hp2-1 phenotype frequencies were found in 21.7% and Hp1-1 in 6.6%. In healthy control group, the maximum serum haptoglobin phenotype frequencies were seen in 58.0% with Hp2-2, whereas for Hp2-1 phenotype frequencies, 32.0% and for Hp1-1, 10.0 phenotype frequencies were obtained. Figure 1 shows determination of serum haptoglobluline phenotypes electrophoresis in polyacrylamid gel using special peroxidases dye.

Table 1. Difference in phenotype frequency of haptoglobin in patient and control groups.

Group / Phenotype	Patient		Normal	
	Number	Percentage	Number	Percentage
1-1	4	6.7	6	10.0
2-1	13	21.7	19	32.0
2-2	43	71.6	35	58.0
Total	60	100	60	100

In relation to comparison of each haptoglobin phenotype together using the X^2 test and SPSS software it was determined that the Hp2-2 phenotype in case group was higher than in control and Hp1-1 and Hp2-1 were higher in the control group than in case but statistically, there was no significant difference between the two groups. The frequency difference of each phenotype in the two groups were analyzed, and statistically, there was no significant difference between the two groups observed (P=0.310) (Table 1). Regarding to some patients clinical features such as history of preterm delivery, previous history of recurrent abortions and history of preterm delivery in their family, statistical correlation was observed with Hp2-2 phenotype compared with healthy control (P=<0.003).

Discussion

Preterm delivery is one of the disorders in women that lead them to have a premature baby birth, and this may increase their mortality rates. On the other hand, suffering from this disease may susceptible them to a variety of disabilities and costly treatments can be imposed on families and society. Therefore reducing premature mortality and disability associated with this complication, is necessary to identify risk factors for preterm delivery compared with the findings of previous studies can be informative and helpful.

Several reports have been published regarding to haptoglobin phenotypes relation with various diseases. But in preterm delivery study, there is no investigation has been done in this field in Iran and the world.

