Determination of serum calcium in patients with malaria by flame photometer

Mohsin Ali Baloch, Dr. Saira Baloch and Bikha Ram Devrajani

Abstract
Malaria is one of the most serious tropical and very widespread disease, it is a health problem in Pakistan. The objective of the present study was to determine the calcium level in the serum of Malaria Patients (n=50) with comparison to the control subjects (n=50), using flame photometer (m-410 corning). Serum calcium was increased in malaria patients as compared to the control subjects. Increased level of serum calcium is an aspect of severe malaria with the association of parasite and complications.

Keywords: Malaria, calcium, flame photometer

Introduction
Malaria is a devastating parasitic disease and the most deadly tropical diseases in the world-wide with a health risk to humans. It is a prevalent disease, cover-up several parts of Europe, North America, South America, Asia and Africa, similarly a health problem in Pakistan. The prevalence of malaria disease is affecting about three to five hundred million people a year [1]. Malaria is related to variations in weather, poverty, malnourishment and anti-malaria medicines and insect repellent [2, 3]. Micronutrients deficiency like calcium is more frequent amongst adults in developing countries [4]. Calcium deficiency is frequently related to enhanced susceptibility to infections like malaria [5].

Calcium is a necessary nutrient requisite through the previous phase of existence for support of body also vital role play for the subsistence of healthiness and nutritive health at all the periods in life [6]. Interested investigation on calcium, by considerable importance on its complicated collaboration through the diverse physical state of human. This happens in correlation with infectious diseases such as malaria. In Pakistan also, Malaria has been major communal and social health problem, it's still imperils millions of persons due to inadequate condition conducive to the spread of infection. Very little is known on the role of calcium in patients with Malaria, Calcium and metabolism are regulated and related to the activities of human being. In order to determine the calcium level in the serum of Malaria Patients (n=50) with comparison to the control subjects (n=50), using flame photometer (m-410 corning).

Materials and Method
Fifty venous blood samples (10ml) from male (n=31) and female (n=19) malarial patients and control subjects were collected into sample tubes without the addition of anticoagulant. The blood samples were centrifuged at 1500 rpm for 20 minutes; the serum was separated and immediately used for the determination of Calcium was analyzed using flame photometer (m-410 corning) [7].

All the chemicals and reagents obtain were of analytical grade obtained from Merck.

Determination of Serum Calcium

Method
The principal reaction is based on measuring, calcium without deprotonization, in human urine and serum. The calcium ion reacts with the methylthymol blue indicator (MTB) in an alkaline medium. The color intensity of the Ca-MTB complex, measured at 612 nm, is proportional to the quantity of calcium present in the sample 8-Hydroxyquinoline eliminates interference from proteins as shown below.
Ca++ +MTB --------------> Ca-MTB complex

Procedure
800 µL of reagent (R1) followed by 200 µL reagent ATP, Lipases, Peroxidase, Glycerol kinase, Glycerol-3-(R2) in a 5ml sample tube containing 10 µL blood serum are mixed and were allowed to stand for 10 minutes to complete the reaction. The absorbance was measured at wavelength 612-623 nm which showed the relationship between the content of alkaline phrophatase and the absorbance of Ca-MTB formation in a linear manner.[7]

Results and Discussion
Figure: 1 shows the serum calcium levels in malarial patients and control subjects, moreover, figure: 2 shows the gender distribution of male and female patients with malaria. All values are expressed as Mean ± S.D.

![Figure 1: Calcium Levels in Malarial Patients and Control Subjects](image)

It was reported that the decreasing of calcium detected in malarial patients who is caused by the symptoms and signs of malaria: temperature, rising pulse rate, perspiration, shaking [8] that interrupts neuromuscular impulsiveness, nerve conduction and muscular contraction. The biological fact involves calcium for their efficiency [9-11]. Low levels of calcium may cause by fatalities in gastrointestinal and kidney complications resulting malaria and that condition may cause a rise in urinary elimination of crystals like calcium. In the present study level of serum calcium was higher in malarial patients as compared to the control subjects. The verdict that hypocalcemia was infrequent at every stage differences by facts sequence's for seriously diseased [12]. In the present study, it was observed that high calcium levels in malaria patients as compared to the control subjects. Assessment the level of calcium may surrounded the specific rate of reduced ionized calcium levels, but rises in calcium levels are probably to contain rises in free ionized calcium levels. Intraerythrocytic calcium levels are significantly higher in parasitized red blood cells [13] and it is recommended that the high calcium levels caused by the intracellular liberation of calcium reduced to the likely erythrocyte lysis because of malaria. This is maintained by the link amongst hypercalcemia and severe malarial anemia, the converse association among serum calcium levels and an earlier record of this relationship with severe malaria [14]. The results of our study indicate that high level of serum calcium is an aspect of severe malaria. There are very few studies highlighted on its occurrence and experimental effects. High level of serum calcium in malaria may biochemical marker for complications.

Conclusion
Increased level of serum calcium is an aspect of severe malaria with the association of parasite and complications. It has a predictive significance in malaria, as improvement from malaria was linked with improving of serum calcium levels to average.

References
9. Planche T, Krishna S. The relevance of malaria

Fig 1: Calcium Levels in Malarial Patients and Control Subjects

Fig 2: Gender Distribution