

## Effects of acute overdose of vitamin A on the hepatic content of K, Na, Mg, Fe, Cu and Zn, in rats

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**SUMMARY.** Interactions among vitamin A metabolism and several metals have been reported in both normal and pathological situations. In the present report we studied, in rats, the effect of daily injections of 100.00 U.I. of vitamin A during seven days on the content of K, Na, Mg, Fe, Cu and Zn in the whole liver. The results were compared with the findings in pair-fed non-treated animals (Control Group). The mean  $\pm$  SEM for the concentrations of these elements in the control group were  $3.433 \pm 234$  (K),  $649 \pm 19$  (Na),  $239 \pm 5$  (Mg),  $169 \pm 7$  (Fe),  $5 \pm 0.1$  (Cu) and  $31 \pm 2$  (Zn)  $\mu\text{g/g}$  wet tissue, respectively. The hypervitaminosis A, confirmed by a significant increase in tissue concentration of the vitamin, altered the hepatic content of the above mentioned cations. While Na and Zn increased, the other cations decreased its concentrations in the whole liver. Possible mechanisms for these findings are discussed and it is concluded that high doses of vitamin A result in marked changes in the hepatic content of the studied metals.

Key words: Acute hypervitaminosis A, electrolytes, liver.

**RESUMEN.** Efecto de la hipervitaminosis A aguda sobre el contenido hepático de K, Na, Mg, Fe, Cu y Zn, en ratas. Publicaciones previas han señalado marcadas interrelaciones entre diversos metales y el metabolismo de la vitamina A, en condiciones normales y/o patológicas. En el presente trabajo se estudió, en ratas blancas, el efecto de 100.000 U.I. de vitamina A inyectadas diariamente, durante siete días sobre el contenido de K, Na, Mg, Fe, Cu y Zn en hígado total y se comparó con lo que sucede en animales no tratados. Las concentraciones de estos elementos (medias  $\pm$  DE) en el grupo control fueron  $3.433 \pm 234$  (K),  $649 \pm 19$  (Na),  $239 \pm 5$  (Mg),  $169 \pm 7$  (Fe),  $5 \pm 0.1$  (Cu) y  $31 \pm 2$  (Zn)  $\mu\text{g/g}$  de tejido húmedo, respectivamente. La hipervitaminosis A aguda, confirmada por el aumento significativo de la concentración hepática de la vitamina, incrementó el contenido tisular de Na y de Zn con una disminución de los restantes. Se discuten los mecanismos que explican estos cambios y se concluye que la hipervitaminosis A aguda determina marcadas alteraciones en el contenido de estos metales en el hígado de ratas blancas.

Palabras Claves: Hipervitaminosis A aguda, electrolitos, hígado.

### INTRODUCTION

Vitamin A (VA) is essential for a variety of cellular functions. Its role in vision has been extensively investigated, and it is also important in the regulation of growth, the control of reproductive functions, the stabilization of biomenbranes, and in intermediate metabolism. An excess of the substance, as well as deficiency, causes disturbances of normal cellular functions.

Metals have an important function in practically every biochemical and physiological process, such as muscle contraction, transport across membranes, nervous conduction, regulation of immune function and also participate in catalytic enzymatic processes.

Interactions among VA metabolism and metals have been reported in both normal and pathological conditions. Several studies have indicated a biological interaction between VA nutriture and Fe nutrition and metabolism (1,2). Smith et al (3), in studies involving humans and experimental animals, suggest a role for Zn in vitamin A metabolism. Theses authors found that low Zn plasma levels coincided with low hepatic and serum levels of retinol. Furthermore, high doses of vitamin A modify the serum and hepatic levels of Ca and P (4). Certain metals like Cd (5,6) Fe (7) and Cu (8) influence several phases of vitamin A metabolism. Thus, high doses of Fe lower the hepatic content of VA (7); whereas, Cd ingestion produces a significant decrease of serum VA levels concomitantly with and increase in liver VA levels (5). Moore et al (8) observed a severe decrease in plasma VA on Cu poisoning in sheep and a rise in plasma VA as plasma Cu levels fell. Little is know about the possible changes in hepatic metal ions concentrations during hypervitaminosis A. Therefore, based on these results we postulate that high doses of VA may significantly change

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the hepatic content of Cu, Zn and other metals as well. In the present study we have determined the potassium, sodium, magnesium, iron, copper and zinc content in the liver after high doses of VA, in rats.

