

# Study of Serum Zinc, Vitamin A and Alkaline Phosphatase Status in Nyctalopia Patients

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## ABSTRACT

**Background:** Nyctalopia is a condition in which the person is unable to see in the dim light/dark since the dark adaptation time increases. The condition is due to deficiency of vitamin A; it is not an only one factor responsible for nyctalopia, along with that Zinc is also the metal ion enzyme responsible for the development of nyctalopia. The zinc ion is responsible for mobilization of stored form of vitamin A (retinyl palmitate to retinol) and results into the alternations of retinol processing in retinal pigment epithelial cells in the visual cycle. Hence the objectives of the present study were the estimation of serum vitamin A, Zn<sup>++</sup> and serum alkaline phosphatase (ALP) (zinc-dependent enzyme) had been added to see the effect in the present study.

**Methods:** The total 30 number of nyctalopia patients and 30 numbers of healthy subjects were selected for the present study.

**Sample collection and processing:** The fasting blood samples were collected in the plain bulb, and then it was used for laboratory processing like centrifugation and estimations. Serum vitamin A was estimated by Bessey method. Serum zinc was estimated by colorimetric (kit) method and serum ALP were estimated by the king and king method.

**Results:** In this study, the estimated serum vitamin A levels in the test was found to be mean  $\pm$  SD = 13.69  $\pm$  2.82 and controls were 29.04  $\pm$  1.97. The calculated  $p < 0.0000$  so it was statistically very highly significant. Serum Zn<sup>++</sup>, test was 18.87  $\pm$  2.47 and controls were found to be 61.86  $\pm$  1.74. The calculated  $p < 0.0000$ , so it was statistically very highly significant. But in case of the calculated serum ALP, it had been found to be nearly the same (very trace increase) in test and control groups of the patients, i.e., 545.97  $\pm$  39.79 and 546.20  $\pm$  39.77, and the  $p$  value was found to be  $p < 0.98$ . So it was not found significant.

**Conclusion:** In this study, the serum vitamin A and Zn<sup>++</sup> had been found to be decreased below the normal levels in nyctalopia patients. But the serum ALP levels had been found to be nearly the same (very trace increase) in test and control group patients.

**Keywords:** Alkaline phosphatase, Nyctalopia, Vitamin A, Zinc.

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## INTRODUCTION

Nyctalopia is one of the earliest symptoms of vitamin A deficiency, in this condition the patient unable to see in dark or dim light, since the dark adaption time is increased. It is an increased dark adaption time in humans, and it may be due to decreased concentration in nutritional vitamin A with serum zinc level.<sup>1,2</sup> Or Nyctalopia is a deficiency of vitamin A associated with visual changes, and depressed ERG.<sup>3</sup> Vitamin A is also necessary for the maintenance of specialized epithelial cell surfaces. Nyctalopia can lead to dryness or xerosis in the conjunctiva loss of goblet cells and squamous cells.<sup>3,4</sup>

Zinc (Zn<sup>++</sup>) is an important metal nutrient for the human body and acts as a cofactor for some important enzymes in the many reactions and transcription factors.<sup>5-7</sup> It is utilized during the growth and development of plants microorganism and animals. It is well distributed in the human body tissues and secretions with different concentrations, with muscle and bones 85%, liver and skin 11% and remaining will found in all other tissues with higher percentages, in the prostate, retina and different parts of the eye.<sup>8,9</sup>

Zinc deficiency may be due to the nutritional supplementation or malabsorption. It can affect ocular development. The severe deficiency of zinc, when studied in rats during gestation, it fails optic cup invagination, colobomata, retinal dysplasia and occasionally an ophthalmic in pumps.<sup>10</sup>

The other childhood diseases like acrodermatitis enteropathica (rare disease) were found in Zn<sup>++</sup> metabolism disorders.

The abnormalities in Zn<sup>++</sup> deficiencies like blepharitis, photophobia, conjunctivitis, corneal opacities, and cataracts are also reported in ocular abnormalities.<sup>11</sup>

In human studies, Zn<sup>++</sup> deficiencies result in poor dark adaptation and nyctalopia and it can be cured appropriate zinc dose.<sup>12-14</sup>

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In the visual cycle, the alternations of the retinol processing in retinal pigment epithelial cells are impaired since  $Zn^{++}$  deficiency, so the retinol dehydrogenase is Zn metalloenzyme which could not catalyze the oxidation of retinol to retinal in the visual cycle.<sup>15</sup>

In older adults, the cataracts are commonly found in all over the world, and it is the effect of lowering zinc level in blood which crystalline the lenses with mature senile cataracts has been reported in the patients with traumatic cataracts.<sup>16</sup>

Some studies reported that zinc induces and accelerate the metallothionein synthesis and metallothionein III which plays a provital role as an endogenous neuroprotectant against light-induced retinal damage.<sup>17,18</sup>

