

2100

A HUMAN MODEL FOR THE STUDY OF THE METABOLISM OF BETA-CAROTENE(BC). S. Moberg, P. Rosen, E. Kaiser, M. Sapuntzakis, H. Friedman, Univ. of Illinois at Chicago, Chicago, IL 60680.

We adequate human or animal model for the study of metabolic effects of BC independent from its provitamin A activity is available. Model requirements are adequate nutritional status, depleted body stores of carotenoids, and a nutritionally adequate and controllable BC-free diet. We postulated that commercial enteral liquid formulas would be BC-free and patients(PTS) receiving long term enteral feedings would have depleted stores of BC. We measured the BC and retinol(RE) levels of Omnilite EN, Criticare NK, Vital EN, Omnilite, Meritene(vanilla supreme), Ensure (strawberry), Ensure EN(vanilla), and Ensure Plus(chocolate) by HPLC separation. No formula contained  $\beta$ -carotene, lycopene or  $\beta$ -cryptoxanthin in measurable amounts. Meritene and Ensure Plus(chocolate) contained BC in trace amounts, 30 ng/ml and 10 ng/ml, respectively, while the others contained no detectable BC. All formulas contained adequate RE levels. Fasting serum from 10 PTS with dysphagia secondary to CNS disorders, normal GI function and receiving enteral feedings for 3-5 weeks were analyzed for RE and BC. Mean serum RE and BC levels were  $41.4 \pm 17.3$  ug/dl and  $2.0 \pm 3.7$  ug/dl, respectively. Four PTS had no detectable BC and three had levels  $< 1.0$  ug/dl. The normal range for RE is 20-70 ug/dl while the reported range for BC is 15-100 ug/dl (mean 38 ug/dl). This PTS population satisfied the model criteria.

2102

WOUND HEALING IN RATS FED  $\beta$ -CAROTENE SUPPLEMENTS. Ahmad Wahid and Leonard E. Gerber, (SPON: A.G. Rand) Department of Food Science and Nutrition, University of Rhode Island, Kingston, Rhode Island 02881.

The effects of the level of dietary protein and  $\beta$ -carotene consumption upon skin wound healing were investigated using young adult male Sprague-Dawley rats. For the 13 days prior to surgery, five different diets were fed to five different groups of rats. Specifically, the diets included combinations of 5% or 20% casein plus 1.3 ug retinyl acetate per gram of diet with or without 28.8 ug  $\beta$ -carotene per gram of diet. One group was maintained on a vitamin A-free diet. Post-surgically all rats were fed a 20% casein diet, incorporating 1.3 ug retinyl acetate per gram of diet for 5 days. After 5 days, all rats were killed, wound tensile strength measured and liver and blood serum removed for retinoid analysis. Rats fed the higher level of protein with  $\beta$ -carotene supplementation were observed to have the greatest wound tensile strength, while rats fed the high level of protein but not receiving  $\beta$ -carotene had lower levels of recovery. Rats fed the low protein diet had lower tensile strength values than any of the higher protein groups, while  $\beta$ -carotene supplementation for these rats did not improve strength compared to rats fed only retinyl acetate. Serum and liver analysis for retinol and its esters indicated that in those rats fed 20% casein,  $\beta$ -carotene feeding elevated the serum and liver levels compared to rats fed only retinyl acetate, while in rats fed low protein, no such elevation was noted.

2104

EFFECTS OF RETINOIC ACID ON THE METABOLISM OF RADIOACTIVE VITAMIN A IN TISSUES OF RATS MAINTAINED ON VITAMIN A DEFICIENT DIET. Pangala V. Unat and Andre Lacroix, Clinical Research Institute of Montreal, (Quebec) H2W 1H7.

The effect of feeding retinoic acid (RA) for 2 and 6 days on the metabolism of labelled retinol in tissues of rats maintained on vitamin A deficient diet was studied. The metabolites of retinol were analyzed by high performance liquid chromatography. Feeding RA for 2 days markedly reduced the blood retinol and retinyl esters (RE) levels without affecting the vitamin A content of the liver. In intestine and testis the formation of labelled RA from labelled retinol was completely inhibited by dietary RA. Further inclusion of RA in the diet for 6 days resulted, in addition to decreased blood retinol and RE values to an increase in the RE values in the liver. The accumulation of RE in the RA fed rat liver was a direct block in the metabolic conversion of retinol to RA as RA could not be detected in these livers. No significant effect of dietary RA on the levels of RA, retinol and RE was found in the kidney tissue during the experimental period. These data provide direct evidence that RA is in the metabolic pathway of retinol, under physiological conditions and suggest that the signal to reduce the blood retinol levels in the presence of dietary RA is generated in the peripheral tissues.

2101

COMPARISON OF SERUM  $\beta$ -CAROTENE REPLETION WITH TWO LEVELS OF CARROTS AND AN ALGAE-DERIVED PRODUCT IN HEALTHY ADULT HUMANS. T. S. Pattison, G. A. Spiller, C. D. Jensen, J. M. Whittam and J. Seala, Shaklee Research Center, Hayward, CA 94545.

