Short communication

Influence of α-tocopherol on nickel induced alteration of serum lipid profile in male albino rats

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Abstract

We studied the effect of the simultaneous treatment with α-tocopherol (100 mg / kg ; im) and nickel sulfate (20 mg/kg , ip) on nickel-induced changes in serum lipid profiles. Nickel-treated rats showed a significant increase in serum LDL cholesterol, total cholesterol, triglycerides, and a significant decrease in serum HDL cholesterol. Simultaneous administration of α-tocopherol with nickel sulfate improved LDL-cholesterol and HDL-cholesterol level when compared with rats receiving nickel sulfate alone. The results indicate that α-tocopherol is beneficial in preventing nickel-induced lipid profile alterations.

Keywords: nickel sulfate; α-tocopherol; serum lipid profile

INTRODUCTION

Nickel is a naturally occurring element, which can be found, in all environmental media like soil, air, sediment and water. In the metal state nickel is silvery white, hard, malleable and ductile. It is somewhat ferromagnetic and a fair conductor of heat and electricity[1]. Nickel is released into environment through the extraction, processing and use of nickel compounds[2]. The single largest use of nickel is in the manufacture of stainless steel[3]. Other major uses are in electroplating industries, nickel-cadmium batteries, electronic components, fuel cells, petroleum products, preparation of colour pigments *etc*[3,4]. Nickel may also be released from natural resources such as volcanoes, windblown dust, weathering of rocks, forest fires *etc*[2]. The typical atmospheric level of nickel for human exposure ranges from 5-35 ng/m^3, leading to nickel uptake of 0.1-0.7 μg/d *via* inhalation, 100-300 μg/d in diet and 10 μg/L in drinking water[5]. After entering into the body nickel penetrates all organs, accumulating primarily in bone, liver and kidney[2,6] and excreted in bile and urine[7]. If nickel enter and accumulate in the tissue faster than the body's detoxification pathways can dispose it, a gradual buildup of toxins may occur. High concentration of nickel is not necessary to produce a state of toxicity in the body, as it accumulates in tissues and over the period of time, it can reach toxic concentration level[4]. Nickel induced severe liver and kidney damage by altering several marker enzymes and alteration of ascorbate-cholesterol metabolism has been reported earlier[6]. It has been also reported that acute nickel toxicity in rats is associated with lipid peroxidation in target organs[8]. Nickel sulfate administration to male albino rat significantly increased the lipid peroxide (LPO) level and simultaneously decreasd the antioxidant enzyme activities[9-11]. There is also report of depletion of extracellular antioxidant like vitamin C after nickel treatment in rats resulting in activation of HIF-1 transcription factor and up-regulation of hypoxia inducible genes[12]. The most plausible mechanism op-

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erating *in vivo* is the generation of reactive oxygen species (ROS), which may initiates lipid peroxidation, oxidative damage to macromolecules such as protein, DNA and cell damage and death[9,13]. Low-density lipoproteins deposited under the endothelial cell of arterial walls may undergo oxidation by these free radicals released from arterial endothelium and smooth muscle cells, which are deficient in α-tocopherol transfer protein and vitamin E[14]. Liver the major site of detoxification is the primary target of environmental and occupational toxicity. So, protection of hepatic cells from nickel-induced alteration of physiological chemistry is necessary[15]. Vitamin E is the primary liposoluble antioxidant, which have an important role in scavenging free radicals and in stabilizing the cell membranes, thus maintaining its permeability. Vitamin E may also effect oxidative changes, which occur in other cell organelles[16]. It has also been reported by Sato *et al*[17] that vitamin E can synergistically inhibit LDL-oxidation. Among the various forms of vitamin E, the α-tocopherol is the only form that is actively maintained in the human body and is therefore the form of vitamin E found in the largest quantities in the blood and tissue[18]. Hence the present study was undertaken to evaluate the effects of α-tocopherol on nickel sulfate ($NiSO_4$, $6H_2O$) induced alteration of serum lipid profile in male albino rats.

MATERIALS AND METHODS

Adult (aged 60 to 70 d) laboratory-bred male Wister strain rats (160 ± 5g) were fed with laboratory stock diet and water ad libitum for 7 days. The animals were kept in an air-conditioned animal house maintained at 22 to 24 ℃ with ~50% relative humidity. The acclimatized animals were divided into four groups of six animals each. Group I served as untreated control. Group II rats were administered nickel sulfate (Sigma Chemicals, St. Louis, Missouri, USA) in double-distilled water at a dose of 20 mg/kg, intraperitoneally (ip) on alternate days until the tenth dose[19]. Group III rats were treated with α-tocopherol (100 mg/kg, im)[20] and Group IV rats were given both nickel sulfate (20 mg/kg, ip.) and α-tocopherol (100 mg/kg, im) simultaneously on every alternate days until tenth dose. We chose the ip route of administration because a large amount of nickel is required to produce a toxic effect by ingestion, whereas nickel salts administered ip or subcutaneously are highly toxic[21]. The entire experimental protocol was approved by an institutional ethical committee, and utmost care was taken during the experimental procedure, as well as at the time of sacrifice, according to the Declaration of Helsinki (1964)[22]. After treatment, the animals were sacrificed by cervical decapitation between 09: 00 and 11: 00 after overnight fasting. Blood samples were collected in centrifuge tube and allowed them to form serum. Serum total cholesterol, HDL cholesterol, LDL cholesterol and triglyceride were measured using enzymatic estimation kit (ERBA-diagnostics Mannheim, GmbH, Germany)[23].

