Turkish Journal of Endocrinology and Metabolism, (2005) 1 : 13-16 **ORIGINAL ARTICLE**

The Effect of L-Carnitine Treatment on Lactic Acid Levels in Normal Subjects and Patients with IGT

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ABSTARCT Principles: Previous studies have demonstrated that L-carnitine has important effects on the mitochondrial beta oxidation of long-chain fatty acids and glucose metabolism. In the present study, effects of the L-carnitine on lactic acid levels and glucose during the oral glucose tolerance test were investigated. Methods: The blood was collected at the initial, first and second hours during the oral glucose tolerance test. The oral glucose tolerance test was repeated after oral carnitine therapy (3 gr/day) for seven days. According to the ADA criteria ten subjects (7 women / 3 men) had normal glucose tolerance (NGT) and eight subjects (4 women / 4 men) had impaired glucose tolerance (IGT). Results: At the baseline, first hour and second hour plasma lactic acid levels showed difference between the NGT and IGT subjects before the carnitine therapy but this difference was not statistically significant. During the glucose loading, plasma lactic acid levels were increased at the first hour and returned to initial levels at the second hour in all subjects both before and after carnitine therapy. Carnitine therapy decreased the lactic acid levels in both groups in the all samples. In NGT group, there was significant decrease in lactic acid levels at the baseline and first hour; P< 0.05, <0.05. In IGT group, also, significant decrease in lactic acid levels at the first and second hour was observed; P<0.05, <0.05. Conclusions : Effect of the carnitine on the lactic acid levels was higher in the subject with IGT than the NGT group at the baseline and first hour during the oral glucose tolerance test. Reduced plasma levels of lactic acid levels following L-carnitine treatment may be related with both improved insulin resistance and increased oxidative glucose use by activating pyruvate dehydrogenase and decraesing intramithochondrial Acetyl CoA / CoA ratio.

Keywords: L-carnitine, Lactic Acid, IGT, Normal Glucose Tolerance

Introduction

L-carnitine (L-3-hydroxy-4-N- trimethylamino-butyric acid) is synthesized mainly in the liver from lysine molecules. Carnitine is rapidly transferred via the plasma to the other tissues (1). L-carnitine is essential for the oxidation of long-chain fatty acids by mammalian tissues because it facilitates the entry of these substrats into the mitochondrial matrix, the sites of the enzymes of beta-oxidation (2). Free fatty acids and many organic acids can not be metabolized directly.

Spesific enzymes first convert them to fatty acyl-CoA compounds can not diffuse through the inner mitochondrial membrane, but are first converted to fatty acylcarnitine. Fatty acylcarnitine can cross the inner mitochondrial membrane under the action of specific translocase enzyme (3). In the mitochondrium, these carnitine derivatives are converted to fatty acyl-CoA. The fatty acyl-CoA molecule then enters the beta oxidation sequence, whereas the carnitin molecule is freed (4). Carnitine has an important regulating effect on the mitochondrial processes in the hypoxic status. Carnitine increases the action of pyruvate dehydrogenase enzyme and oxidative metabolism of glucose (5). During the oral glucose tolerance test, plasma glucose, insulin and lactic acid levels are increased (6). The present study has investigated the effects of oral carnitine therapy for seven days on plasma glucose and lactic acid levels in subjects with normal glucose tolerance (NGT) and impaired glucose tolerance (IGT).

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Material and Methods

10 subjects with NGT and 8 subjects with IGT participated in the study. Both group were matched for age (39 ± 10 vs 45 ± 8 years), body mass index (29 ± 2,8 vs 30 ± 4 kg/m^2), gender ratio (7 women/3 men vs 4 women/4 men) and physical activity. None of them had a family history of diabetes mellitus, liver or kidney disease. All subjects were on a weight maintaining diet containing at least 250 g/day carbohydrate. After 12- hours fasting, subject were given the equivalent of 75 g anhydrous glucose dissolved in 250 ml water. Blood was drawn from antecubital vein to determine plasma glucose and lactic acid content at the baseline, first and second hours after glucose ingestion using vacutainer tubes. The oral glucose test was repeated after seven days oral carnitine therapy. Carnitine was obtained from Santa Farma Co, Turkey. (Carnitene tab. 1g) and was given orally 3 g/day. Plasma glucose and lactic acid were determined by the glucose oxidaze method (Sigma Chem. Cat. No 16-50 U.S.A.) and lactat oxidaze enzymatic method (Bio Merieux, Cat. No: 61192, France) respectively. American Diabetes Association criteria was used to identify whether subjects are NGT (2-hour glucose <140 mg/dl) or have IGT (2-hour glucose between 140-200 mg/dl). All statistical comparisons were made using paired two tailed t-test and unpaired two tailed t-test. Unpaired and paired t-test was also validated by the nonparametric Mann-Whitney U and Wilcoxon tests respectively. Analysis of variance (ANOVA) was used to compare multiple group means. All data are given as means ± standart deviation. Area under curve method was used to compare the total glucose levels variations before and after carnitine treatment during the oral glucose loading (7).