Alkaline phosphatase (ALP; EC-3.1.3.1) is a hydrolase enzyme responsible for removing phosphate groups from different molecules like nucleotides, proteins, and alkaloids.<sup>19</sup> It is a zinc-containing diametric enzyme with mol.wt.86000Da, stable to heat.<sup>20</sup> Serum ALP levels are significantly higher in children, pregnant women, during bone formation, osteoblastic activity and Paget's diseased bone. Hypophosphatasia seen in aging, postmenopausal women, heart surgery, malnutrition,  $Mg^{++}$  deficiency anemia, hypothyroidism, cretinism, pernicious anemia, aplastic anemia, and Wilson disease.<sup>21,22</sup>

Selection of the above biomarkers for the present study having the great importance in nyctalopia condition. The previous studies show that there was a deficiency of vitamin A in nyctalopia patients. The metal ion co-enzyme  $Zn^{++}$  equally important and also responsible for the mobilization of retinyl palmitate (the stored form of vitamin A in the liver) to all-trans retinol from the liver and then, it can be directly picked up by the retina of eyes for the rhodopsin cycle for the normal vision. ALP is one of the zinc-dependent enzymes which blood level might be affected during the nyctalopia condition, so the above-mentioned parameters are added in the present study.

## MATERIALS AND METHODS

The study was conducted in the Department of Biochemistry, in collaboration with ophthalmology department at SMBT Institute of Medical Sciences and Research Center, Dhamangaon, Igatpuri, Nasik, Maharashtra, India.

### Sample Size Estimation

The sample size has been estimated by the prevalence of nyctalopia condition in Maharashtra. The prevalence is 0.48%.<sup>23</sup> So, by calculation, the sample size is found to be 7.64 and rounding up by 8. But it was a very small number

so; thirty numbers of patients have been included in the study in 2.5 years of time period.

### Study Design

The study is hospital-based, case-control study. The samples are collected from the SMBT hospital and peripheral hospitals from Nasik city and district.

The patients had been hospitalized with the complaints of night/dark visions were selected, and collected complete history of all the 30 patients. The visual activity was measured and eyes of all the patients were examined with a slit lamp test, and ophthalmoscope.<sup>24</sup>

The papillary thresholds of the admitted patients were assessed in a dark room by using a prototype scotopic (dim light vision) sensitivity instruments.<sup>25</sup> Then the patients were selected with their increased dark adaptation time. They were also cross-checked with their physical characters in detail including with vitamin A deficiency.

### Inclusion Criteria

The subjects, who described their inadequate or poor vision in the dark, evening and night, but had their adequate day time vision, were included as eligible cases for the study.

### Exclusion Criteria

The subjects with impaired their day time vision, were considered as false positive cases and excluded from the study.

### Sample Collection and Processing

The fasting blood sample was collected in the plain bulb, and then it was used for laboratory processing like centrifugation and estimations of the different parameters serum vitamin A was estimated by Bessey method.<sup>26</sup> Serum zinc was estimated by a colorimetric method ( $Zn^{++}$  kit method)<sup>27</sup> and serum ALP was estimated by the king and king method.<sup>28</sup>

### Statistical Analysis

Statistical analysis (calculations) of the data was performed by using Statistical Package for the Social Sciences (SPSS) latest software version. Student 'T' test was suitable so, applied for the calculation. A  $p < 0.005$  was supposed to be statistically highly significant.

## RESULTS

In this study, serum vitamin A,  $Zn^{++}$  and ALP were estimated and compared with the controls. In Table 1,

the statistically calculated serum vitamin A level very significantly decreased with controls ( $13.69 \pm 2.82$  and  $29.04 \pm 1.97$ ), In Table 2, serum  $Zn^{++}$  was also very significantly decreased in the test group ( $18.87 \pm 2.47$  and  $61.86 \pm 1.74$ ), and the  $p < 0.000$ .

Table 3, the serum ALP level had been found to be nearly the same (very trace increase) in test and control group patients,  $545.97 \pm 39.79$  and  $546.20 \pm 39.77$ . The  $p$  value is not significant ( $p$  is less than 0.98).

In the graphical study, serum vitamin A and serum  $Zn^{++}$  was significantly decreased as compared with

control. But serum alkaline phosphatase was nearly the same (Graph 1).

**DISCUSSION**

The study showed that the serum vitamin A and serum  $Zn^{++}$  is significantly decreased in nyctalopia. It might be due to the result of malnutrition, malabsorption, and poor vitamin metabolism or due to liver diseases (Graph 2).<sup>29</sup>

Along with the decreased level of vitamin A, serum  $Zn^{++}$  is also decreases below the normal in nyctalopia condition, here zinc does not interact with taurine and vitamin A in the retina, which depresses the modification of plasma membranes in the photoreceptors that causes or inhibits the regulation of light-rhodopsin reaction in the photoreceptors.<sup>11</sup>

Zinc deficiency can impaired the mobilization of retinyl palmitate to retinol and retinyl binding protein (RBP) synthesis in liver and results into decreases the concentration of RBP in plasma.<sup>30</sup> Besides, there may be a specific role in the conversion of  $\beta$ -carotene to retinol through the 15'-15' dioxygenase enzyme.<sup>31</sup>

Thus, the zinc deficiency may result in a secondary vitamin—A deficiency that is reflected in low serum vitamin A concentrations.

