DOI: 10.4149/BLL_2015_023

Bratisl Lek Listy 2015; 116 (2)

119 – 123

EXPERIMENTAL STUDY

Effect of melatonin on element distribution in the liver tissue of diabetic rats subjected to forced exercise

Bicer M[1], Akil M[2], Baltaci AK[3], Mogulkoc R[3], Sivrikaya A[4], Akkus H[1]

Selcuk University School of Physical Education and Sports, Konya, Turkey. **rasimmogulkoc@yahoo.com**

Abstract: The objective of the present study was to investigate the effects of melatonin supplementation on elements in the liver of diabetic rats subjected to acute swimming exercise. Eighty adult male rats were equally divided into eight groups. Group 1, general control. Group 2, melatonin-supplemented control. Group 3, melatonin-supplemented diabetic control. Group 4, swimming control. Group 5, melatonin-supplemented swimming. Group 6, melatonin-supplemented diabetic swimming. Group 7, diabetic swimming. Group 8, diabetic control. Liver tissue samples were analyzed for lead, cobalt, molybdenum, chrome, sulphur, magnesium, manganese, sodium, potassium, phosphorus, copper, iron, calcium, zinc, selenium. The highest cobalt, chrome values were found in the groups 7, 8 and the groups 5, 6 respectively. Groups 3 and 7 had the highest copper values. Iron and potassium values were higher in the groups 1 and 4. Group 6 had increased magnesium value, and groups 6, 7, 8 were found to have the highest manganese levels. The highest lead values were found in the groups 5 and 6. Group 6 had the highest selenium levels. The highest zinc levels were established in 1 and 2. Groups 1, 2, 5 and 6 were found to have the highest calcium values. The results of our study indicate that melatonin supplementation in diabetes and forced exercise significantly alters the element metabolism in the liver *(Tab. 3, Ref. 33)*. Text in PDF *www.elis.sk.*

Key words: melatonin supplementation, diabetes, exercise, liver, elements.

Introduction

Diabetes mellitus is a chronic metabolic disease characterized by hyperglycaemia and inadequate secretion or action of endogenous insulin (1). Type I diabetes develops as a result of severe damage to pancreatic cells and commonly results in insulin dependence (1, 2). The metabolic irregularity in diabetes brings about various complications involving both macro- and micro-vascular disorders (2). Oxidative stress is the primary mechanism among the mechanisms presumed to be involved in beta cell damage (3).

It has been noted that the melatonin hormone, which is synthesized by the pineal gland in all mammals and secreted by some tissues including the retina and the gastrointestinal system, is protective against diabetes (4, 5, 6).

Regular exercise and physical activity are known to have a positive effect on cardiovascular diseases and mortality (7). Therefore, exercise is strongly recommended to diabetic patients (8).

Diabetes has been reported to lead to significant changes in the levels of elements in the blood and various tissues (9). Similarly, it has been argued that physical activity causes crucial differences in element levels, which may have resulted from the changes in the distribution of various elements in the liver (10). The protective role of melatonin against diabetes seems to be associated with its free radical scavenging effect (11). We have not found any report concerning the effects of melatonin on the distribution of elements, the blood and tissue levels of which are known to change in diabetes and exercise. The objective of the present study is to investigate the effects of melatonin supplementation on element distribution in the liver of rats in which diabetes was induced with streptozotocin and which were subjected to acute swimming exercise.

[1]Selcuk University School of Physical Education and Sports, Konya, Turkey, [2]Karabuk University, Hasan Dogan High School of Physical Education and Sports, Karabuk, Turkey, [3]Selcuk University, Selcuklu Medical School, Department of Physiology, Konya, Turkey, and [4]Selcuk University, Selcuklu Medical School, Department of Biochemistry, Konya, Turkey

Address for correspondence: R. Mogulkoc, MD, Selcuk University, Selcuklu Medical School, Department of Physiology, 42031, Konya, Turkey.

Materials and methods

Animal groups

This study was conducted on 80 Spraque-Dawley type male rats obtained from Mediterranean University Experimental Medicine Application and Research Centre in the Experimental Animals Unit of Selcuk University School of Veterinary Medicine. The study protocol was approved by the local ethics committee. Experimental animals used in the study were divided into 8 groups:

Group 1 (n:10) General Control Group: the group, which was not subjected to any procedure and was fed on a normal diet.

Group 2 (n:10) Melatonin-Supplemented Control Group: the group, which was fed on a normal diet and supplemented with 3 mg/kg/day intraperitoneal melatonin for 4 weeks.