In view of the recent interest in  $\beta$ -carotene (BC) 30 healthy adults were placed on a low carotene diet (10 days), then randomized into 5 groups and placed for 7 days on: ALG24 = 24 mg/day BC from the alga *Dunaliella salina* (DS); CAR24 = 24 mg/day BC from raw carrots (RC); ALG8 = 8 mg/day BC from DS; CAR8 = 8 mg/day BC from RC. Another group (PLAC) was given a placebo. After 7 days on treatment, all subjects were placed on placebo for 7 days. Fasting serum  $\beta$ -carotene levels (SBC) were measured at end of depletion (day 10), on days 14 and 17 (days 4 and 7 of treatment) and on day 24 (end of post-treatment depletion). SBC mean  $\pm$  SD values in mcg/dl were:

	day 10	day 14	day 17	day 24
ALG24	11.0 $\pm$ 2.4(a)	32.9 $\pm$ 5.9(a)	31.3 $\pm$ 9.4(a)	21.6 $\pm$ 3.5(a)
CAR24	11.7 $\pm$ 2.1(a)	31.3 $\pm$ 5.2(a)	38.2 $\pm$ 5.0(a,b)	19.7 $\pm$ 3.2(a,b)
ALG8	13.1 $\pm$ 3.2(a)	25.8 $\pm$ 5.4(a,b)	26.4 $\pm$ 6.3(b,c)	16.0 $\pm$ 3.8(a,b)
CAR8	13.1 $\pm$ 4.3(a)	23.3 $\pm$ 4.7(a,b)	18.5 $\pm$ 4.1(b,c)	13.2 $\pm$ 3.2(b,c)
PLAC	10.7 $\pm$ 2.5(a)	11.7 $\pm$ 3.2(b)	9.9 $\pm$ 2.5(c,d)	7.0 $\pm$ 1.9(c)

In each column, means without a common letter are different ( $p < 0.05$ ). At end of treatment (day 17), combined values for SBC for ALG24 and ALG8 were higher ( $p < 0.05$ ) than for the combined CAR24 and CAR8. 8 mg/day BC from either DS or RC led to higher SBC per mg BC consumed than 24 mg/day from the same sources. DS extracts seem to be a good source of BC.

2103

HEPATIC SUBCELLULAR DISTRIBUTION OF  $\beta$ -CAROTENE, RETINOL, AND  $\alpha$ -TOCOPHEROL IN RAT AND CHICK. S. Taylor and R. Parker, (SPON: G. Combs), Cornell University, DIV. OF NUTRITION SCIENCES, Ithaca, NY, 14853.

Young male S-D rats and male White Leghorn chicks were fed 0.1 and 0.05 percent  $\beta$ -carotene (BC), respectively, as stabilized beadlets in the diet for six weeks. Subcellular fractions were prepared from perfused livers by differential centrifugation and purity assessed by marker enzymes. Concentrations of BC, retinol (A), and  $\alpha$ -tocopherol (E) were determined by reverse phase HPLC. Both rats and chicks fed BC had elevated total liver A and E. In rat liver the relative BC concentration in subcellular fractions was lysosomes > purified mitochondria, microsomes > purified nuclei, on a phospholipid basis. Dietary BC increased the mean rat liver mitochondrial E three-fold and A five-fold when compared to controls, with no effect in lysosomes. A positive correlation was seen between total liver BC and total liver E. In chick liver the relative BC concentration in subcellular fractions was purified mitochondria > lysosomes > microsomes > nuclei, on a phospholipid basis. Dietary BC increased the mean mitochondrial and lysosomal E and A content four fold relative to controls. Thus in both species dietary BC was nonuniformly associated with several hepatic subcellular fractions, and BC distribution was not a function of phospholipid content. Dietary BC was also shown to influence A and E content in total liver and certain subcellular fractions. (NIH CA 33638).

2105

AN IMPROVED GC/MS TECHNIQUE FOR ANALYSIS OF VITAMIN A. A.J. Clifford, Y. Tondeur, R. Lautamo, W. Jennings, A.D. Jones, H.C. Furr, J.A. Olson, and L.H. De Luca, U. of Calif., Davis, CA 95616, PRI-NCI, Frederick, MD 21701, J&W Scientific, Rancho Cordova, CA 95670, Iowa State U., Ames, IA 50011, and NCI-NIH, Bethesda, MD 20205.

Present stable isotope dilution techniques, using deuterated retinol ( $^2$ H-ROH) and gas chromatography (GC)/mass spectrometry (MS) to assess body stores of vitamin A, lack desirable sensitivity because intact retinol (ROH), due to its thermal instability and ease of isomerization is destroyed (dehydrated and isomerized) during the derivatization and GC. This report describes a GC/MS procedure where picogram quantities of intact retinol can be measured directly without derivatization or loss. ROH was directly injected on a DB-1 column (15 m x 0.25 mm ID, 0.1  $\mu$ m thick film) and one minute later the oven temperature was ballistically brought to 150°C, held for 2 min. at this temperature, and then increased to 250°C at 10°C/min. Sample loading was on column at room temperature to prevent degradation, the carrier gas was He and the solvent was THF. Isomeric ROHs were separated during chromatography from the all trans isomer and both were eluted in about 8 min. at 210°C. MS was performed with electron ionization at 70 eV and selected ion monitoring at high resolution was performed on the molecular ion  $m/z$  286 of ROH. Using this GC/MS procedure ROH gave a molecular ion  $m/z$  286 with a relative abundance in excess of 90% and a sensitivity of detection of about 100 picograms.

# FEDERATION PROCEEDINGS

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## Abstracts

Abstracts 1-2720

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