Results

Plasma lactic acid and glucose levels in NGT and IGT subjects during oral glucose tolerance test before and after the carnitine treatment are shown in the table 1 and 2. At the baseline, first and second hour, there were differences in plasma lactic acid levels between the NGT and IGT before the carnitine treatment but statistically not significant. During the oral glucose tolerance test, plasma lactic acid levels increased at the first hour and returned to initial levels at the second hour in all subjects both before and after the carnitine treatment. Lactic acid levels decreased significantly after the carnitine therapy at the baseline and first hour in NGT group (P<0.05, <0.05). In IGT group, lactic acid level significantly reduced at the first and second hour after carnitine therapy (P<0.05, <0.05). Lowering effect of carnitine treatment on lactic acid levels was more specific in IGT group than the normal group at the baseline, first and second hour. During the oral glucose tolerance test there were no significant differences in plasma glucose levels between the NGT and IGT subjects before and after the carnitine therapy according to area under curve method. The results are shown at the table 1 and 2

Table 1. The levels of plasma lactic acid before and after L-carnitine during the oral glucose tolerance test

	Groups	L. Acid (mg/dl) Basal	L. Acid (mg/dl) 1. hour	L. Acid (mg/dl) 2. hour
NGT	Before carnitine	16.01 ± 5.23	20.74 ± 6.40	17.71 ± 6.95
	After carnitine	$10.46 \pm 2.51_a$	$14.88 \pm 3.39_a$	12.63 ± 4.56
IGT	Before carnitine	19.79 ± 6.46	22.35 ± 7.01	19.07 ± 6.46
	After carnitine	11.50 ± 4.41	$12.10 \pm 5.49_b$	$9.84 \pm 5.63_b$

a: Differences in NGT group before and after carnitine.(p<0.05)
b. Diffrerences in IGT group before and after carnitine. (p<0.05)

Table 2. Plasma glucose levels in NGT and IGT during the oral glucose tolerance test before and after carnitine therapy.

Groups	Glucose (mg/dl), Before carnitine	Glucose (mg/dl), After carnitine
NGT	17571.42 ± 2951.34	17738.57 ± 1981.19
IGT	33744.00 ± 6085.46	33708.00 ± 5366.33

Discussion

L-carnitine promotes mitochondrial beta oxidation of long-chain fatty acids by facilitating their transfer across the inner mitochondrial membrane (8). During the mitochondrial beta oxidation, fatty acyl-CoA increase in the mitochondria. Fatty acyl-CoA is an important metabolic intermediate. Excessive levels of fatty acyl-CoA inhibite many enzymes including pyruvate dehydrogenase, and pyruvate carboxylase. Fatty acyl-CoA levels are modulated in several ways. First, some enzymes synthesizing them are inhibited by high levels of fatty acyl-CoA. Second, fatty acyl-CoA is metabolized further in energy-generating pathways. Finally, toxic fatty acyl-CoA can be converted to non-toxic

