BIOCHEMICAL STUDIES ON THE EFFECT OF D VITAMINS AND CO-CARBOXYLASE ON METABOLISM OF EXPERIMENTAL ANIMALS

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TO

MY BELOVED WIFE AND MY DEAR

SONS M., A. AND SH.

TO SOUL OF MY PARENTAL

CONTENTS

1- INTRODUCTION

In the past few years, much attention has been directed towards re-evalution of the nutritive requirements of animals. A part of this attention was directed towards the action of vitamin D and Co-carboxylase (Vitamin B_1).

Vitamin D is required for normal growth of animals. This is probably due to its effect on calcium and phosphorus metabolism .When the rate of bone growth is below normal, as in case of vitamin D difficiency, the rate of body weight is retarded. It appears that the effect of vitamin D on growth is closely related to its effect on bone development (Harrison and Harrison., 1960). Vitamin D is also necessary for normal calcification of bone, the absence of adequate amount from the diet results rickets in infants and osteomalacia in adults. When vitamin D is given to rachitic subjects the metabolic abnormalities are corrected and the zones of provisional calcification reassume a more normal appearance (Jeans, 1950).

Vitamin B_1 is an essential nutrient for all mammalian through the role of its function form (thiamin pyrophosphate or Co-carboxylase). It has been known the lack of thiamin results in the deficiency disease known as beriberi and that the coenzyme activity of thiamin diphosphate plays an important role in carbohydrate metabolism (Nishins ,1986). Thiamin deficiency was evidenced by anorexia, weight-loss (Mclane et al., 1987) and disturbances in calcium and phosphorus metabolism (Tazhibaev and Mamyrvaev,1982).

The present work was designed to study the influence of Vitamin D or Vitamin B_1 free diet on biochemical changes in serum of immature rats. The disturbances in total protein, albumin, calcium,calcium - binding protein, phosphorus and retardation of body weight were taken under consideration.

It is worthwhile to carry out the present investigation in order to reveal the differences between variable diet and different types of Vitamin D or Vitamin B_1 upon disturbances in total protein, albumin, calcium, calcium binding protein, phosphorus, phospholipids, total lipids and alkaline phosphatase activity in experimental animals.

inactive formsand Vitamin D_7 has about one-tenth the activity of Vitamin D_2 (Boer, et al ., 1939).

The elucidation of the chemistry of various Vitamin D and the photochemical changes proceeding during the ultraviolet irradiation of the Precursors were markedly hastened. It became evident that two forms of Vitamin D are of importance in human nutrition and medicine: Calciferol (Vitamin D_2), produced from irradiation of the plant sterol (ergosterol), and vitamin D_3, the activation product of the animal sterol 7-dehydrocholesterol (IUPAC, 1966).

2- Chemical structure of vitamin B_1

Williams., (1936) studied the chemistry of vitamin B_1 after purification and crystallization. He proved that vitamin contain a pyrimidine nucleus as well as thiazole ring. An important derivative of thiamine is the pyrophosphate. This molecule is known as co-carboxylase of the enzyme decarboxylase, which is involved in the decarboxylation of α -keto acids in the body. Structure of thiamin pyrophosphate is as illustrated in (Fig. 2). Thiaminokinase has been prepared from rat liver. This enzyme in the presence of ATP and Mg^{++} synthesizes Co-carboxylase from thiamin by transfer

