Effect of Magnesium Deficiency on Δ6 Desaturase Activity and Fatty Acid Composition of Rat Liver Microsomes

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Experimental Mg^{2+} deficiency was induced in a group of rats by feeding them a Mg^{2+}-deficient diet for 23 days. They were pair-fed to compare with a control group of rats fed a Mg^{2+}-sufficient diet. In the Mg^{2+}-deficient group the plasma total cholesterol and triglyceride levels were increased while HDL-cholesterol was decreased. In the Mg^{2+}-deficient group the plasma level of thiobarbituric acid reacting substances (TBARS) used as a measure for lipid peroxidation was increased. The increase was attributed to the increased cytosolic Ca^{2+} in Mg^{2+}-deficiency which can cause: 1) increase of hydro and endoperoxide levels as a consequence of the increase of arachidonic acid release and eicosanoid synthesis in Mg^{2+}-deficiency, and 2) inhibition of the mitochondrial respiratory activity and activation of Ca^{2+}-dependent proteases which may activate the conversion of xanthine dehydrogenase to xanthine oxidase which generates active O_2 species. In the Mg^{2+}-deficient group, the fatty acid composition of the liver microsomes indicated a slower rate of conversion of linoleic acid to arachidonic acid which was consistent with the decrease of Δ6 desaturase activity in liver microsomes of Mg^{2+}-deficient rats as measured *in vitro*. The decrease of Δ6 desaturase activity was attributed to the lower concentration of actual enzyme molecules as a result of the decreased rate of protein synthesis in Mg^{2+}-deficiency. The possible effects of the increased catecholamine release in Mg^{2+}-deficiency are discussed.
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Several studies provided ample evidence that Mg^{2+}-deficiency affects lipid metabolism. Mg^{2+}-deficiency produced hypercholesterolemia, hypertriglyceridemia and dyslipoproteinemia characterized by an increase of VLDL and LDL and a decrease of HDL (1). In Mg^{2+}-deficient rats an increase of plasma-free cholesterol and a decrease of esterified cholesterol as a result of the reduced lecithin-cholesterol acyltransferase activity (LCAT) were observed (1). Changes in plasma fatty acids were also reported in Mg^{2+}-deficient rats. These changes were characterized by a decrease of stearic and arachidonic acids and an increase of oleic and linoleic acids (2). They were related to the hypertriglyceridemia and the increase of VLDL and LDL and decrease of HDL since a notable alteration of fatty acids can occur as the density of the lipoprotein increases (2). In patients with latent tetany, a disease characterized by a magnesium deficiency (3), the plasma phospholipid fatty acids showed an increase in linoleic acid and a decrease in dihomo-(gamma)-linolenic (20:3ω6) and arachidonic (20:4ω6) acids (4).

Many cellular functions and responses are affected when the membrane lipid or fatty acid unsaturation is modified. These include carrier-mediated transport, membrane-bound enzymes and receptor properties (5). The present experiment was conducted to determine the origin of the fatty acid modification in Mg^{2+}-deficiency. We studied the effect of Mg^{2+}-deficiency on Δ6 desaturase, the enzyme which regulates the biosynthesis of essential polyunsaturated fatty acids (such as 20:4ω6 and 22:6ω3) derived from essential fatty acids. We also studied the changes in membrane fatty acids in Mg^{2+}-deficiency using liver microsomal membrane as a model.

MATERIALS AND METHODS

Chemicals and reagents. [1-^{14}C]Linoleic acid (sp. act. 58.7 mCi/mmol) was purchased from Amersham Corporation (Arlington Heights, IL); [1-^{14}C]linoleoyl-coenzyme A (sp. act. 54.4 mCi/mmol) was purchased from New England Nuclear (Boston, MA). Linoleoyl-coenzyme A, ATP, COA (lithium salt), NADH, bovine serum albumin V fraction (essentially free of fatty acid), thiobarbituric acid (TBA), 1,1,3,3-tetramethoxypropane, and the diagnostic kits for the enzymatic assay of total cholesterol, HDL-cholesterol, and triglycerides in plasma were purchased from Sigma Chemical Company (St. Louis, MO). Linoleic acid and methyl esters of linoleic and γ-linolenic acids were purchased from Nu-Chek Prep. Inc. (Elysian, MN).