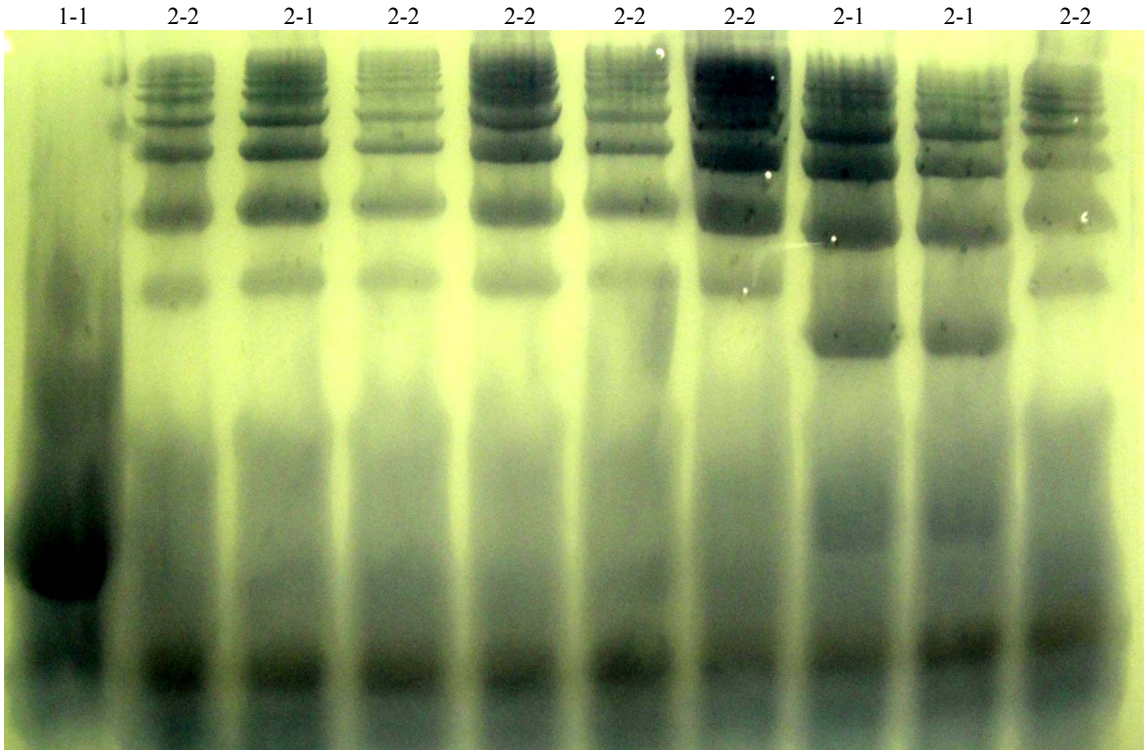

Figure 1. Determination of serum haptoglobluline phenotypes electrophoresed in polyacrylamid gel using special peroxidases dye.

So some reports indicating clearly the role of haptoglobin phenotypes in some diseases. Wobeto and his colleague in 2008 reported the polymorphism of human haptoglobin and its clinical importance in verity diseases (20). In TB patients who have had Hp2-2 phenotype, die has been likely happened six times more compared with those who had Hp1-1 phenotypes (23). Hp2-2 phenotypes have been reported by Nakhoul *et al.* in 2007 in patients suffered from diabetes type 1 and 2 (24).

Haptoglobin phenotypes associated with cardiovascular disorders, cancer, infectious diseases and neurological disorders are also investigated (20). Premature rupture of membranes associated with Hp1-1 (25) and preeclampsia with Hp1-1 (26). Serum concentration of this protein was reported lesser than out of normal in 39% of pregnant women (27).

In our study, we examined the possible role of Hp phenotype associated with clinical features of patients suffered from preterm labor compared with term labor. 120 pregnant women were studied and divided in two groups, 60 cases of preterm and 60 controls with normal delivery, The higher serum haptoglobin phenotype was found in patients with Hp2-2 associated in 43 cases (71.7% percent), whereas 35 healthy controls (58.0%), had this phenotype. The phenotype Hp2-1 in 13 cases (21.7%) and Hp1-1 in 4 cases (6.6%) were found whereas Hp2-1 phenotype in 19 cases (32.0%), and Hp1-1 in 6 cases (10.0%) in the control group were found.

Comparison of haptoglobin phenotype frequency between two groups showed that Hp2-2 levels in cases were higher than controls, whereas Hp1-1 and Hp2-1 were higher than the control group. So there was no statistically significant difference between the two groups (P=0.310) but regarding to the clinical status, this relation may exist. On the other hand based on some clinical features of patients such as their history of preterm delivery, previous history of recurrent abortions and history of preterm delivery in their family, significant association was found with Hp2-2 compared with healthy control (P=<0.003). As Hp2-2 phenotype likely to play a role in the regulation of humoral and cellular immune responses and can be stimulated antibodies syntheses after active immunization and under circumstances, prevent the proliferation of lymphocytes and also affect the balance of Th1/Th2 responses and increase Th1 responses (22), so this relation is also confirmed. Other clinical features of disease showed no significant differences.

A similar finding in studies on patients suffering with allergic rhinitis compared with healthy individuals has already been observed (29).

The lack of significant difference between the two groups studied, indicating that specific haptoglobin phenotypes have no role in causing preterm labor. So apart from haptoglobin phenotypes, other genes may have effect on the disease that needs further investigations in the future.

It can be also said that environmental factors and life styles may have an important role in preterm birth than the possible role of genetic factors. Thus, these results are consistent with the results obtained by other investigators (30,31).

On the other hand, the majority of patients had experience of one time pregnancy with low educational level and their job was housekeeping. So these can be other influence factors for their disease. These all suggesting further study in women with a history of several preterm deliveries would need to follow the footsteps for genetic factors. It would be also suggested that the amount of serum haptoglobin levels in women with preterm labor should be measured.

The results showed that the Hp2-2 phenotype in the case and Hp1-1 and Hp2-1 in the control group were higher, but specific haptoglobin phenotype influencing disease, is not create or cause preterm labor. Based on some clinical features of patients such as their history of preterm delivery, previous history of recurrent abortions and history of preterm delivery in their family Hp2-2 phenotype have a role in this disease.

Thus the study results suggest that in women with preterm labor, other genetic factors and measurement the amount of serum haptoglobin should be considered in those with a history of several preterm deliveries.

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