The Turkish Journal of Pediatrics
Volume 25, No. 1, January - March 1983

SERUM HAPTOGLOBIN LEVELS IN PATIENTS WITH ACTIVE AND INACTIVE RHEUMATIC FEVER*

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Key words: acute phase reactants, rheumatic fever, serum haptoglobins

When there is an inflammation in an organism caused by infectious agents certain changes take place which all together are called the acute phase reaction. The production of some proteins such as fibrinogen, haptoglobin, ceruloplasmin, orosomucoid and C-reactive protein, and perhaps others, is increased in the organism, especially in the liver[1].

Haptoglobin is a glycoprotein which is present in plasma and other biological fluids[2,3] and combines with free hemoglobin. Electrophoretically it moves in the alpha$_2$ fraction, and the major part of haptoglobin synthesis occurs in the liver[4-6]. The body synthesizes approximately 1 gram of haptoglobin per day[7]. It has been shown by isotope technique that a human fetus synthesizes haptoglobin in the liver as early as nine weeks of age[8].

The levels of haptoglobin may increase in inflammatory conditions or infections. This elevation is thought to be caused by hepatic production, initiated in an unknown way by abnormal conditions, but during the initial phase of inflammation, haptoglobin concentration decreases for several hours. The reason for this has not been clarified[9].

This study investigates whether plasma haptoglobin concentration, as an acute phase reactant, has any significance in the diagnosis of acute rheumatic fever. In one of our previous studies[10], we measured the serum haptoglobin values by using the peroxydase activity method in 68 patients with definite acute rheumatic fever and demonstrated an increase in the acute phase of the disease. When the other acute phase reactants returned to normal limits, the serum haptoglobins were still

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above the normal values[10]. But at that time we could not undertake further prospective studies in the inactive period to investigate the exact point at which the serum haptoglobins returned to normal values, and we were not able to find any information in the literature on this subject.

The purpose of the present study was to identify the exact point at which the serum haptoglobins returned to their normal values following acute rheumatic fever.

Material and Methods

Thirty-seven patients between the ages of 6 and 12 years (mean age 9 years) with acute rheumatic fever who were examined in the Department of Pediatrics at the Uludağ University Hospital and 21 age-matched controls were included in this study. Sterile serum samples were obtained from each of the 37 subjects, all manifesting acute rheumatic fever by rigid Jones criteria[11], and from the controls. All serum samples were taken between 5 and 80 days after the clinical onset of the disease and stored at 4 °C prior to the study.

In selecting the patients for this research, we took care that they did not have any infections other than rheumatic fever. The specimens were obtained during hospitalization and at follow-up visits in the outpatient clinic of the hospital, and no patient was receiving steroids at the time of collection of the blood samples.

Patients and controls were divided into 3 groups:

Group I consisted of 20 children in whom acute rheumatic fever was active. The follow-up samples were taken at various stages of active and inactive periods of the disease in these patients. In 10 of them serum haptoglobins were determined more frequently (1 sample every fortnight, totaling 5-8 samples).

Group II was composed of 37 children with inactive rheumatic fever. Twenty cases in this group were the patients in Group I, in whom the disease had entered an inactive stage. The remaining 17 patients were in an inactive period when we began this investigation. Nevertheless the acute phase of their illness had been previously observed by one of us. The erythrocyte sedimentation rate and C-reactive protein values in these patients were normal.

Group III consisted of 21 control children between the ages of 5 and 12, in good health with no history of rheumatic fever or other infections. Their economic and racial background, as well as their geographic location, were essentally the same as those of the study patients.

The haptoglobin-hemoglobin complex was separated from free hemoglobin by gel filtration using Sephadex G-100. Estimation is made by measuring the bound hemo-

globin or the ratio of free and bound hemoglobin to the total amount of hemoglobin [12].

Student's t test was used for statistical analysis.