Animals and diets. Male Sprague-Dawley weanling rats weighing about 30 grams were housed individually in suspended cages with wire-mesh bottoms. They were subjected to alternating 12 hr periods of light and darkness and were divided randomly to provide a control (12 animals) and an experimental magnesium-deficient group (30 animals). The rats were pair-fed fresh food daily with a modified AIN-76 semipurified diet (Table 1). The magnesium content determined by analysis was a 45 mg/kg diet for the deficient diet group and this was adjusted to a 960 mg/kg diet for the control group by addition of magnesium oxide. Distilled water was available *ad libitum*. The rats were fed their respective diets for 23 days before blood and tissue were collected.

Analysis. Blood was collected by cardiac puncture after ether anaesthesia at the end of the dark period after 12 hr fasting. A single blood sample was taken from each rat. Twelve rats from the control and 20 rats from the deficient groups were used. Plasma from heparinized blood was obtained by immediate centrifugation at 2000 *g* for 15 minutes at 4°C and then frozen immediately for subsequent lipid and mineral analyses.

The enzymatic methods for total cholesterol (6), HDL-cholesterol (7) and triglyceride (8) determination in plasma were used according to the instruction manuals accompanying the diagnostic kits obtained from

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Abbreviations: ATP, adenosine triphosphate; COA, coenzyme A; FFA, free fatty acid; HDL, high density lipoprotein; HCl, hydrochloric acid; LCAT, lecithin cholesterol acyltransferase activity; LDL, low density lipoprotein; MDA, malondialdehyde; NADH, nicotinamide adenine dinucleotide; TBARS, thiobarbituric acid reacting substances. TBA, thiobarbituric acid; VLDL, very low density protein.

TABLE 1

Composition of Semipurified Magnesium-Deficient Diet[a]

Component	gm/100 gm
Casein	20.0
DL-methionine	0.3
Corn starch	15.0
Sucrose	50.0
Fiber	5.0
Corn oil	5.0
Choline bitartrate	0.2
AIN mineral mix (omitting Mg^{2+})	3.5
AIN vitamin mix	1.0
Mg^{2+}	Varied

[a]Modified AIN-76 semipurified diet from which magnesium was omitted. The magnesium level of this diet was 45 mg magnesium/kg diet (deficient diet). The magnesium hydroxide was added to reach the level of 960 mg magnesium/kg diet which represents the control diet in this experiment. The modified AIN-76 semipurified diet from which magnesium omitted was obtained from ICN-Biochemical, Cleveland, OH.

Sigma Co. Lipid oxidation was estimated by assaying thiobarbituric acid reactive substances (TBARS) using the fluorometric method of Yagi (9). Fluorometric measurements were made at 553 nm with 515 nm excitation. The amount of malondialydehyde (MDA) equivalent was determined against a standard curve obtained by using freshly diluted 1,1,3,3-tetramethoxypropane. The degree of lipid oxidation was expressed in nmoles of MDA equivalent per 100 ml plasma.

Magnesium was estimated by atomic absorption flame spectrophotometry (Thermal Jarell Ash Video 12-E) and was carried out at the Atomic Absorption Center, Rodger Adams Laboratory, University of Illinois at Urbana. The plasma samples were diluted 1:50 with a $LaCl_3$ solution (0.1% lanthanum). Microsomes were dried at 105°C for 24 hr and ashed at 550°C for 24 hr. The ashed residue was dissolved in dilute HCl before dilution with the lanthanum solution.