acylcarnitine and transferred to cytoplasma (3). These conditions can explain the effect of carnitine in the glucose and lactic acid metabolisms. Lactic acid, a metabolite of glycolysis, is synthesized from pyruvate by the lactic acid dehydrogenase and increases in the conditions where the pyruvate catabolism is blocked, like in poor controlled diabetes mellitus, IGT and hypoxic states. Glucose-induced metabolic imbalances that cause an increase in the reduced nicotinamid-adenine dinucleotid/ nicotinic acid dehydrogenase ratio (NADH/H) and lactic acid increasing are linked to imbalances in carnitine metabolism (9). In carnitine deficiency, fatty acyl-CoA metabolites inhibit the pyruvate dehydrogenase complex and pyruvate conversion in to acetyl-CoA. Carnitine deficiency stimulates pyruvate production from its two major sources. Enhanced glycolysis in liver increases pyruvate formation and the catabolism of amino acids in muscle contributes a substantial quantity of pyruvate and resulted lactic-pyruvic acidemia (3). Inokuchi et al (10) demonstrated that the plasma short-chain acylcarnitine concentration and acylcarnitine/free carnitine ratio were similar in the IGT and type 2 groups and significantly greater than those in the NGT group. Winter et al. (11) reported that acylcarnitine increases and free carnitine decreases in the type 1 diabetes. Bach et al. (12) showed that the plasma carnitine level was increased following 2 g of L-carnitine administration in healhty subjects. Our results are in accordance with previous reports showing that the plasma lactic acid level increases after the oral glucose loading. According to Siliprandi et al. (13) L-carnitine administration prior to intense exercise stimulates pyruvate dehidrogenase activity, thus decreases plasma pyruvate, lactic acid levels and at the same time induce the increase of acetyl-carnitin. On the contrary, Colombani et al. (14) demonstrated that acute administration of carnitine did not affect the metabolism in the athletes during the marathon running. However it is widely believed that carnitine administration decreases lactic acid levels in the hypoxic states. NADH/H ratio and lactic acid production are increased in subjects with diabetes mellitus and IGT when compare to healthy subjects (15). L-carnitine administration significantly decreases lactic acid levels in subject with diabetes mellitus and IGT. Our results demonstrated that carnitine treatment can be more effective in reducing lactic acid levels in IGT patients than NGT subjects. Abdel Kader et al. (16) reported that diabetic subjects treated with carnitine during oral glucose tolerance test showed delayed peak and decrease in plasma pyruvate levels. In our study, during the oral glucose loading, lactic acid levels were highest at the first hour in the all subjects and decreased at the second hour to basal levels. Probably, this result is due to lactic acid release from intestinal cells which produced lactic acid during the glucose absorption. Normally, intestinal cells use free fatty acids, glutamin and only limited amount of glucose for energy production.When the glucose content of intestine is increased, intestinal glucose metabolism is increased in order to provide energy fot the absorbtion of glucose and lactic acid release from the intestinal cells is increased (17). It has been reported that L-carnitine restored glycolysis and glucose oxidation in the heart of rats with carnitin deficiency (18). Negro et al. (19) reported that L-carnitine decreased high plasma glucose levels induced by glucose infusion in healthy people. L-carnitine treatment significantly reduced serum glucose, free fatty acids, triglyserides and ketones in diabetic rats. In nondiabetics rats, carnitine increased serum ketones while free fatty acids and triglserid decreased (20). But we did not find any significant difference in glucose levels between IGT and NGT groups during oral glucose tolerance test before and after carnitin therapy.

In a study evaluating the effect of parenteral administration of L-carnitine (either two or four grams per day) on metabolic parameters after surgical stress, Heller et al concluded that carnitine called prevent worseninig in insulin resistance (21). Gunal et al also reported a single intravenous dose of L-carnitine (one gram) had a positive effect on insulin sensitivity in patients with chronic renal failure (22). L-carnitine constant infusion improves insulin sensitivity in insulin resistant diabetic patients; a significant improvement in whole body insulin mediated glucose uptake is also observed in normal subject (23).

L-carnitine is an important drug in the energy metabolism. After administration of L-carnitine, reduced plasma levels of lactic acid suggest that this effect might be exerted through the activation of pyruvate dehydrogenase, which is depressed in the insulin resistance state. Addition of carnitine to the treatment of metabolic disorders, which cause

lactic acid elevation, may be helpful by this mechanism.

As a conclusion, reduced plasma lactic acid levels after L-carnitine treatment may be an indicator of improved insulin resistance which also increases oxidative glucose use by activating pyruvate dehyrogenase and decreasing intramitho-chondrial Acetyl CoA/CoA ratio (24) as pointed out in literature.

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