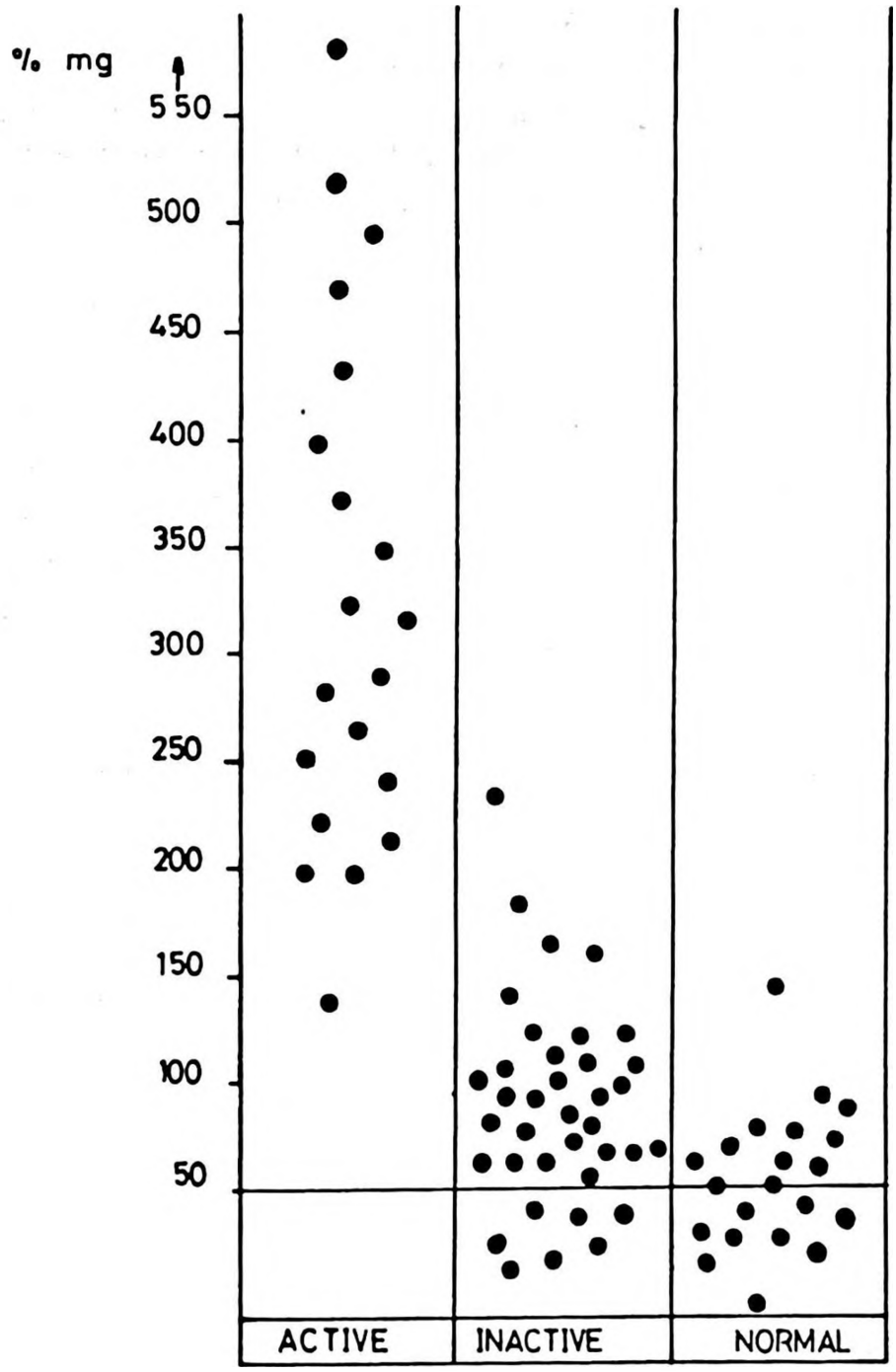

Figure 1

The mean haptoglobin values in active and inactive rheumatic fever patients and normal healthy controls.

Results

In Figure 1 the individual values of serum haptoglobin for patients with acute rheumatic fever as well as for patients in the early inactive stage of rheumatic fever and normals are shown. As can be seen from the figure, the serum haptoglobin levels markedly increase in the acute stage of the disease.

In Table I, the results of serum haptoglobin determination in the 20 children with acute rheumatic fever are compared with the 37 inactive rheumatic fever patients and 21 normal healthy controls. The mean level of serum haptoglobin in the children with active rheumatic fever was 319 mg/dl, well above the mean level of the children with inactive rheumatic fever (82.1 mg/dl), ($p < 0.01$), and the normal level of 52.5 mg/dl ($p < 0.001$).