Incubation conditions and assay of desaturase. For microsomal preparation the rats were killed by decapitation after which the abdomen was opened and livers removed. Livers were washed three times in fresh, ice-cold, homogenization solution and immediately processed to obtain the microsomal preparation. The livers of two rats from the same group were pooled to yield enough microsomal preparation for fatty acid analysis and desaturase assay. The rats were not fasted before being killed so that the maximal activities of the liver desaturase could be measured (10).

The liver microsomes were assayed for Δ6 desaturase activity as previously described (11). Each incubation in 1 ml of 0.15 M KCl-0.25 M sucrose solution contained (in μmoles): ATP, 5.0; CoA, 0.25; NADH, 1.0; $MgCl_2$, 5.0; glutathine, 1.5; NaF, 45.0; nicotinamide, 0.5; phosphate buffer (pH 7.0), 100.0; and 2 mg protein of a microsomal suspension. The microsomal protein concentration was measured according to Lowry *et al.* (12). The microsomes were separated by centrifugation at 105,000 × *g* for 2 hr (13). One hundred nmol (containing an amount of ^{14}C-labeled acid equivalent to 0.1 μCi) of linoleic acid in the form of the sodium salt bovine albumin complex (1 μg free fatty acid/11.5 μg bovine serum albumin) were used as substrate. Under these conditions, the enzyme was saturated by the substrate. The incubations were carried out for 20 min in a Dubnoff metabolic shaker at 37°C. The products of the reaction under the assay conditions as described were proportional to the 2 mg protein concentration and the 20 min reaction time.

The reaction was terminated by the addition of 5% HCl in methanol, and the lipids were extracted with chloroform/methanol (2:1, v/v). The extract was dried under a stream of N_2 and transesterified with 3 N methanolic-HCl (Supelco, Inc., Bellefonte, PA) at 70°C for 2 hr (14). The HCl-methanol was evaporated under N_2, and the methyl esters were dissolved in petroleum ether. A mixture of unlabeled carriers of methyl esters of 18:2 + 18:3 was added to the incubation products. The esters were separated on 10% $AgNO_3$ Silica Gel GHL plates (Analtech, Newark, DE) as previously described (15). The separated bands were scraped into scintillation vials, 10 ml of scintillation fluid (Beckman, Ready Solv) were added, and the activity was counted in a Beckman LS 3801 Scintillation Counter. The percentage of desaturation was calculated as the ratio of the counts in the desaturated products to the sum of the counts in the substrate plus product, corrected for background. The nmol of the product was then calculated. The recovery of the radioactivity was more than 85% of the amount used.

Extraction of lipids and analysis of fatty acids. In order to study the effect of magnesium deficiency on the fatty acid composition of the liver microsomal fraction, the microsomes were extracted by the method of Folch *et al.* (16). The methyl esters of fatty acids were prepared from the microsomal lipids following saponification and methylation (17). All operations were carried out under N_2. A Packard model 428 gas chromatograph (Hewlett-Packard Instrument Co., Inc., Chicago, IL), equipped with an all-glass injection splitter and flame ionization detector (FID), was used to separate the methyl esters on a Supelcowax-10, 30 m × 0.25 mm ID fused silica column. The oven temperature was programmed from 160–220° at 1°C/min; the injector and detector temperatures were 280° and 300°C, respectively. The N_2 flow rate was 0.7 ml/min with a split ratio of 110:1. Retention time, peak areas and peak relative area percentages were determined electronically using a Hewlett-Packard Model 3390 A Reporting Integrator. The mol% of fatty acids was then calculated. Identification of methyl esters of fatty acids was accomplished by comparing relative retention time with authentic standards (Nu-Chek Prep Inc.), Elysian, MN, and Supelco, Inc., Bellefonte, PA).

Statistical analysis. Results were expressed as means with their standard deviations. The statistical significance of differences between means was assessed by Student's t-test using group analysis.