Group 3 (n:10) Melatonin-Supplemented Diabetic Control Group: the group, in which diabetes was induced by subcutaneous 40 mg/kg streptozotocin (STZ) and which was then supplemented with 3 mg/kg/day intraperitoneal melatonin for 4 weeks.

Group 4 (n:10) Swimming Control Group: the group, which was fed on a normal diet and subjected to 30-minute acute swimming exercise.

Group 5 (n:10) Melatonin-Supplemented Swimming Group: the group, which was fed on a normal diet, supplemented with 3 mg/kg/day intraperitoneal melatonin for 4 weeks, and subjected to 30-minute acute swimming exercise.

Group 6 (n:10) Melatonin-Supplemented Diabetic Swimming Group: the group, in which diabetes was induced by subcutaneous 40 mg/kg streptozotocin (STZ), and which was then supplemented with 3 mg/kg/day intraperitoneal melatonin for 4 weeks and subjected to 30-minute acute swimming exercise.

Group 7 (n:10) Diabetic Swimming Group: the group, in which diabetes was induced by subcutaneous 40 mg/kg streptozotocin (STZ), and which was then subjected to 30-minute swimming exercise.

Group 8 (n:10) Diabetes Group: the group, in which diabetes was induced by subcutaneous 40 mg/kg streptozotocin (STZ).

Experimental animals

The experimental animals were fed in steel cages, which were washed clean daily. Their feed was provided in special steel bowls and their water (normal tap water) in glass feeding bottles. The animals were given about 10 g feed per 100 g of body weight daily. They were kept in an environment where it was 12 hours dark/12 hours light and the standard room temperature (21 ± 1 °C) was maintained. All injections were given between 09.00 a.m. and 10.00 a.m. hours. At the end of the four-week procedures, the animals were decapitated at 09.00–10.00 a.m. and liver tissue samples were collected to determine levels of lead, cobalt, molybdenum, chrome, sulphur, magnesium, manganese, sodium, potassium, phosphorus, copper, iron, calcium, zinc and selenium.

Experimental procedures

Inducement of diabetes in experimental animals

In order to induce diabetes, 40 rats were picked as the diabetes groups. The rats were injected with 40 mg/kg subcutaneous streptozotocin (STZ) "Sigma, S-0130". The injections were repeated at the same dose 24 hours later. Six days after the last injection, blood glucose levels of the animals were determined from the tail vein using a diagnostic glucose kit. The animals whose blood glucose was found at or above 300 mg/dl were accepted as diabetic (12).

Melatonin supplementation

After dissolving 40 mg of melatonin (Sigma M-5250) in 3 ml pure ethanol, this suspension was sealed and stored in dark in the deep freeze until the time of use. From the stock solution 0.1 ml was added 0.9 ml NaCl (3 mg/kg/day) and injected to the rats through the intraperitoneal route at 09.00 a.m. Melatonin supplementation was carried out at the same hours for 4 weeks.

Swimming exercise

Swimming exercise was performed in a heat-resistant glass swimming pool, 50 cm in depth and width, with a thermostat that kept the temperature fixed at 37 °C. The exercise was conducted 24 hours after the end of procedures for once and for a period of 30 minutes. The experimental animals were made to swim in pairs, and decapitated immediately after the exercise to collect liver tissue samples.

Determination of lead, cobalt, molybdenum, chrome, sulphur, magnesium, manganese, sodium, potassium, phosphorus, copper, iron, calcium, zinc and selenium in liver tissue

Liver tissue samples were put into capped, polyethylene tubes washed with NHO_3 and deionised water to avoid contamination and kept at –35 °C until the time of analysis. For the analysis, liver tissues were pounded in a mortar to powder and the wet weight of the tissue was recorded. Then, it was added concentrated H_2SO_4 and concentrated HNO_3 (gram tissue/ml H_2SO_4/ml HNO_3 = 1/1/10). The mixture was kept in a closed-system microwave oven (CEM – Mrasx5) at 170 psa pressure and 200 °C temperature for 20 minutes. The final volume was completed to 25 ml by deionised water, and the samples were read after a maximum 30-minute waiting period. Analyses were conducted in the atomic emission (ICP-AES) equipment in the S.U. Agriculture Faculty, Department of Soil. The results were calculated as mg/L.