TABLE I

Mean Haptoglobin Values

Groups	n	Mean haptoglobin level ($\bar{x}$) mg/dl	SD	SE	Range
Acute rheumatic fever	20	319	180	45	130-873
Inactive rheumatic fever (early stage)	37	82.1	44.1	7.3	27-230
Normal controls	21	52.5	26.1	5.7	10-136

When serial determinations of serum haptoglobin were carried out in 10 patients with the disease, it became apparent that the levels of haptoglobin were not returning to normal values as the activity of the disease subsided. In Figure 2, the haptoglobin, erythrocyte sedimentation rate and anti-streptolysin O values and C-reactive proteins are shown for one patient from the beginning of the disease until four months later. As can be seen in the figure, when the erythrocyte sedimentation rate had returned to the normal range, the haptoglobins still remained above their normal limits. The other cases from whom we obtained serial determinations of haptoglobin, erythrocyte sedimentation rate, anti-streptolysin O and C-reactive protein gave results similar to those shown in Figure 3.

The mean values for serum haptoglobin, erythrocyte sedimentation rate and C-reactive protein in the patients with acute rheumatic fever over a 2.5-month period are shown in Figure 4. Even though erythrocyte sedimentation values returned to normal levels within a month, the same process took much longer (approximately 2.5 months) for haptoglobins.

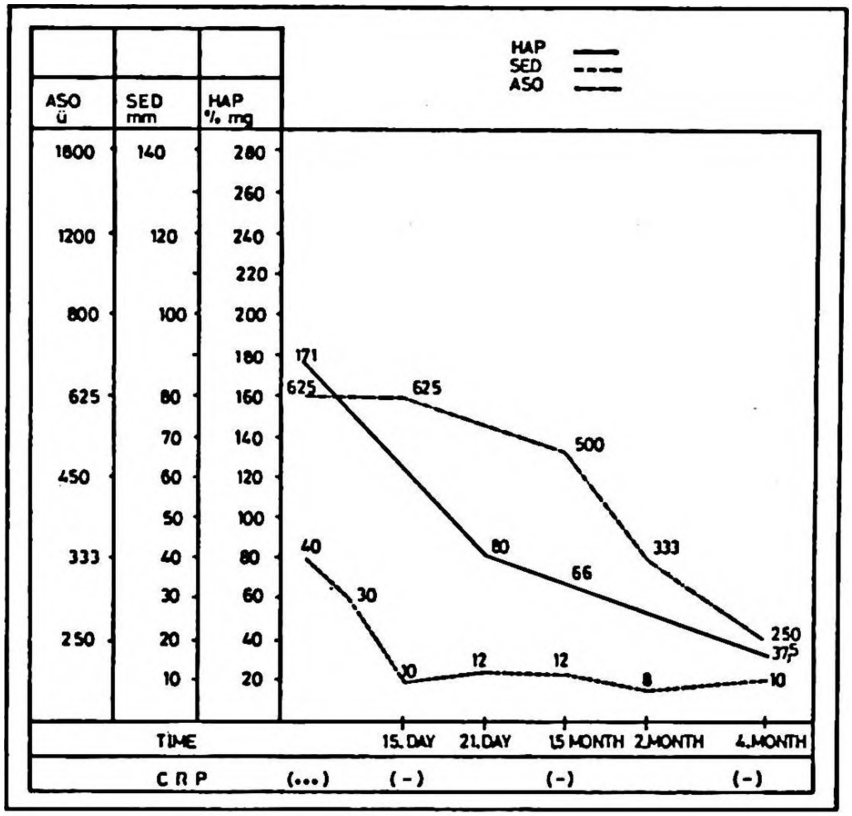

Figure 2

Serial determinations of serum haptoglobin, erythrocyte sedimentation rate, anti-streptolysin O titer and C-reactive protein in one patient with acute rheumatic fever.

Discussion

The present study has been undertaken to determine whether serum haptoglobin values can be utilized as a true acute phase reactant. The data presented in this study demonstrate that the amount of serum haptoglobin markedly increases in the acute phase of rheumatic fever. The reason for acceleration of haptoglobin production in rheumatic fever is not clear. It is probably due to release of a material from the inflamed tissue that can situmulate its hepatic synthesis. As suggested by Jayle and Moretti[13] haptoglobin might represent the building material produced by the liver, which would be fixed by injured tissue, supplying the elements necessary for biosynthesis of the scar tissue.

According to the present study, the increase in the serum concentrations of haptoglobin is, however, not limited to the acute phase but lasts for about two months. More rapid normalization of the serum haptoglobins has been observed in patients with a favorable evolution and adversely, a more prolonged elevation or a new peak has been observed in cases with a less favorable evolution.