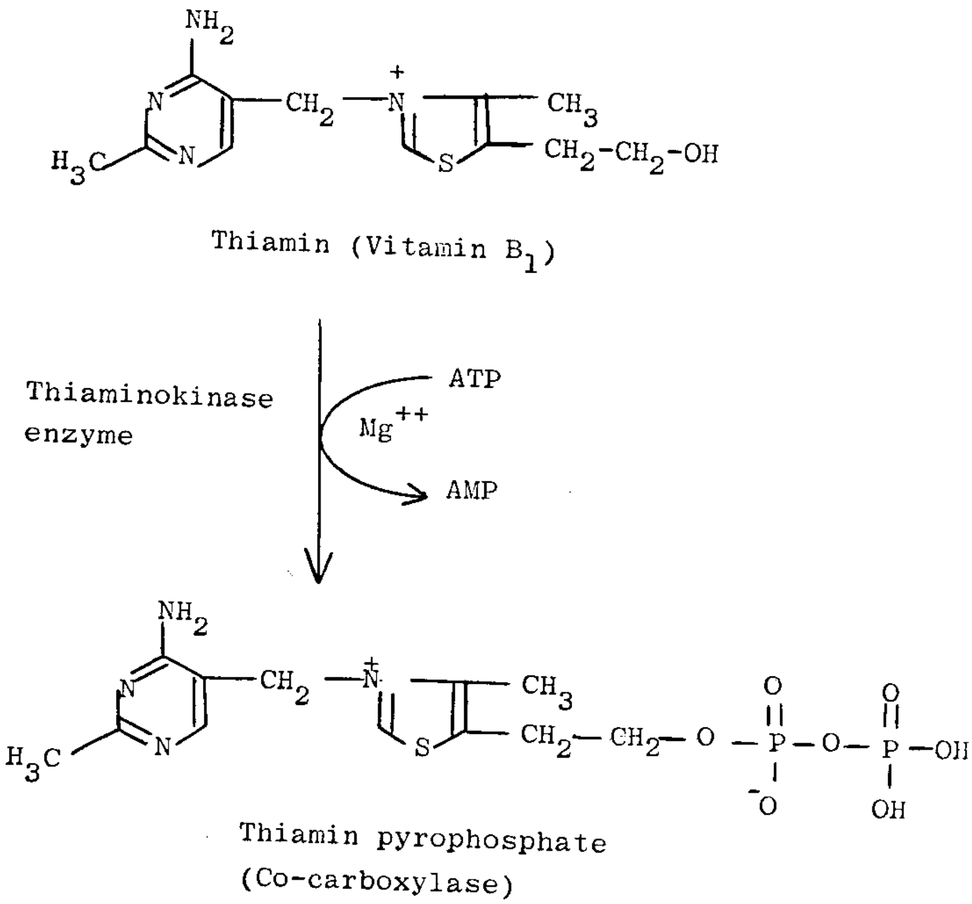

Fig. (2): Structural formules of thiamin and thiamin pyrophosphat (Co-Carboxylase).

of pyrophosphate from ATP to thiamin (Barron *et al*., 1941).

Sanemori, (1979), pointed out that, thiamin pyrophosphate was synthesized from hydroxymethylpyrimidine and hydroxyethylthiazole. Thiamin pyrophosphokinas catalyzed the synthesis of thiamin pyrophosphate from thiamin.

b- Natural Occurrence and Distribution of Vitamin D and Vitamin B_1

1- Natural Occurrence and Distribution of Vitamin D

Heymann, (1937) , studied the tissue distribution of vitamin D. He administered 5 mg of viosterol (vitamin D_2) preparation to rabbits and followed the vitamin activity in various tissues by the biological assay method. Some vitamin D activity was found in blood, liver and skin.

Warkany *et al*. , (1945), evaluated the concentration of vitamin D activity in human serum during and after periods of injection of high doses of vitamin D. Their results suggested the hypothesis that vitamin was stored in tissues and subsequently released quite slowly into the general circulation, where it was utilized to produce vitamin D- released biological responses.

Norman and Deluca (1964), reported that microsomes isolated from intestine and kidney contain the highest concentration of radioactivity, followed by mitochondria and the nuclear fraction, with the cytoplasmic fraction conntaining little or no radioactivity. These data were obtained 24 hours after an oral dose of 500 Iu of ^{3}H -vitamin D_3 to rats.

Neville and Deluca (1966), administered 10.7 IU of (1,2 ^{3}H) vitamin D_3 to rats maintained on vitamin D-deficient rations. When data were expressed in terms of the mass of each tissue, the following pattern of selective accumulation emerged at 24 hours post injection: bone cells > intestinal mucosa > kidney > Small intestinal Contents > blood > liver > muscles. These data correlate well with the known sites of vitamin D action on, bone, intestine, and kidney. Liver had a high relative uptake 0.5 hour after dosing. From these data it appears that, after absorption, the vitamin is first transported to the liver, where it may be altered before becoming located in target organs.

The same authors, orally administered (^{3}H)vitamin D_3 into rats. After the rats were killed and the tissue radioactivity was determined. One interesting aspect of those data was the rapid accumulation of vitamin D by liver (28.5 %), the bone (26.78 %), blood (12.92 %), the muscle (8.61 %), kidney (2.35 %), the small intestine (1.73 %), lung (1.72 %) and the spleen (1.04 %).

Nair and Bucana (1966), using gasliquid chromatography as the detection system, observed that the majority of vitamin D_2 and vitamin D_3 could be found in the cytoplasmic fraction of liver cells 15 minutes after an oral dose of 8000 IU to rats.

Imrie et al., (1967) , studied the tissue distribution of radioactivity and found a 10 IU dose of ^{3}H-vitamin D_3 in rachitic chicken was qualitatively similar to what had been abserved in rats. A high relative incorportion was observed in bone cells and intestinal mucosa. Vitamin D_2 had the same pattern of distribution as vitamin D_3 rachitic chicken.

Myrtle and Norman (1971), found 1,25 dihydroxyvitamin D_3 (the biologically active form of vitamin D_3) in the intestine.

2- Natural Occurrence and Distribution of vitamin B_1

It appears from the studies of various authors that vitamin B_1 is localized in several places in the