Statistical evaluations

Statistical evaluation of the results was carried out using acomputer software. Arithmetic means and standard errors of all parameters were calculated. Variance analysis was employed in the determination of differences between groups. The Least Significant Difference (LSD) Test was used to compare group means, which were found statistically significant in variance analysis results. Differences for which $p < 0.05$ were considered significant.

Results

The highest liver lead levels in the present study were found in the groups 5 and 6 ($p < 0.001$). Lead levels in groups 7 and 8 were lower than those in the groups 5 and 6, but higher than the levels in all other groups ($p < 0.001$). Groups 7 and 8 had cobalt levels significantly higher than all other groups ($p < 0.001$). Levels of molybdenum in liver tissue did not differ significantly among groups. The highest chrome levels were established in the groups 5 and 6 ($p < 0.001$) and the lowest chrome levels in the groups 3 and 4 ($p < 0.001$). Groups 6, 7 and 8 had the highest ($p < 0.001$) and the group 2 had the lowest levels of sulphur ($p < 0.001$) (Tab. 1).

The lowest liver magnesium levels in our study were found in the groups 1 and 2 ($p < 0.001$), while the group 6 had the highest magnesium levels ($p < 0.001$). Manganese levels were the highest in the groups 6, 7 and 8 ($p < 0.001$) and the lowest in the group 4 ($p < 0.001$). Groups 5 and 6 had the highest sodium levels ($p < 0.001$), whereas the groups 4, 7 and 8 had sodium levels lower than all other groups ($p < 0.001$). The highest potassium levels were obtained in the groups 1 and 4 ($p < 0.001$) and the lowest potassium levels in the group 2 ($p < 0.001$). Phosphorus levels in the liver tissue did not differ among groups (Tab. 2).

In the present study, we obtained the highest copper levels in the groups 3 and 7 ($p < 0.001$) and the lowest copper levels in the

Tab. 1. Levels of lead, cobalt, molybdenum, chrome and sulphur in liver tissue (mg/L).

Groups	lead	cobalt	molybdenum	chrome	sulphur
1 General Control	2.43±0.81^{C}	0.22±0.19^{B}	0.72±0.12	1.37±0.34^{B}	1161.1±59.1^{B}
2 Melatonin-Supplemented Control	2.56±0.85^{C}	0.17±0.18^{B}	0.76±0.15	1.70±0.53^{B}	816.3±144.8^{C}
3 Melatonin-Supplemented Diabetic Control	0.97±0.11^{C}	0.16±0.16^{B}	1.12±0.98	0.92±0.14^{C}	1161.3±84.5^{B}
4 Swimming Control	1.13±0.30^{C}	0.11±0.15^{B}	1.01±0.58	0.89±0.96^{C}	1139.8±245.6^{B}
5 Melatonin-Supplemented Swimming	8.64±2.64^{A}	0.13±0.12^{B}	1.07±0.27	2.12±0.61^{A}	1107.4±90.7^{B}
6 Melatonin-Supplemented Diabetic Swimming	9.91±2.16^{A}	0.22±0.20^{B}	1.16±0.16	2.33±0.53^{A}	1287.2±67.4^{A}
7 Diabetic Swimming	6.96±2.24^{B}	0.54±0.26^{A}	1.09±0.41	1.83±0.68^{B}	1308.0±66.3^{A}
8 Diabetes	5.81±1.06^{B}	0.53±0.30^{A}	0.92±0.12	1.61±0.26^{B}	1203.2±61.6^{A}

*Means with different superscripted letters in the same column are statistically significant ($p < 0.001$).

Tab. 2. Levels of magnesium, manganese, sodium, potassium and phosphorus in liver tissue (mg/L).

Groups	magnesium	manganese	sodium	potassium	phosphorus
1 General Control	151.56±37.44^{C}	1.39±0.20^{B}	1775.7±72.1^{B}	2.7±0.2^{A}	3503.2±179.2
2 Melatonin-Supplemented Control	160.50±36.72^{C}	1.45±0.33^{B}	1819.2±311.8^{B}	1.0±0.3^{C}	3417.9±281.8
3 Melatonin-Supplemented Diabetic Control	287.26±30.30^{B}	1.48±0.35^{B}	1854.4±210.4^{B}	1.8±0.3^{B}	3397.7±284.8
4 Swimming Control	284.33±62.95^{B}	0.48±0.52^{C}	1255.3±81.7^{C}	2.9±0.5^{A}	3377.1±144.6
5 Melatonin-Supplemented Swimming	297.67±35.26^{B}	1.46±0.17^{B}	2572.3±388.8^{A}	2.0±0.5^{B}	3557.1±188.5
6 Melatonin-Supplemented Diabetic Swimming	366.39±18.25^{A}	2.09±0.27^{A}	2587.6±327.3^{A}	2.0±0.3^{B}	3370.5±220.0
7 Diabetic Swimming	295.78±42.77^{B}	2.40±0.67^{A}	1290.4±92.5^{C}	1.8±0.2^{B}	3528.2±353.6
8 Diabetes	264.93±21.02^{B}	2.06±0.37^{A}	1275.6±95.3^{C}	1.9±0.2^{B}	3530.9±240.2

*Means with different superscripted letters in the same column have statistical significance ($p < 0.001$).