#### MATERIALS AND METHODS

Sixty albino Sprague Dawley rats weighing between 180 and 200 g were included in the study. They were housed in individual metabolic cages. The animals had free access to water and food during the experimental period. The formula for the basal diet was similar to that recommended by the American Institute of Nutrition (9). After 10 days of adaptation the animals were randomly divided in two groups:

Group I (n=30) received a daily intramuscular injection of 100.000 U.I. of retinol palmitate for 7 days (Total dose for rat= 700.000 I.U). (10-12).

Group II (n=30) received a daily intramuscular injection of 1 mg of palmitate for seven days.

At the end of the seven day period, all the animals were sacrificed by decapitation under light ether anesthesia. To minimize blood contamination, the liver was perfused in situ through the portal vein, with a cold 0.25M sucrose (w/v) solution. At the end of the perfusion, the liver was dissected and placed on a Petri dish on ice. Aliquots of 1g were prepared and digested with acid according to Burguera et al (13). The flow injection analysis/atomic absorption spectrophotometric technique was used for the determination of metal concentrations in the digest (14,15). The vitamin A content in liver was estimated by the method of Neeld and Pearson (16). Student's t-test was used for statistical analyses.

#### RESULTS AND DISCUSSION

The rats treated with high doses of vitamin A showed the following abnormalities: edema of the eyelide with secretion of mucus and pus and generalized hemorrhages. These findings were present in 75% of the animals. Other authors have reported the same phenomena (17).

Table 1 shows the hepatic concentrations of different metals. Vitamin A administration significantly ( $p < 0.05$ ) decreased the tissue content of potassium, magnesium, iron and copper. In contrast, sodium and zinc significantly increased. The hepatic level of vitamin A was significantly augmented ( $1.400 \pm 15.83$  ug/g) in comparison to control value ( $24.23 \pm 7.98$  ug/g).

TABLE 1  
HEPATIC CONTENT OF CATIONS\* IN THE CONTROL GROUP (GROUP II) AND IN THE GROUP THAT RECEIVED HIGH DOSES OF VITAMIN A (GROUP I)

Cations	Group II	Group I
K	$3.433 \pm 234$	$2.359 \pm 101^{**}$
Na	$649 \pm 19$	$1.150 \pm 60^{**}$
Mg	$239 \pm 5$	$147 \pm 9^{**}$
Fe	$169 \pm 7$	$106 \pm 5^{**}$
Cu	$5 \pm 0.1$	$2 \pm 0.2^{**}$
Zn	$31 \pm 2$	$86 \pm 4^{**}$

\* ug per gram of wet tissue (mean  $\pm$  SEM).

\*\* statistically significant difference respect to control at the  $p < 0.001$  level.

N= 30 rats by group

The liver concentrations of Na, K, Mg, Fe, Cu, Zn and vitamin A, in the control group, were very similar to those previously reported (18). The presence of acute hypervitaminosis A, as suggested by generalized hemorrhages (19) and confirmed by the elevated hepatic vitamin A levels, determined different changes in the tissue level of those cations. This corroborates our original hypothesis. The decrease of Fe content confirms the findings of Staab et al (7). These authors have suggested that VA is involved in the regulation of Fe release from the liver. The modifications in the liver content of Na, K and Mg seen here, might contribute to the hyponatremia, hyperpotasemia and hypermagnesemia previously observed in rats receiving overdoses of VA (20).

The changes in the activity of hepatic enzymes (21), and the decrease of basal metabolism (4) described in albino rats receiving high doses of VA, could be partially explained by the different changes in the hepatic levels of cations under study. Our results also support the hypothesis of Modis et al (11). According to this theory, «the polymorphic symptomatology of hypervitaminosis A involves a complex mechanism of action which compromises the integrity of cell membranes, the synthesis processes of the body and the great majority of intermediary metabolisms».

Based on our results, we propose the following mechanisms to explain the changes on hepatic cations provoked by acute overdoses of VA: a) A direct action of VA on cation metabolism, which would increase or decrease their release from the liver. This proposed mechanism has been confirmed for Fe (7). b) Necrosis of the hepatocytes, due to hypervitaminosis A (22), would cause a redistribution of Na, K and Mg (23) within the liver. c) Massive proteolysis in the liver (17), and the concomitant K loss (24). d) Coexistence of secondary infections that stimulate the production of Interleukin-1, a key mediator of animal response to infection, inflammation, injury and other stress factors that also modifies the metabolism of metal

ions (25,26). Interleukin-1 is now recognized as a cytokine, but it was originally identified as «leukocytic endogenous mediator» (25,26). This Interleukin-1 increases the hepatic levels of Zn and Fe and decreases the liver content of Cu (27) and f) the antagonism of the Fe, Cu and Zn in the liver (28).

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