RESULTS AND DISCUSSION

The effectiveness of the diet (Table 1) used to induce Mg^{2+} deficiency was clearly shown by the usual depression in plasma Mg^{2+} (Table 2) as well as the clinical manifestations of the syndrome (18). The Mg^{2+}-deficient diet resulted in a decreased efficiency of food utilization and a lower body weight of Mg^{2+}-deficient

TABLE 2

Magnesium Concentrations in Plasma and Liver Microsomal Fraction of Rats Fed on a Control or Magnesium-Deficient Diet[a]

	Control	Mg-deficient	P
Plasma (mg Mg^{2+}/L)	20.10 ± 1.56 (n=10)	4.60 ± 1.71 (n=19)	<0.001
Microsomes (μg Mg^{2+}/mg Protein)	521.00 ± 35.00 (n=5)	389.00 ± 29.00 (n=8)	<0.020

[a]Number in parenthesis represents the number of samples analyzed. In case of microsomes each sample represents microsomes obtained from two pooled livers. Values are given as means ± standard deviations. Statistical analysis was by Student t-test.

rats compared with pair-fed controls. Clinical symptoms of Mg^{2+} deficiency such as hyperaemia of the ears (manifested during the first two weeks), alopecia and hypoxia were also observed: Mg^{2+}-deficient rats were notably excitable. It was not possible to follow their weight increase after 10 days on the deficient diet because once they were touched or disturbed they started convulsing and died within one or two minutes. The mortality rate was 33% after 23 days on the Mg^{2+}-deficient diet.

In Mg^{2+}-deficient rats the decrease of Mg^{2+} level was more severe in plasma than in the microsomal fraction (Table 2). The decrease in plasma Mg^{2+} level agrees with previous reports (19,20). In a previous study (21) a decrease in Mg^{2+} concentration of the liver was reported while Mg^{2+} levels remained unchanged in the liver microsomes of Mg^{2+}-deficient rats. In the latter study, however, adult rats which weighed 100 g were used; while in the present experiment, we used weanling rats weighing only 30 g. This may indicate that the response of adult rats to Mg^{2+}-deficient diets may differ from the weanling rats which are more severely affected by Mg^{2+} deprivation as previously reported (1).

Plasma total cholesterol and triglyceride concentrations were significantly elevated in Mg^{2+}-deficient rats and the increase in triglycerides was much higher than the increase in total cholesterol (Table 3). The total cholesterol/HDL cholesterol ratio was significantly increased in Mg^{2+}-deficient rats, while HDL cholesterol was decreased which indicates that total cholesterol in the lower density lipoproteins such as VLDL and LDL was increased. The increase of plasma triglycerides and decrease of HDL-cholesterol in Mg^{2+}-deficient rats are in agreement with the previous reports (22,23). The significant increase of plasma total cholesterol of Mg^{2+}-deficient rats in our experiment is in agreement with that reported by Jaya and Kurup (22) while it disagrees with that reported by Rayssiguier and Gueux (24) who observed no change in total serum cholesterol of Mg^{2+}-deficient rats. This could be due to the short duration of their experiment which continued for only 8 days on the Mg^{2+}-deficient diet.

The elevated level of plasma-total cholesterol in Mg^{2+}-deficient rats was mainly attributed to: 1) increased activity of hepatic HMG-CoA reductase and the increased incorporation of [^{14}C]acetate into the cholesterol of liver and intestine (22) and 2) a decrease in the degradation of cholesterol to bile acids (22).

An elevated plasma triglyceride level in Mg^{2+}-deficient rats may arise either from an increased synthesis of triglycerides in the liver and increased secretion of VLDL particles, from a decreased removal of lipids from the blood, or from a combination of both. Previous studies (22,23) showed that the triglyceride accumulation in plasma of Mg^{2+}-deficient rats was mainly due to the decreased uptake of triglyceride lipoproteins from the circulation by extrahepatic tissues (heart and adipose tissues) because of the decrease in the lipoprotein lipase activity in these extrahepatic tissues of Mg^{2+}-deficient rats. After intravenous heparin administration, a significant reduction in plasma post-heparin lipolytic activity in Mg^{2+}-deficient rats was observed (1).

