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CHOLECALCIFEROL INDUCED REGULATION OF CALCIUM AND INORGANIC PHOSPHATE IN CAPRA AEGAGRUS HIRCUS EYE LENS

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ABSTRACT

Capra aegagrus hircus eye lenses were incubated in Ringer's solution in presence of Cholecalciferol (vitamin D_3) (40,000I.U) for different time period *i.e.* 0, 15, 30, 45 minutes and 1.0, 2.0, 4.0, 8.0 and 12.0 hours. The result indicated that the fortification of vitamin D_3 in the medium was more effective within first 15 minutes of incubation period consequently decreased the inorganic phosphate and calcium levels in lens.

Figures : 02 References : 03 Table : 00

KEY WORDS : *Capra aegagrus hircus,* Calcium and inorganic phosphate, Cholecalciferol (vitamin D_3), eye lenses, Ringer's solution.

Introduction

Cells not only maintain the Ca^{2+} concentration of cytoplasm below that of the surrounding fluids, but also possess the capacity to regulate their internal calcium concentration. Amongst all tissues in the mammal, normal eye has the highest concentration of calcium. Calcium has an important role in lens including the maintenance of its metabolic activity, permeability and configuration of proteins which are considered important for the maintenance of transparency of eye lens. However, when calcium concentration increases or decreases outside the normal value, the cortical cells of the lens is restructured and lead to the opacification of lens with eventual formation of cataracts. Calcium has been associated with the formation of cataract in both human and calf lenses. In the bovine lens concentration of ATP, ADP, AMP, Pi (inorganic phosphate) and lactate decrease during aging. Calcium ions have been observed to induce the formation of high molecular weight proteins aggregates in lens homogenates which lead to the opacification. It was reported that in young rats cataractogenic subcutaneous dose of sodium selinite causes a threefold increase in calcium and a two fold increase in inorganic phosphate in the lens before development of nuclear cataract[2]. The purpose of this investigation was to understand the regulation of calcium and phosphorus