Tab. 3. Levels of copper, iron, calcium, zinc and selenium in liver tissue (mg/L).

Groups	copper	iron	calcium	zinc	selenium
1 General Control	3.08±0.23^{C}	175.66±22.19^{A}	160.22±46.63^{A}	19.73±1.49^{A}	0.28±0.15^{B}
2 Melatonin-Supplemented Control	3.11±0.65^{C}	85.63±28.50^{C}	169.98±29.24^{A}	18.01±1.76^{A}	0.27±0.20^{B}
3 Melatonin-Supplemented Diabetic Control	5.95±1.78^{A}	130.43±25.26^{B}	118.51±24.81^{B}	13.45±1.46^{B}	0.10±0.03^{C}
4 Swimming Control	4.60±0.97^{B}	181.37±44.52^{A}	115.11±57.49^{B}	13.81±5.89^{B}	0.09±0.03^{C}
5 Melatonin-Supplemented Swimming	4.65±0.52^{B}	138.25±22.79^{B}	173.63±76.24^{A}	14.76±1.78^{B}	0.27±0.06^{B}
6 Melatonin-Supplemented Diabetic Swimming	4.80±0.40^{B}	137.45±20.04^{B}	163.49±70.02^{A}	13.52±1.18^{B}	0.39±0.08^{A}
7 Diabetic Swimming	6.32±2.42^{A}	138.69±17.71^{B}	120.31±44.93^{B}	09.07±1.64^{C}	0.27±0.08^{B}
8 Diabetes	4.57±0.23^{B}	136.74±21.15^{B}	124.36±32.09^{B}	10.60±1.23^{C}	0.26±0.19^{B}

*Means with different superscripted letters in the same column have statistical significance ($p < 0.001$).

groups 1 and 2 ($p < 0.001$). Groups 1 and 4 had the highest iron levels ($p < 0.001$) and the group 2 had the lowest ($p < 0.001$). Liver calcium was found higher than all other groups in the groups 1, 2, 5 and 6 ($p < 0.001$). The highest liver zinc was established in the groups 1 and 2 ($p < 0.001$), while the lowest zinc levels were in the groups 7 and 8 ($p < 0.001$). The group 6 had the highest selenium levels ($p < 0.001$), while selenium levels were the lowest in the groups 3 and 4 ($p < 0.001$) (Tab. 3).

Discussion

When compared to the controls, lead, cobalt, chrome and sulphur values in the liver tissue of diabetic rats were found elevated in the present study. However, only melatonin supplementation was observed to reduce particularly lead, chrome and sulphur values in the liver tissue of diabetic rats. Similar results were obtained in diabetic animals subjected to swimming exercise as well. It was noted that element distribution not only in the blood, but also in the tissues was impaired in diabetic rats subjected to maximal exercise (13). Likewise, only intense exercise was reported to affect the element metabolism (14) and it was accepted that these post-exercise differences could have resulted particularly from the exchange of elements between the extracellular fluid and tissues (10). That melatonin supplementation reduced the elevated levels of lead and sulphur we obtained in diabetic rats in particular suggested that melatonin supplementation maight be important in inhibiting the possible toxic effects of these elements. Similarly, it was already reported that melatonin supplementation prevented toxic effects caused by both trace and major elements in various tissues of rats, including the liver tissue (15). Dogukan et al (16) reported reduced chrome levels in the liver tissue of diabetic rats in their study. In our study, chrome levels in the liver tissue of diabetic rats were not different from those in the controls. However, melatonin administration led to an increase in liver tissue chrome levels in both swimming (group 5) and diabetic swimming (group 6) groups. In consideration of the importance of chrome in carbohydrate and lipid metabolisms (17) and its effect in the regulation of blood glucose (18), elevated chrome levels we found with melatonin supplementation in diabetic rats seemed to be a crucial finding. In our study, we established the highest liver cobalt levels in both diabetic exercise (group 7) and non-exercised diabetes (group 8) groups. High cobalt levels are known to stimulate the production of free radicals (19). Our finding may be pointing out the possible role of cobalt in the pathogenesis of lipid peroxidation, which increases in diabetes. We could not find any literature study with which we could directly compare the molybdenum pa-

rameter in liver tissue. But then, molybdenum levels of the study groups did not differ anyway.