Previously it was shown Owen et al[14] and Bernstein and Allerhand[15] that serum haptoglobins rise in acute rheumatic fever. However, Bernstein et al[16] also put

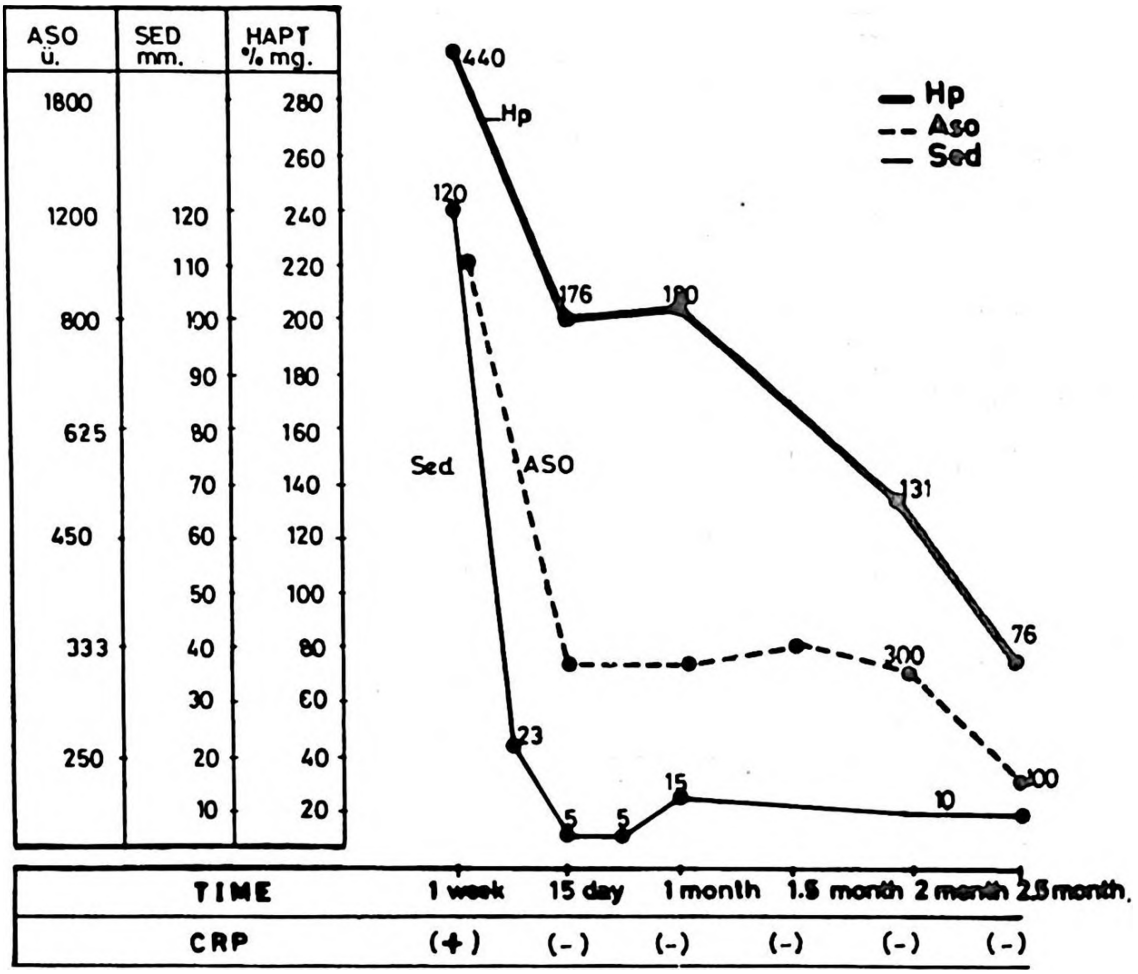

Figure 3

Erythrocyte sedimentation rates, ASO titers and serum haptoglobin values in a patient with acute rheumatic fever.

forward the theory that the levels of haptoglobin returned to normal values when the activity of the disease subsided. This suggestion is not in accordance with our results. Our observations have shown that the increase in serum haptoglobins is not limited to the acute phase of the disease. When the erythrocyte sedimentation rate decreased and returned to normal, serum haptoglobins were still above the normal values. Though it differed from case to case, the normalization of serum haptoglobin occurred between the second and fourth months, generally within two and a half months in our studies.

No significant difference in mean haptoglobin levels was noticed when the rheumatics were grouped according to their major clinical signs.

In our previous study we had also demonstrated an apparent elevation in serum haptoglobin levels by applying the peroxydase activity method to 67 patients with acute rheumatic fever, and these elevations were not different whether the patients had carditis or not [10].

