DIETARY LINOLEIC ACID SUPPRESSES GENE EXPRESSION OF RAT LIVER α-AMINO-β-CARBOXYMUCONATE-ε-SEMIALDEHYDE DECARBOXYLASE (ACMSD) AND INCREASES QUINOLINIC ACID IN SERUM

Yukari Egashira, Makiko Sato, Atsushi Tanabe, Kuniaki Saito*, Suwako Fujigaki* and Hiroo Sanada#

ABSTRACT

Hepatic ACMSD [EC4.1.1.45] plays a key role in regulating NAD biosynthesis from tryptophan. We previously reported that ingestion of polyunsaturated fatty acids by rats leads to a decrease in their hepatic ACMSD activity. We purified ACMSD and cloned cDNA encoding rat ACMSD. Therefore, in this study, we examined whether dietary linoleic acid altered ACMSD gene expression and its protein level. Moreover we measured the tryptophan catabolite quinolinic acid level in rats.

In the rats fed with linoleic acid, ACMSD mRNA and its protein levels in the liver were strongly suppressed and serum quinolinic acid was significantly increased as compared with the rats fed on a fat-free diet. These results suggest that the transcription level of ACMSD is modulated by linoleic acids or their metabolites and probably there is an inverse relationship between ACMSD activity and the production of quinolinic acid converted from tryptophan.

1. INTRODUCTION

Hepatic ACMSD [EC4.1.1.45] plays a key role in regulating NAD biosynthesis from tryptophan. It has been reported that there is an inverse relationship between ACMSD activity and the production of NAD converted from tryptophan. ACMSD activity is

Laboratory of Food and Nutrition, Department of Bioproduction Science, Chiba University, Matsudo, Chiba 271-8510, Japan; Tel: +81-47-308-8861; Fax:+81-47-308-8720: e-mail: egashira@midori.chiba-u.ac.jp
*Department of Laboratory Medicine, Gifu University School of Medicine, 40 Tsukasa-machi, Gifu 500-8705, Japan

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greatly affected by many factors such as nutrients, hormones and diseases[1-5]. The tryptophan catabolite quinolinic acid has been reported to be associated with the pathogenesis of certain neurodegenerative diseases, since it acts as an excitotoxic agonist of the N-methyl-D-aspartate receptor. ACMSD was postulated to affect the production of quinolinate[6]. We previously reported that ingestion of polyunsaturated fatty acids by rats leads to a decrease in their hepatic ACMSD activity[7]. We purified ACMSD and cloned cDNA encoding rat ACMSD [8,9]. In this study, we examined whether dietary linoleic acid altered ACMSD gene expression, its protein level and quinolinic acid concentration in rat.

2. RESULTS AND DISCUSSION

We investigated the effect of short-chain, middle-chain, and long-chain fatty acids on the activity of rat liver ACMSD[7]. When diets containing 2%, 5%, and 10% levels of fatty acids were given to rats for a week, saturated fatty acids and elaidic acid (trans form) did not suppress the ACMSD activity in liver. But polyunsaturated fatty acids such as linoleic acid, linolenic acid, eicosapentaenoic acid, and docosahexaenoic acid strongly suppressed the liver ACMSD activity. Five % sorbic acid and oleic acid tended to suppress the liver ACMSD activity weakly (Fig.1).

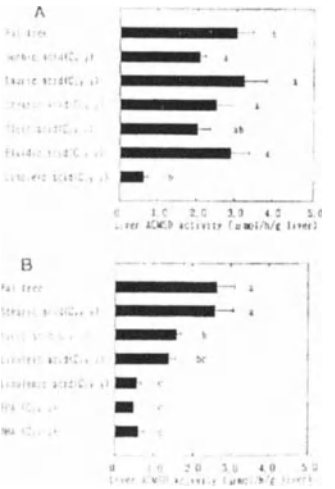

Figure 1. Comparison among the effects of various kind polyunsaturated fatty acids in diets on the liver ACMSD activity. A:The rats were fed on the diets containing 5% level of fatty acids. B: The rats were fed on the diets containing 2% level of fatty acids. Each bar represents mean±SE of 6 rats. Values without a common superscript letter are significantly different at $p<0.05$.

