THE ROLE OF PYRIDOXINE AS A COUNTERMEASURE FOR IN-FLIGHT LOSS OF LEAN BODY MASS

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ABSTRACT

Ground based and inflight research has shown that humans, under conditions of microgravity, sustain a loss of lean body tissue (protein) and changes in several biological processes including, reductions in red blood cell mass, and neurotransmitters. The maintenance of muscle mass, the major component of lean body mass, is required to meet the needs of space station EVAs. Central to the biosynthesis of amino acids, the building blocks of protein, is pyridoxine (vitamin B-6). Muscle mass integrity requires the availability of vitamin B-6 for protein metabolism and neurotransmitter synthesis. Furthermore, the formation of red blood cells require pyridoxine as a cofactor in the biosynthesis of hemoglobin, a protein that carries oxygen to tissues. In its active form, pyridoxal-5'-phosphate (PLP), vitamin B-6 serves as a link between amino acid and carbohydrate metabolism through intermediates of glycolysis and the tricarboxylic acid cycle. In addition to its role in energy metabolism, PLP is involved in the biosynthesis of hemoglobin and neurotransmitter which are necessary for neurological functions. Alterations in pyridoxine metabolism may affect countermeasures designed to overcome some of these biochemical changes.

The focus of this research is to determine the effects of microgravity on the metabolic utilization of vitamin B-6, integrating nutrition as an integral component of the countermeasure (exercise) to maintain lean body mass and muscle strength. The objectives are: 1) to determine whether microgravity effects the metabolic utilization of pyridoxine and 2) to quantitate changes in B-6 vitamer distribution in tissue and excreta relative to loss of lean body tissue. The rational for this study encompasses the unique challenge to control biochemical mechanisms effected during space travel and the significance of pyridoxine to maintain and counter muscle integrity for EVA activities. This experiment will begin to elucidate the importance of biochemical interactions between micronutrients and the homeostasis condition of biological processes in the space environment.

To address this research topic a simulated microgravity model has been developed. The experiment uses radioisotopically labelled pyridoxine administered as an oral dose to rats which are maintained by tail suspension to simulate a microgravity environment. At the termination of the study, liver, muscle, blood and urine are collected and analyzed by reverse phase high pressure liquid chromatography to determine the quantity and distribution of the B-6 vitamers in tissue and excreta relative to lean body tissue loss. Earlier studies, published by this investigator, have shown that differences in vitamer distribution among samples from experimental verses control subjects indicate changes in metabolic utilization and storage of vitamin B-6.

INTRODUCTION

Ground based and inflight research has shown that humans, under conditions of microgravity, sustain a loss of lean body tissue (protein) and changes in several biological processes including, reductions in red blood cell mass, and neurotransmitters. The maintenance of muscle mass, the major component of lean body mass, is required to meet the needs of space station EVAs. Central to the biosynthesis of amino acids, the building blocks of protein, is pyridoxine (vitamin B-6). Muscle mass integrity requires the availability of vitamin B-6 for protein metabolism and neurotransmitter synthesis. Furthermore, the formation of red blood cells require pyridoxine as a cofactor in the biosynthesis of hemoglobin, a protein that carries oxygen to tissues. In its active form, pyridoxal-5'-phosphate (PLP), vitamin B-6 serves as a link between amino acid and carbohydrate metabolism through intermediates of glycolysis and the tricarboxylic acid cycle. In addition to its role in energy metabolism, PLP is involved in the biosynthesis of hemoglobin and neurotransmitter which are necessary for neurological functions. Alterations in pyridoxine metabolism may affect countermeasures designed to overcome some of these biochemical changes.

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MATERIALS AND METHODS

To address this research topic a simulated microgravity model has been developed. The experiment uses radioisotopically labelled pyridoxine administered as an oral dose to rats which are maintained by tail suspension to simulate a microgravity environment. At the termination of the study, liver, muscle, blood and urine are collected and analyzed by reverse phase high pressure liquid chromatography to determine the quantity and distribution of the B-6 vitamers in tissue and excreta relative to lean body tissue loss. Earlier studies, published by this investigator, have shown that differences in vitamer distribution among samples from

experimental verses control subjects indicate changes in metabolic utilization and storage of vitamin B-6. The chromatographic methodologies are being adapted to the HPLC equipment at NASA. All of the above analytical methodologies are being performed on a Hewlett-Packard HPLC with a loop injection valve, a fluorometric detector, and an electronic integrator. Excitation and emission wavelength are 295 nm and 405 nm respectively. Two mobile phases are being employed in a gradient elution procedure using a Bondapak IP column. Mobile phase A contains 0.033 mol/l phosphoric acid, and 8 mmol/l 1-octanesulfonic acid, adjusted to pH 2.2. Mobile phase B contains 0.033 mol/l phosphoric acid and 3.4 mol/l acetonitrile, adjusted to pH 2.2 and no ion pairing reagent.

RESULTS

The results of this study will determine the effects of microgravity on the metabolic utilization of pyridoxine and establish the requirement for vitamin B-6 during space travel. Alterations in pyridoxine metabolism may effect vitamin B-6 nutriture and the maintenance of lean body mass and muscle strength, which are necessary components for EVA activities. Statistical analysis on the distribution of vitamin B-6 vitamers between groups will be compared by the method of least squares ANOVA using general linear model procedures SAS, following a logarithmic transformation of the data to normalize the variance if necessary. Data will be reported as least squares mean +/- the pooled standard error of the least squares mean (SEM). Orthogonal contrasts will be made to examine the linear and quadratic effects of different independent variables on dependent variables.

DISCUSSION

The bioavailability of vitamin B-6 vitamers is a function of their extent of absorption and metabolic conversion to active coenzymatic forms. Pyridoxine that is absorbed from the intestinal tract is concentrated initially in the liver and is the sole source of plasma PLP. Thus, liver plays a central role in the overall metabolism of vitamin B-6. The major transformations in hepatic vitamin B-6 metabolism involve phosphorylation catalyzed by pyridoxal kinase, oxidation of PNP and PMP by pyridoxine (pyridoxamine) 5'-phosphate oxidase, along with interconversion of PLP and PMP through transamination reactions. The principal forms of vitamin B-6 in liver are PLP and PMP. Pyridoxine 5'-phosphate is usually present in only trace quantities because of its rapid oxidation to PLP. The non-phosphorylated B-6 vitamers constitute <10% of the total vitamin B-6 content in the liver. The metabolic pathway for the degradation of PLP involves enzymatic hydrolysis of the phosphate ester bond, and the oxidation of pyridoxal to 4PA. As a terminal product of vitamin B-6 metabolism, urinary 4PA reflects the in vivo metabolic utillization of the vitamin.