Serum ALP level was not significantly affected in nyctalopia condition (Graph 3).

**CONCLUSION**

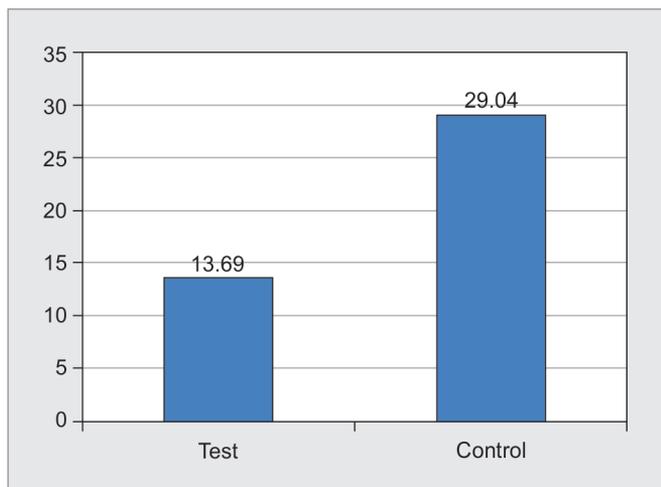
Serum vitamin A and Sr. zinc levels were decreases in nyctalopia patients and serum alkaline phosphatase level was not significantly affected.

**Table 1:** Sr. Vitamin A levels in nyctalopia patients

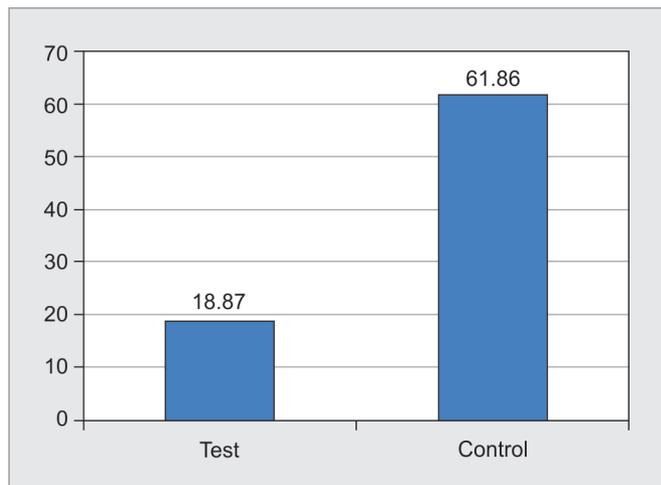
Group	Mean $\pm$ SD	p value
Test	$13.69 \pm 2.82$	0.0000
Control	$29.04 \pm 1.97$	

**Table 2:** Serum  $Zn^{++}$  levels in nyctalopia patients

Group	Mean $\pm$ SD	p value
Test	$18.87 \pm 2.47$	0.0000
Control	$61.86 \pm 1.74$	



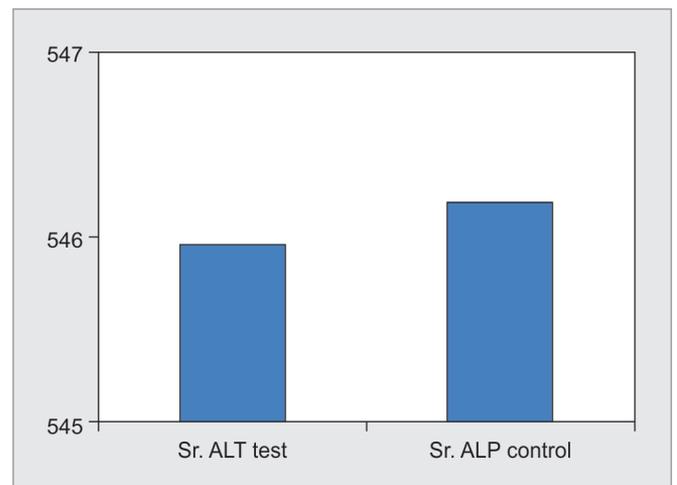
**Graph 1:** Sr. Vitamin A levels in nyctalopia patients



**Graph 2:** Serum  $Zn^{++}$  levels in nyctalopia patients

**Table 3:** Serum ALP levels in nyctalopia patients

Group	Mean $\pm$ SD	p value
Test	$545.97 \pm 39.79$	0.98
Control	$546.20 \pm 39.77$	



**Graph 3:** Serum ALP levels in nyctalopia patients

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