Assessment of serum bilirubin and hepatic enzymes in malaria patients

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Abstract
Objective: The present study was conducted on malaria patients to observe the correlation between liver enzymes (SGOT, SGPT & ALP) and bilirubin.

Material & Methods: The study population contained 100 subjects divided into two groups, 60 malaria patients and 40 healthy control subjects of varying age groups and both sex. All biochemical parameters Total bilirubin, Direct bilirubin, Indirect bilirubin, SGOT (aspartate transaminase), SGPT (alanine transaminase) & ALP (alkaline phosphatise) were analyzed by semiautoanalyser. Statistical analysis was done by, science (SPSS 16) Software

Results: In our study we have found that (Mean ± SD) of Total serum bilirubin in malaria patients were 1.63±2.27 & controls were 0.67±0.08. Direct bilirubin in malarial patients were 0.92±1.3 & control were 0.37±0.07. Indirect bilirubin in malarial patients was 0.68±0.95. We observed that (Mean ±SD) of SGOT in malaria patients was 48.29±28.89 & in control were 29.28±7.44, the level of SGPT in malarial subjects were 44.9±27.15 & control subjects were 29.98±7.77 & the level of ALP in malarial patients were 98.47±60.67 & in control 71.4±25.12. We also found that both aminotransferases (SGOT & SGPT) showed significance positive correlation with serum total bilirubin whereas in case of ALP, significance correlation could not be obtained.

Conclusion: Our study indicates that liver enzymes SGOT, SGPT and ALP significantly increases in malaria patients as compared to control subjects. Therefore these enzymes may be useful in diagnosis of malaria subjects.

Keywords: Total bilirubin, Metabolic syndrome, Antioxidant

1. Introduction
Malaria is a devastating parasite transmitted by the bite of infected Anopheles mosquitoes. It is responsible for infecting 300-500 million and 1-3 deaths annually. In humans, malaria is caused by Plasmodium falciparum, Plasmodium vivax, Plasmodium ovale, Plasmodium malariae, and rarely Plasmodium Knowlesi. Plasmodium vivax is the commonest of the malaria species. Malaria can be transmitted by three known ways: vector transmission, blood transfusion and congenital transmission. The malaria parasite interferes with 3 major organs in the body, namely: the brain, kidney and liver.

Malarial transmission to the human host is established by sporozoites infection to the liver. The malarial sporozoite once injected into the blood by the bite of female anopheline mosquitoes is attached to the hepatocytes through the receptor for thrombospondin and receptor. Here these sporozoites become mature to form tissue schizonts or become dormant hypnozoites. Tissue schizonts amplify the infection by producing large number of merozoites. Merozoite infects and ruptures the liver cells in an attempt to escape back into the circulation and continues the infection. The infection of liver cells by the sporozoites form of the malarial parasite can cause organ congestion, sinusoidal blockage and cellular inflammation. These changes in hepatocytes can lead to the leakage of parenchymal (transaminases) and membranous (alkaline phosphatase) enzymes of the liver to the circulation. Hence increase in liver enzymes AST, ALT and ALP observed in malaria infected patients also demonstrated that the serum activities of these liver enzymes increased with the increase in malarial parasite density and confirmed that the hepatic stage of the parasite’s life cycle in human host is accompanied by significant perturbation in the hepatocyte’s parenchymia and membrane leading to leakage of liver enzymes into the general circulation. Jaundice is also one of the common manifestations of severe malaria in adults causing high mortality rate and incidence of jaundice vary from 10-45% in different regions.

In India, the epidemiology of malaria is complex because of geo-economical diversity, multiethnicity and wide distribution of nine anopheline vectors transmitting three Plasmodial species: P. falciparum, P. vivax and P. malariae. Anopheles culicifacies is widely distributed and is the principal vector of rural malaria. The proportion of P. vivax and P. falciparum varies in different parts of India. Mostly indo-gangatic plains and northern hilly states, north-western India and southern Tamil Nadu state have <10% P. falciparum and rest are P. vivax infections. In the forested area inhabited by the ethnic tribes, the situation is reversed with P. falciparum proportion 30-90% whereas in the remaining areas it is between 10-30%.