The mean ± SEM values were calculated for each group. For determining the significance of intergroup differences, each parameter was analyzed separately. A one way analysis of variance (ANOVA) was carried out at 1% of Fischer's distribution to determine which of the groups differed among them. Duncan's multiple range test was applied with the level of significance fixed at $P < 0.05$[24].

RESULTS

In Group II, nickel induced a significant increase in serum LDL cholesterol (LDL-C), total cholesterol (TC) and triglyceride (TG) levels and a significant decrease in serum HDL cholesterol (HDL-C) level (Table 1) in comparison with the control group (Group I). Group IV (nickel sulfate + α-tocopherol) showed a non-significant alteration of LDL cholesterol level, a significant elevated level of serum TC, TG, and a lower level of serum HDL cholesterol in comparison with Group I, but LDL cholesterol level was significantly lower and serum HDL cholesterol level was significantly higher than those in Group II. Our results also showed a significant decrease in serum LDL-C level when the rats were treated with α-tocopherol alone in Group III.

The percentage increase in serum LDL-C, TC and TG level and percentage decrease in serum HDL-C level in Group IV rats in comparison

Table1 Effect of nickel sulfate on serum lipid profile in rats (mean ± SEM, n: 6)

Group[a]	LDL-C(mg/dL)	TC(mg/dL)	HDL-C(mg/dL)	TG(mg/dL)
I	42.15 ±5.32	52.13 ± 6.43	21.45 ± 2.46	76.23 ± 6.56
II	66.14 ± 5.63	80.67 ± 6.56	09.35 ±2.21	145.36 ±12.35
III	32.38 ± 3.56	49.66 ± 4.67	22.53 ±4.34	73.9 ±7.34
IV	46.34 ± 4.84	78.45 ± 4.29	15.39 ±3.67	139.70 ±16.77

I: Untreated control; II: Nickel sulfate; III: α - tocopherol; IV: Nickel sulfate + α- tocopherol. ([a]$P < 0.05$) from each other.