The highest liver magnesium was found in the group 6 (melatonin-supplemented diabetic swimming) and the highest manganese levels were established in the groups 6 (melatonin-supplemented diabetic swimming), 7 (diabetic swimming) and 8 (diabetes). Magnesium is associated with enzyme activities that involve insulin (20). Therefore, magnesium is involved in mechanisms regulating the blood glucose (21). It is known that magnesium levels drop in diabetic patients (22). Elevated magnesium levels we found with melatonin supplementation in the group 6 (melatonin-supplemented diabetic swimming) in our study maight be evaluated as a striking result, in consideration of the effects of this element in the glucose metabolism (20, 21). Physical activity was shown to result in an increase in manganese activity in rats (22) However, it is also known that excessive accumulation of manganese causes lipid peroxidation through a toxic effect (23). Therefore, elevated manganese levels we established in the diabetes groups are important with respect to the relation between diabetes and lipid peroxidation. In our study, the highest sodium levels were found in the groups 5 (melatonin-supplemented swimming) and 6 (melatonin-supplemented diabetic swimming), while the lowest sodium levels were established in the groups 4 (swimming), 7 (diabetic swimming) and 8 (diabetes). Diabetic patients were reported to have reduced sodium levels, in comparison to their controls (24). The reduced sodium levels we found in diabetic groups in our study are consistent with the results of Harmer and colleagues (2006). What is more important and needs emphasis here is that the reduction in sodium levels in diabetic animals was inhibited by melatonin supplementation. Potassium levels did not differ among diabetic groups and melatonin supplementation did not affect the potassium parameter of diabetic animals. Phosphorus levels in the liver tissue of study groups were not different either.

Groups 1 (general control) and 4 (swimming control) had the highest liver iron. It was argued that exercise modified distribution of iron in various tissues (25) and also that moderate exercise had a regulatory effect on body iron values (26). The results we found in relation to the iron parameter in our study are congruous with the results of the above-cited researchers. Our study results pertinent to liver iron demonstrate that diabetes and melatonin supplementation did not have a noteworthy effect on liver iron in diabetic animals.

The lowest liver calcium was found in the groups 3 (melatonin-supplemented diabetes), 4 (swimming), 7 (diabetic swimming) and 8 (diabetes). It was stated that calcium release was impaired in diabetes (27) and that diabetic rats suffered from a decrease in calcium levels and a parallel increase in the urinary excretion of calcium (28). Reduced liver calcium we found in diabetic animals in the present study was parallel to the results of the above researchers.

All the diabetic and exercised groups in our study had elevated copper values, relative to the control groups (groups 1 and 2). Diabetic animals were shown to have elevated blood and tissue copper levels (16). Elevated copper levels in the liver tissue, relative to the controls, indicate that increased liver copper is involved in the events associated with lipid peroxidation, which increase in diabetes (3).

The lowest liver zinc was found in the groups 7 and 8 in our study. It was shown that zinc levels dropped significantly in the blood and tissues of diabetic rats (16, 29) and that this decrease in zinc levels was also observed in humans (4). Reduced zinc levels we established in diabetic animals (groups 7 and 8) in our study are consistent with literature data. However, what is more important is that melatonin supplementation offsets the decrease in liver zinc in diabetes. Liver zinc in melatonin-supplemented groups in our study was higher than those in non-supplemented diabetic groups (groups 7 and 8). There are numerous studies showing the relation between melatonin and zinc, an important trace element (30, 31). Zinc is found in the structure of serotonin, which is required for the synthesis of melatonin (32). Melatonin, in turn, increases the absorption of zinc in the digestive system (30, 31). Results of the cited researchers are in harmony with elevated zinc levels we obtained with melatonin supplementation.

In the study we established that the group 6 (melatonin-supplemented swimming) had the highest liver selenium. It was reported that liver selenium decreased in diabetes (16) and that this decrease might lead to a reduction in antioxidant activity (33). We could not find any study, with which we could compare the melatonin-selenium relation on a one-to-one basis. However, elevated liver selenium we found in melatonin-supplemented group 6, in comparison to diabetic groups, what seems to be an interesting result.