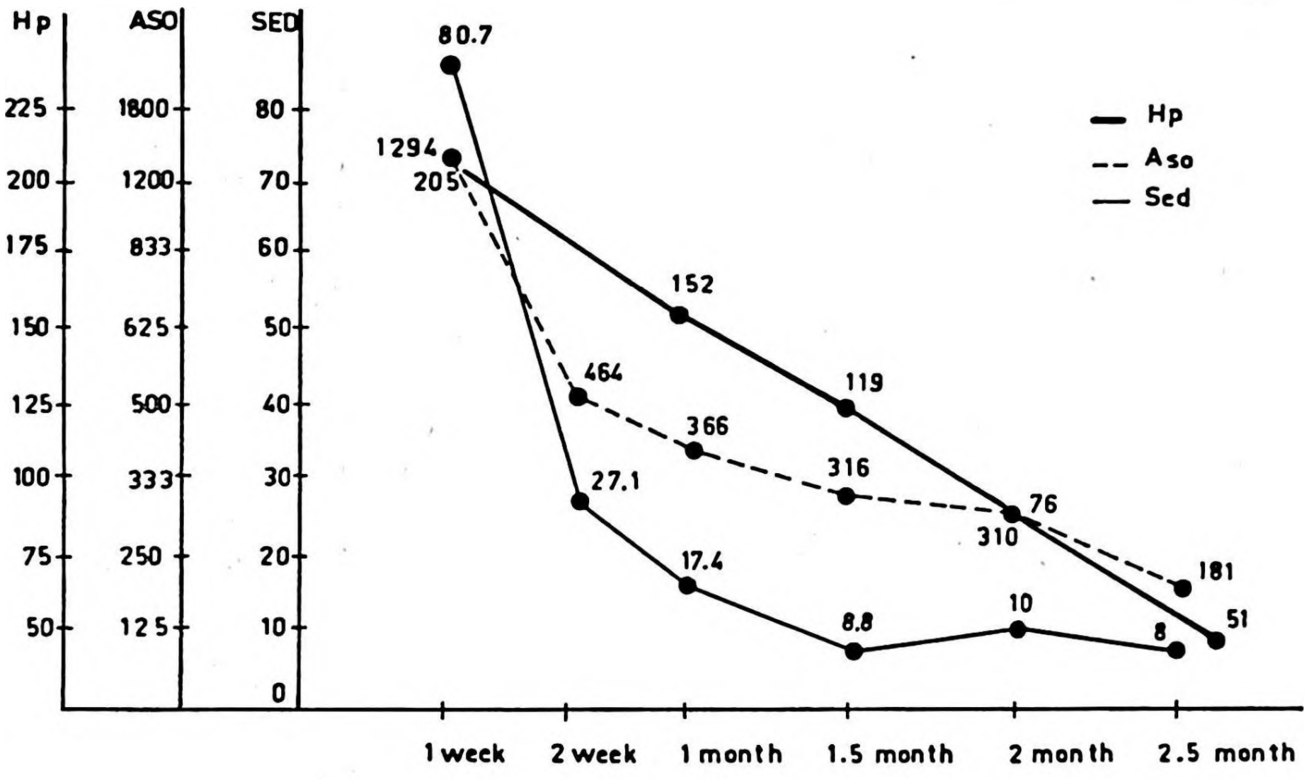

Figure 4

Mean values of serum haptoglobin, erythrocyte sedimentation rate and ASO titers in patients with active rheumatic fever.

Murray et al[17] investigated the haptoglobin phenotypes in 408 patients with rheumatic fever. Among these cases they found a number of patients with hypohaptoglobinemia. In our series we had only one case with a low haptoglobin level.

In conclusion, from a clinical point of view the normalization of haptoglobin indicates the healing of the infected areas whereas high levels may indicate that the infection is still present.

Summary

Utilizing the Sephadex-100 gel filtration technique, haptoglobin levels were determined in specimens taken from 20 patients with active and 37 patients with inactive rheumatic fever, and these groups were matched with 21 normal subjects of the same ages who served as controls.

A definite elevation in serum haptoglobin levels (mean: 319 mg ± 180) was noted in the active rheumatic group, but serial determinations revealed that in the early inac-

tive stage of the disease serum haptoglobins (82 mg ± 44.1) were still above the normal values (52 mg ± 26.1).

Normal haptoglobin levels were obtained within two to four months after the onset of the disease. From a clinical point of view, it is probable that the normalization of the haptoglobin level may be a reliable indicator of the complete inactivity of the disease.

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