Another possible mechanism may involve hypomagnesaemia increasing the catecholamines released (25) with activation of adenylate cyclase through their β-adrenergic effect and enhancing the synthesis of cAMP. Elevated cAMP activates triglyceride lipase of adipose tissue, lipolysis is thus increased, and free fatty acids (FFA) are formed. The FFA enter the blood stream where they are transported to the liver and enhance lipogenesis (26,27). This mechanism is supported by the results of Itokawa *et al.* (28) who demonstrated an acceleration of the incorporation of [^{14}C]acetate into the hepatic lipids in Mg^{2+}-deficient rats. Thus, the increase in plasma cholesterol and triglycerides observed in Mg^{2+}-deficient rats may be the result of increased hepatic synthesis, increased release of lipoproteins into the circulation and their decreased uptake from circulation by the extrahepatic tissues.

The thiobarbituric acid reacting substances (TBARS) used as a measure for the lipid peroxide levels in plasma were significantly increased in Mg^{2+}-deficient rats (Table 3). It is not known if these lipid peroxidation products originated within the vascular compartment of the body or were released from other organs or tissues.

In magnesium deficiency, an increase in cell membrane permeability produces a decrease in cytosolic [K^+] and [Mg^{2+}] and an increase in cytosolic [Na^+] and [Ca^{2+}]. The increased cytosolic [Na^+] induces a release of mitochondrial Ca^{2+} (29,30) and a further elevation of cytosolic [Ca^{2+}]. The calcium-dependent activation of phospholipases with the subsequent release of free fatty acids from membrane phospholipids and activation of the arachidonic acid cascade then occur. This will result in an increase of eicosanoid synthesis as previously reported in different organs and tissues of Mg^{2+}-deficient rats (31). Therefore, higher levels of the

TABLE 3

Plasma Lipids and Lipid Peroxidation Levels in Rats Fed on a Control or Magnesium-Deficient Diet for 23 Days[a]

	Control	Mg-deficient	P
Total cholesterol (mg/100 ml)	125.20 ± 17.80 (9)	178.50 ± 23.44 (20)	<0.001
HDL-cholesterol (mg/100 ml)	51.20 ± 3.71 (8)	45.60 ± 6.24 (16)	<0.05
$\frac{\text{Total cholesterol}}{\text{HDL-cholesterol}}$ %	2.45 ± 0.20 (8)	3.96 ± 0.31 (16)	<0.001
Triglycerides (mg/100 ml)	46.00 ± 3.80 (9)	189.20 ± 50.50 (17)	<0.001
TBARS (nmol MDA/100 ml)	559.00 ± 52.00 (9)	935.00 ± 120.00 (20)	<0.001

[a]Number in parenthesis indicates number of animals used for each assay. Values are given as means ± standard deviations. Statistical analysis was by Student t-test.

hydro-, and endoperoxides should be produced from arachidonic acid through action of lipoxygenase and cyclooxygenase within the tissues of Mg^{2+}-deficient rats. These lipid peroxides are both intermediates in and regulators of prostaglandin synthesis: they have a short lifetime. The increased levels of these peroxides above a certain level within the cell may have an initiating effect on lipid peroxidation if the glutathione peroxidase and glutathione transferase enzyme levels are not enough to protect against their peroxidative effect. The increase of intracellular $[Ca^{2+}]_i$ also may inhibit the mitochondrial respiratory activity and activate Ca^{2+}-dependent proteases which may catalyze conversion of xanthine dehydrogenase to xanthine oxidase (32) and lead to the increase of lipid peroxidation through the generation of active O_2 species. A similar effect can be produced also as a result of the stress accompanying Mg^{2+} deficiency. This stress can increase xanthine and hypoxanthine concentrations within the cells which result in a greater production of H_2O_2 and O_2 (33).