Malaria causes catastrophes such as maternal/infant death and abortion. Pregnant women are also especially vulnerable. Susceptible groups are children and adults who have lost or never acquired immunity. Morbidity and mortality due to malaria have remained unabated primarily as a result of the unavailability of suitable vaccines and the spread and intensification of drug resistant Plasmodium parasites. Thus it is...
rational to assess the current status of malaria related complications in order to estimate the burden among biologically risked groups, children and endemic areas.

2. Materials and methods

The subjects included in the study were 60 clinically diagnosed patients suffering from malaria of both sex and varying age groups, attending the out-patients department (OPD), emergency ward and from indoor patients, admitted in the ward of department of Medicine Gold Field Institute of Medical Sciences & research Village Chhainsa, Ballabgarh, Faridabad, Haryana. Forty healthy controls were selected for the study from volunteers such as paramedical staff, healthy relatives / attendants of patients. The patients comprised 42 males and 18 females. The control group comprised of 18 males and 22 females.

Patients selection criteria: Patients whose case history showed a concomitant presentation with the following conditions; pregnancy, renal diseases, liver diseases including cirrhosis, hepatitis, obstructive jaundice, alcoholism, cancer, metabolic bone diseases, gastrointestinal tract infection, protein energy malnutrition, diabetes, heart failure, infectious mononucleosis and magnesium /vitamin D deficiencies, were excluded from the study. This is because these conditions are associated with significant changes in serum alkaline phosphatase, alanine and aspartate transaminases activities. Similarly, patients on self medication with any antimalarial drug prior to presentation were also excluded from the study. Blood samples were collected by clean veinpuncture and centrifuged. Sera was collected and analysed for serum bilirubin and enzyme activity of ALP, SGOT and SGPT using kit method by erba chem semiautoanalyzer.

2.1 Statistical analysis

Statistical analysis was done, using the statistical package for social science (SPSS 16) for Windows Software. Differences in the parameters between the groups were analyzed by means of the t test. Variables were presented as mean ± standard deviation (S.D.). The accepted level of significance for all statistical analyses used in the study was P ≤ 0.05.

3. Results

Levels of serum bilirubin and liver enzymes (SGOT, SGPT, and ALP) were increased in the patients with malarial infection as compared to the controls and the increase was statistically highly significant (p<0.01). (Table 1)

Table 1: Comparison of serum bilirubin and liver enzymes among controls and malaria patients

| Variables | Controls | Malaria patients | p value |
|-----------|----------|-----------------|--------|
| Total bilirubin (mg/dl) | 0.67±0.08 | 1.63±0.27 | 0.001* |
| Direct bilirubin (mg/dl) | 0.37±0.07 | 0.92±0.13 | 0.001* |
| Indirect bilirubin (mg/dl) | 0.3±0.02 | 0.68±0.95 | 0.001* |
| SGOT (IU/L) | 29.2±7.44 | 48.29±28.89 | <0.001** |
| SGPT (IU/L) | 29.98±7.77 | 44.9±27.15 | <0.001** |
| ALP (IU/L) | 71.4±25.12 | 98.47±60.67 | <0.001** |

*→significant, **→highly significant

Significant increase in serum bilirubin & liver enzymes was obtained in malarial female patients as compared to the control (Table 2).

Table 2: Comparison of serum total bilirubin and liver enzymes among female controls and female malarial patients

| Parameters | Control | Test | p value |
|------------|---------|------|--------|
| Total bilirubin | 0.67±0.07 | 1.25±0.76 | <0.01* |
| SGOT | 29.17±6.67 | 51.67±3.67 | <0.01* |
| SGPT | 29.61±7.58 | 48.65±22.95 | <0.01* |
| ALP | 71.81±19.27 | 104.87±95.85 | <0.01* |

*→significant

Significant increase in bilirubin and hepatic enzymes levels was obtained in malarial male patients as compared to the controls (Table 3).

Table 3: Comparison of serum total bilirubin and liver enzymes among male controls and male malarial patients

| Parameters | Control | Test | p value |
|------------|---------|------|--------|
| Total bilirubin | 0.68±0.09 | 1.84±2.66 | <0.01* |
| SGOT | 29.41±6.43 | 47.13±30.43 | <0.01* |
| SGPT | 30.24±7.84 | 43.37±28.28 | <0.01* |
| ALP | 72.61±31.4 | 95.17±36.54 | <0.01* |

*→significant

Significant difference was not obtained in liver function parameters. (Table 4)