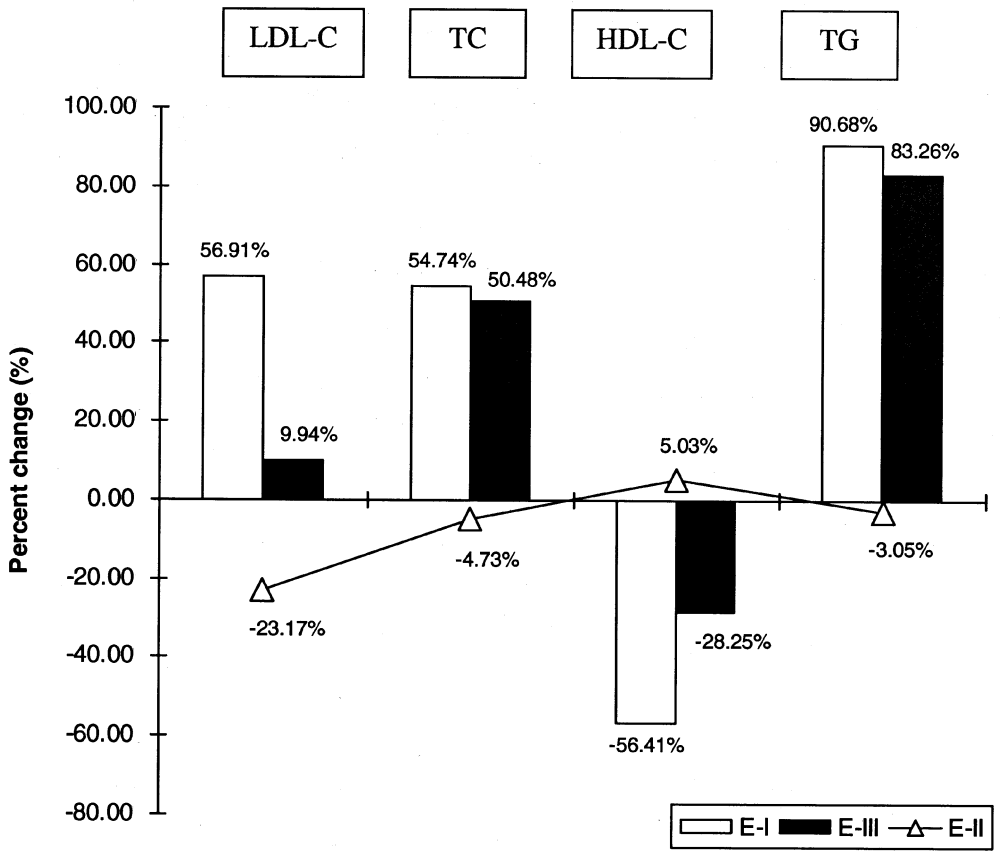

Figure 1 Percent change chart of serum lipid profile after nickel sulfate treatment E I, Group I (control) *vs* Group II (+ $NiSO_4$); E II, Group I *vs* Group III (+ α- tocopherol); E III, Group I *vs* Group IV ($NiSO_4$ + α- tocopherol)

with the control Group I(Figure 1). But compared to Group II, receiving nickel sulfate only, the rise in serum LDL-C and fall in serum HDL-C level were remarkably less (E I *vs* E III). Rats receiving α-tocopherol alone (Group III) showed a significant decrease in serum LDL-C level when compared to control group (E I *vs* E II).

DISCUSSION

Metals like iron, copper, cadmium, nicke, chromium *etc* have been reported to have the ability to produce ROS and free radicals resulting in DNA damage, lipid peroxidation, depletion of protein, sulfhydryls leading to hepatotoxicity, nephrotoxicity and neurotoxicity. Liver, with its metabolic detoxifying function, is extremely vulnerable to harmful substances. The most important consequences of free radical production are lipid peroxidation and change in the permeability of cell membrane[25]. In the present study, nickel induced rise of serum LDL cholesterol, total cholesterol and triglycerides and fall in serum HDL cholesterol in nickel treated rats (Group II) could be due to changes in the gene expression of

hepatic enzymes like HMG-CoA reductase (3-hydroxy 3-methylglutaryl-CoA reductase), which in turn depresses LDL-receptor gene expression. Defects in the LDL-receptor interfere with cholesterol uptake from the blood stream, which in turn causes excess cholesterol synthesis in the liver and high levels of serum total cholesterol and LDL-cholesterol[26]. Possibilities of hypoactivities of lipoprotein lipase in blood vessels after nickel treatment have also been reported by Terasawa *et al*[14]. As a result, triglyceride breaks down inside the chylomicrons, releasing fatty acids in this process resulting in increased risk of atherosclerosis. Also this hyperlipidemia is associated with low level of HDL-C, insulin resistance and poorly controlled diabetes mellitus after chronic nickel treatment [14,27]. High level of serum LDL-C level in Group II rat in present study may also be a result of low level of androgen, which helps in the synthesis of LDL-receptors[28]. Above all, it is referred that nickel toxicity causes a high level of intracellular depletion of vitamin C, which is a potent antioxidant as well as helps in recycling of inactive vitamin E to its active form during the scavenging process[29,30]. This may account to decreased level of active α-tocopherol in the cell membrane resulting in increased lipid peroxidation and an increased level of LDL-C, TC and TG in this study. A significant improvement of lipid profiles in Group IV (nickel sulfate + α-tocopherol) rats in comparison to Group II rats may be due to the presence of vitamin E in all cellular membrane near cytochrome P450 which can sweeps the free radicals formed there. By this way, it can protect polyunsaturated fatty acid from peroxidation. This postulation can be supported by a study by Rao *et al* where α-tocopherol have been found to ameliorate metal induced oxidative stress in liver[31]. It has been also reported that vitamin E can be useful in preventing hepatocellular damage by inhibiting lipid peroxidation by free radicals generated by heavy metals[32]. The restoration of LDL-C and HDL-C level after simultaneous treatment with α-tocopherol in Group IV rats is also supported by another study which shows a significant rise in HDL-C level along with a significant fall in serum LDL-C, TC and TG level when vitamin E was co-supplemented with crude oil[33]. In Group IV rats, α-tocopherol could counteract nickel-induced changes in HMG-CoA reductase activities to some extent and thereby improve the serum lipid profile[34]. α-tocopherol acting in conjugation with glutathione peroxide (GSH-Px) could directly reduce phospholipids hydroperoxides within the membrane and lipoproteins to inhibit lipid peroxidation[35]. The improvement of serum lipid profile also reflects normalization of liver P450 enzyme system function by α-tocopherol[36]. As nickel inhibits the cellular transportation of ascorbic acid and depletes intracellular ascorbate level resulting in decreased level of active α-tocopherol, hence supplementation with extra load of α-tocopherol could reverse the nickel-induced alteration of hepatic enzyme activities that regulate cholesterol biosynthesis.

Thus, nickel sulfate, a toxic heavy metal, adversely affects the serum lipid profile and causes degenerative histopathological changes in rat liver. Simultaneous treatment with α-tocopherol partially improved nickel-induced alterations in serum lipid profiles.

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