In comparison to the control group, a significant decrease in microsomal arachidonic (20:4ω6) and 22:4ω6 acids and an increase in 20:3ω6 and 18:2ω6 acids were noted in the Mg^{2+}-deficient rats (Table 4). These changes are not fully in agreement with the changes previously reported in other studies (34,35) regarding the effect of Mg^{2+} deficiency on fatty acid composition. These studies (34,35) reported an increase of arachidonic acid in the triglyceride fraction and an increase of 22:4ω6 and 22:5ω3 acids in the triglycerides and phospholipid fraction of the arterial bed of the magnesium-deficient rats as compared to the controls. These discrepancies between our results and the previously reported results (34,35) could be attributed to the difference in the degree of Mg^{2+} deficiency. In the present study, Mg-deficient rats were fed for only 23 days a diet which contained 45 mg Mg/kg diet; while in the other studies (34,35), the rats were fed a diet containing 120 mg Mg^{2+}/Kg diet for 14 weeks.

The increase of 18:2ω6 and the decrease of 20:4ω6 in Mg^{2+}-deficient rats may possibly indicate a decrease in the rate of conversion of 18:2ω6 to 20:4ω6. Another possible reason for the decrease of arachidonic acid could be the increased rate of eicosanoid synthesis in the tissues and organs of Mg^{2+}-deficient rats (31) which can increase 20:4ω6 utilization in these tissues. In addition, the increased rate of lipid peroxidation in Mg deficiency could contribute to the decrease of 20:4ω6 acid since it is one of the highly susceptible acids to oxidation.

Since Δ6 desaturase is the key enzyme for the conversion of 18:2ω6 acid to any of ω6-metabolites, the activity of this enzyme then can be measured by the ratio of total ω6 metabolites total ω6 acids (36). From Table 4 it is clear that this ratio was significantly decreased in Mg^{2+}-deficient rats. These changes in fatty acids are consistent with the lower Δ6 desaturase activity of liver microsomes derived from Mg^{2+}-deficient rats, as measured *in vitro* (Table 5).

The true substrates for the desaturase enzymes *in vivo* are the Coenzyme-A (CoA) esters of fatty acids which require Mg^{2+}, ATP and CoA for their formation. A low cellular Mg^{2+} level could decrease the cellular ATP level (37,38) or directly affect the rate of Acyl-CoA formation. This activation step may become a rate-limiting step in the fatty acid desaturation reactions in Mg^{2+} deficiency which could explain the lower conversion rate of 18:2ω6 to 20:4ω6 in the liver microsomes of Mg^{2+}-deficient rats *in vivo*. However, when the liver microsomes of Mg^{2+}-deficient rats were incubated *in vitro* with 18:2ω6 acid as substrate in the presence of $MgCl_2$, CoA and ATP as cofactors, the Δ6 desaturase activity was not restored but remained significantly lower than that of controls (Table 5). Similarly, when linoleoyl-CoA was used as substrate instead of linoleic acid the Δ6 desaturase activity also remained lower in Mg^{2+}-deficient rats than in the controls. These results indicate that the activation step of the fatty acids to their CoA-ester is not the contributing factor for the lower microsomal Δ6 desaturase activity in Mg^{2+} deficiency. It is possible that Mg^{2+} deficiency affects the concentration of actual enzyme molecules since a decreased rate of liver protein synthesis and hypoproteinemia are early symptoms of Mg^{2+} deficiency (39).