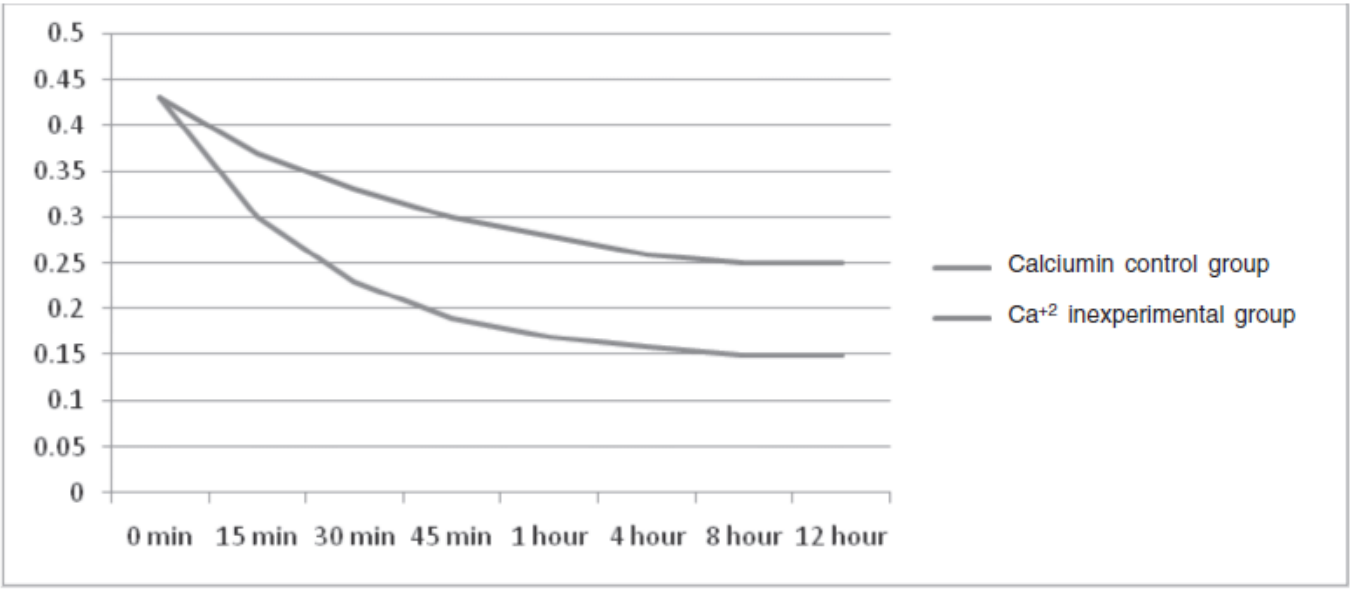

Fig.1 : Effect of vitamin D_3 on calcium

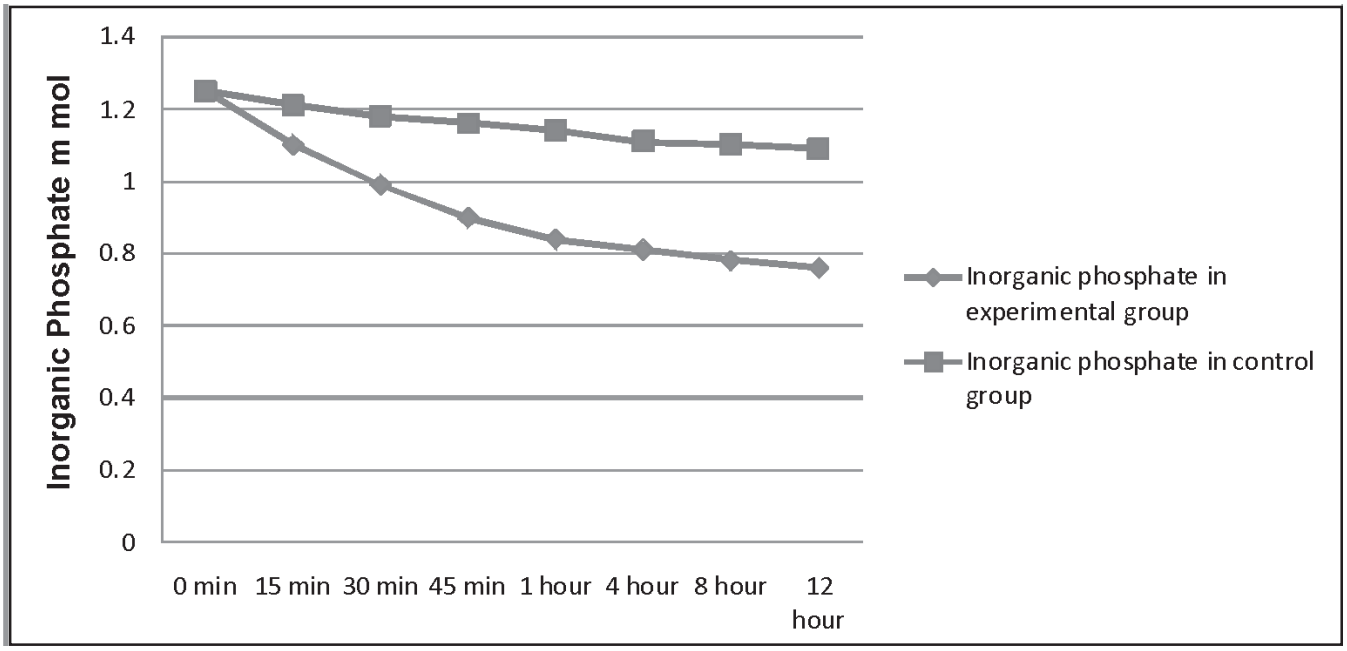

Fig.2 : Effect of vitamin D_3 on Inorganic Phosphate (HPO_4^{-2})

levels in goat lens in the presence of vitamin D_3.

Materials and Methods

Fresh goat eyes were collected from local butcher's shop. The eyes were put in pairs into ice cooled flasks and brought to the laboratory; operations were performed under strictly sterile conditions. All incubation experiments were carried out in parallel for experimental and control. The lenses dissected from the eye ball were incubated in two groups (I & II). In group I (control), lenses were incubated in 5.0 ml Ringer solution (containing NaCl 140 mM $MgCl_2$ 2mM, KCl 5m M, glucose 5m M, $NaHCO_3$ 25 mM & $CaCl_2$ 0.2m M10) while in group II (experimental) lenses were incubated in same volume of Ringer's solution containing 40000I.U of vitamin D_3. After incubation periods of 0, 15,30,45, minutes and 1.0, 2.0, 4.0, 8.0 & 12 hours, lenses from both control and experimental groups were taken out and blotted on a filter paper moistened with EGTA (ethylene glycol tetra acetic acid) (1m M) to remove capsular water containing calcium from the medium. Lenses were then homogenized in 2 ml of tris HCl (pH 7.4, 0.1 M) and protein was precipitated by adding equal volume of TCA (Trichloroacetic-Acid) (10%). Calcium was determined by atomic absorption spectrophotometry from the supernatant obtained after 3000 rpm for 10 minutes and inorganic phosphate from the supernatant by spectrophotometric method.

Results

The results of this investigation (Fig.1) indicate that vitamin D_3 produced pronounced effect during first hours of incubation period. The results show that vitamin D_3 was more effective within first 15 minutes of incubation period. This is apparent by significant decrease in the level of calcium by 0.15 m mol and 0.06 m mol (Fig. 1) in case of experimental and control groups respectively. However, the level of inorganic phosphate decreased in the initial stage of experimental group whereas there was no apparent change in the level of inorganic phosphate in control group (Fig. 1)

Discussion

The results of the present investigation suggest that the decrease in the level of calcium and inorganic phosphate in lens can occur either as a result of direct effect of vitamin D_3 on lens or through the conversion of its active metabolite, or else through some other mechanisms. It is possible that the enzyme responsible for the production of 1á, 25 dihydroxy vitamin is present in the lens epithelium. Such an enzyme is present in renal mitochondria of human, rat and dog. It can thus be suggested that vitamin D_3 mobilises calcium and inorganic phosphates directly from the lens matrix this results in accumulation of calcium from the lens.

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