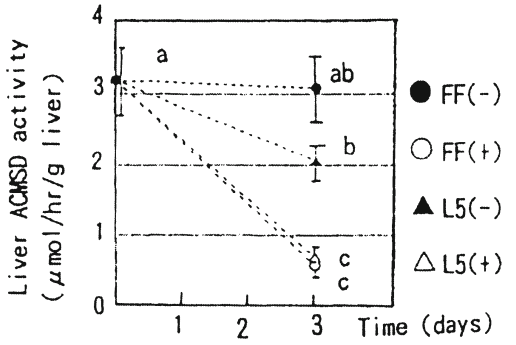

Figure 2. Effect of linoleic acid on the ACMSD activity in puromycin-treated rats.
The rats were fed on a fat free diet or 5% linoleic acid diet for 3 days, and injected with 0.5ml of 0.14M NaCl or 0.5ml of the same NaCl solution containing neutralized puromycin hydrochloride intraperitoneally at 08:30 and again at 20:30 during 3 days. Values without a common superscript letter are significantly different at $p<0.05$. FF(-),fat free(no puromycin): FF(+),fat free(+ puromycin): L5(-),linoleic acid (5%)(no puromycin): L5(+),linoleic acid (5%)(+ puromycin)

Next, we attempted to clarify whether linoleic acid would inhibit ACMSD synthesis or enhance the enzyme degradation or inactivation[10]. Puromycin suppressed ACMSD activity, but no additional suppression of hepatic ACMSD was observed by dietary linoleic acid. If linoleic acid had accelerated ACMSD degradation and inactivation, dietary linoleic acid should have reduced the hepatic ACMSD activity in the puromycin-treated rats less than that in the puromycin-treated rats without dietary fat. Therefore, the present results indicate two possibilities: one is that linoleic acid might not promote ACMSD degradation or inactivation, and the other is that dietary linoleic acid might influence the putative protein which acts to influence ACMSD turnover (Fig.2).

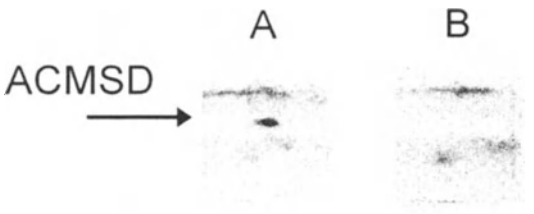

Figure 3. Immunodetection of rat liver ACMSD.
Cytosolic fractions were subjected to 2D-PAGE and transferred to PVDF membrane. A: fat free group, B: 20% linoleic acid group.

We purified ACMSD to homogeneous state and prepared the antibody against rat liver ACMSD by injecting mice with the purified enzyme[8,9]. With the use of this polyclonal antibody and analysis by two-dimensional electrophoresis, we studied the mechanism by which the level of liver ACMSD activity was varied in rats fed a linoleic acid diet [11]. In the rats fed a dietary linoleic acid, ACMSD protein levels in the liver were strongly suppressed as compared with the rats fed a fat-free diet (Fig.3). The result of this experiment suggest that the linoleic acid suppresses ACMSD synthesis.

We cloned the cDNA encoding rat ACMSD[9]. We mesured ACMSD mRNA levels of rats fed with diet containing linoleic acid with Northern blott analysis. When the Sprague-Dawley strain rats were fed with diets containing 0-20% linoleic acid for 8 days, ACMSD activity was decreased in the linoleic acid groups according to the levels of linoleic acid administered. In the rats fed with linoleic acid, ACMSD mRNA levels in the liver were strongly suppressed and serum quinolinic acid was significantly increased as compared with the rats fed a fat-free diet. These results suggest that the transcription level of ACMSD is modulated by linoleic acid or their metabolites and probably there is an inverse relationship between ACMSD activity and the production of quinolinic acid converted from tryptophan.

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