As previously indicated a reduced plasma Mg^{2+} concentration released increased catecholamines (40). In previous studies it was shown that epinephrine regulates the biosynthesis of polyunsaturated fatty acids in rat liver microsomes (41). A single dose of this hormone (1 mg/kg body wt) administered to normal

TABLE 4

Fatty Acid Composition of Liver Microsomal Lipids of Rats Fed on a Control or Magnesium-Deficient Diet[a]

Fatty acid	Control (n=6) mol%	Mg^{2+}-deficient (n=6) mol %	P
14:0	1.00 ± 0.2	0.96 ± 0.2	N.S.[b]
16:0	18.30 ± 2.1	18.24 ± 0.7	N.S.
16:1	3.00 ± 0.7	3.20 ± 0.2	N.S.
18:0	6.80 ± 0.9	5.40 ± 0.6	<0.05
18:1ω9	8.10 ± 1.3	6.70 ± 0.5	N.S.
18:1ω7	2.00 ± 0.2	2.26 ± 0.3	N.S.
18:2ω6	16.40 ± 0.7	20.44 ± 1.3	<0.01
18:3ω6	0.60 ± 0.1	0.52 ± 0.1	N.S.
20:3ω6	0.80 ± 0.3	1.20 ± 0.2	<0.02
20:4ω6	31.60 ± 1.9	28.48 ± 0.9	<0.02
22:4ω6	1.20 ± 0.2	0.85 ± 0.1	<0.05
22:5ω6	3.30 ± 0.3	3.20 ± 0.6	N.S.
22:5ω3	1.10 ± 0.2	0.86 ± 0.2	N.S.
22:6ω3	6.20 ± 0.7	5.48 ± 0.8	N.S.
20:4ω6 / 18:2ω6	1.93 ± 0.1	1.40 ± 0.1	<0.001
Total ω6 acids	53.88 ± 1.7	55.08 ± 1.4	N.S.
Total ω6 metabolites	37.51 ± 1.3	34.64 ± 0.5	<0.01
ω6 Metabolites[c] / ω6 Acids	0.70	0.63	<0.001

[a]Results are expressed as means ± standard deviations. Statistical significant differences were calculated by Student t-test.

[b]N.S. - not significant.

[c]ω6 Metabolites are total ω6 acids other than 18:2ω6 acid.

TABLE 5

Liver Microsomal Δ6 Desaturase Activity in Rats Fed on a Control or Magnesium-Deficient Diet[a]

Substrate	Controls (n=6)	Mg-deficient (n=8)	P
Linoleic acid +Cofactors nmol 18:3 produced/ min/mg microsomal protein	0.238 ± 0.018	0.192 ± 0.44	<0.01
Linoleoyl-CoA nmol 18:3 produced/ min/mg microsomal protein	0.282 ± 0.018	0.245 ± 0.025	<0.01

[a]Results are expressed as mean ± standard deviation. Values are given as nmol of 18:3ω6 produced/min/mg microsomal protein. Statistical analysis was by Student t-test.

rats produced a significant decrease in Δ6 and Δ5 desaturase activities in rat liver microsomes. In the liver, this action was attributed to an increase in the intracellular level of cAMP (39) and operated through a β-adrenergic mechanism (42). The increase of cAMP was followed by a glycogen breakdown in liver which led to an increase in blood glucose and a decrease in Δ6 desaturase activity (41). Since epinephrine injection promoted only a modification of the V_{max} of linoleic acid desaturation while K_m remained constant, it was concluded that the inhibitory effect of epinephrine is evoked through a decrease in the amount of active enzyme (41) which could be evoked through an inhibition of the synthesis of Δ6 desaturase.

The results of the present study confirmed the previous findings that Mg^{2+} deficiency increases the plasma total cholesterol and triglyceride levels while decreasing the HDL-cholesterol. It also shows, to the best of our knowledge, for the first time that Mg^{2+} deficiency increases the plasma lipid peroxidation products' level. In addition, it modifies the membrane fatty

acid composition due to the decrease of Δ6 desaturase activity.

The impairment of essential fatty acid metabolism could have an effect on the polyunsaturated essential fatty acid content in the cell membranes this can affect the cellular membrane fluidity and permeability. A known relationship exists between thrombotic-induced aggregation and fatty acid composition of the platelets (43). The possibility exists that Mg^{2+} deficiency may have an affective role on platelet functions. All these changes favor increased risk of atherosclerosis.

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