Table 4: Comparison of serum total bilirubin and liver enzymes among male and female malarial patients

| Parameters | Male | Female | p value |
|------------|------|--------|--------|
| Total bilirubin | 1.84±2.66 | 1.25±0.76 | 0.19 |
| SGOT | 47.13±30.43 | 51.67±3.67 | 0.54 |
| SGPT | 43.37±28.28 | 48.65±22.95 | 0.45 |
| ALP | 95.17±36.54 | 104.87±95.85 | 0.68 |

Both aminotransferases (SGOT and SGPT) showed statistically significant positive correlation with serum total bilirubin levels whereas in case of ALP, significant correlation could not be obtained. (Table 5)

Table 5: Pearson correlation coefficient among bilirubin and liver enzymes

| Enzymes | Bilirubin | p value |
|---------|----------|--------|
| SGOT | 0.642* | <0.001** |
| SGPT | 0.673* | <0.001** |
| ALP | 0.037 | 0.39 |

**→highly significant
4. Discussion

Investigations into the effects of *Plasmodium* parasites on the levels of serum enzymes have gained recognition as an important area of research in the pathogenesis of malaria. Malaria involves the liver where infective sporozoites invade and multiply in the hepatocytes and in the erythrocyte stage the merozoites cause the destruction of infected red blood cells. Malyneux et al suggested that jaundice, which may be deep, is usually accompanied by only moderate elevation of hepatic enzymes and results more from hemolysis than from hepatic damage. The role of liver injury or hepatocellular damage in the malarial patients has been proposed by many workers especially in the Indian subcontinent. Raised serum bilirubins, hepatomegaly along with increase in liver enzymes are important denominator of liver injury in these patients.

In our study, level of serum total bilirubin ranged from 0.5-15.2 mg/dl. Mean serum total bilirubin was significantly higher as compared to controls (p<0.01). Out of 60 malarial patients, 23.3% had increased serum total bilirubin level. Of these 78.6% patients had mild jaundice (2-5 mg/dl), 14.3% had moderate jaundice (5-10 mg/dl) and 7.1% had severe jaundice (>10mg/dl). Our finding was in accordance with Kocher et al who showed that 70.6% of children suffering from malaria (P. vivax) had mild jaundice where moderate and severe jaundice was present in 23.5% and 5.9% of the children respectively. Patwari et al observed jaundice in only 8.7% on P. vivax malarial cases.

We observed statistically significant increase in levels of enzymes SGOT, SGPT and ALP in malarial patients (p<0.001) as compared to controls. These enzyme activities were also significantly higher in test males and test females as compared to their respective controls (p<0.01). But we could not find the significant difference in the mean enzyme activities in both male and female malarial patients (p>0.05). Our finding was supported by that of Jigam AA et al. According to them transaminases and ALP activities were significantly higher in patient groups but when compared (p<0.05). Mohmad Ali et al showed increased activities of SGOT, LDH, ALP and CPK in patients with P. vivax malaria where in cases of *P. falciparum* malaria, enzymes ALP, SGOT and CPK activities decreased and LDH activity increased significantly. The increase in serum levels of hepatic enzymes; transaminases and alkaline phosphatase are the markers of liver damage. SGPT is a specific enzyme of liver. In this study we found that serum SGOT, SGPT and Alp levels were increased in 53%, 50% and 11.7% of the malarial patients. The mean level of AST was higher than that of ALS. It was similar to the findings of Kocher et al. Study of Noppadon et al showed that AST levels were increased in 26% of *P. vivax* infection, 31% of *P. malariae* infection and 40% of *P. ovale* infection. Similarly, increased ALT levels was observed in 21% *P. vivax*, 30% *P. malariae* and 40% of *P. ovale* infections. ALP was increased in 20%, 22% and 20% of *P. vivax*, *malariae* and *ovale* infections respectively.

Both the aminotransferases (AST, ALT) showed statistically significant positive correlation with bilirubin (p<0.001) whereas it was not significant in case of ALP (p>0.05). Kauser MW et al, in their study obtained significant positive correlation of serum transaminases and ALP with serum bilirubin (2). Kocher et al also showed an excellent positive correlation of SGOT with bilirubin (p<0.01).

5. Conclusion

Malaria is a disease whose pathogenesis is not clearly defined as it is species-specific and of geographical variability. Thus the assessment of bilirubin and liver enzymes (like SGPT, SGOT and ALP) in malaria patients could represent additional and useful parameters in determining the clinical and prognostic aspects of the disease.

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