Conclusions

The results of our study indicate that melatonin supplementation in diabetes and acute exercise significantly changes the element metabolism of the liver tissue. Prevention of the decrease in liver zinc in diabetes by melatonin supplementation in particular suggests that melatonin treatment can be beneficial in diabetes.

References

1. Laakso M. Cardiovascular disease in type 2 diabetes: challenge for treatment and prevention. J Intern Med 2001; 249: 225–235.

2. Rahimi R, Nikfar S, Larijani B, Abdollahi M. A review on the role of antioxidants in the management of diabetes and its complications. Biomed Pharmacother 2005; 59: 365–373.

3. Islam MS, Loots Du T. Diabetes, metallothionein, and zinc interactions: a review. Biofactors 2007; 29: 203–212.

4. Al-Maroof RA, Al-Sharbatti SS. Serum zinc levels in diabetic patients and effect of zinc supplementation on glycemic control of type 2 diabetics. Saudi Med J 2006;. 27: 344–350.

5. Abdel-Wahab MH, Abd-Allah AR. Possible protective effect of melatonin and/or desferrioxamine against streptozotocin-induced hyperglycaemia in mice. Pharmacol Res 2000; 41: 533–537.

6. Gorgun FM, Ozturk Z, Gumustas MK, Kokogu E. Melatonin administration affects plasma total sialic acid and lipid peroxidation levels in streptozotocin induced diabetic rats. J Toxicol Environ Health A 2002; 65: 695–700.

7. Paffenbarger RS, Kampert JB, Lee IM, Hyde RT, Leung RW, Wing AL. Changes in physical activity and other lifeway patterns influencing longevity. Med Sci Sports Exerc 1994; 26: 857–865.

8. Kim JD, Yu BP, McCarter RJM, Lee SY, Herlihy JT. Exercise and diet modulate cardiac lipid peroxidation and antioxidant defenses. Free Radic Biol Med 1996; 20: 83–88.

9. Jansen J, Karges W, Rink L. Zinc and diabetes -clinical links and molecular mechanisms J Nutr Biochem 2009; 20: 399–417.

10. Cordova A, Gimenez M, Escanero JF. Changes of plasma zinc and copper at various times of swimming until exhaustion in the rat. J. Trace Elem. Electrolytes Health Dis 1990; 4: 189–192.

11. Baydas G, Tuzcu M, Yasar A, Baydas B. Early changes in glial reactivity and lipid peroxidation in diabetic rat retina: effects of melatonin. Acta Diabetol 2004; 41: 123–128.

12. Havel PJ, Uriu-Hare JY, Liu T, Stanhope KL, Stern JS, Keen CL, Ahrén B. Marked and rapid decreases of circulating leptin in streptozotocin diabetic rats: reversal by insulin. Am J Physiol 1998; 274: 1482–1491.

13. Cordova A. Zinc content in selected tissues in streptozotocin-diabetic rats after maximal exercise. Biol Trace Elem Res 1994; 42: 209–216.

14. Marrella M, Guerrini F, Solero PL, Tregnaghi PL, Schena F, Velo GP. Blood copper and zinc changes in runners after a marathon. J Trace Elem Electrolytes Health Dis 1993; 7: 248–250.

15. Cemek M, Büyükokuroğlu M, Yürümez Y, Yavuz Y, Aslan A, Büyükben A, Aymelek F. Tissue trace and major element levels in organophosphate insecticide fenthion (Lebaycid) toxicity in rats: prophylactic and therapeutic effect of exogenous melatonin. Toxicol Environ. Saf 2010; . 73: 206–212.

16. Dogukan A, Sahin N, Tuzcu M, Juturu V, Orhan C, Onderci M, Komorowski J, Sahin K. The effects of chromium histidinate on mineral status of serum and tissue in fat-fed and streptozotocin-treated type II diabetic rats. Biol Trace Elem Res 2009; 131: 124–132.

17. Anderson RA, Polansky MM, Bryden NA, Roginski EE, Patterson KY, Reamer DC. Effect of exercise (running) on serum glucose, insulin, glucagon, and chromium excretion. Diabetes 1982; 31: 212–216.

18. Anderson RA. Chromium metabolism and its role in disease processes in man. Clin Physiol Biochem 1986; 4: 31–41.

19. Valko M, Morris H, Cronin MT. Metals, toxicity and oxidative stress. Curr Med Chem 2005; 12: 1161–1208.

20. Wells IC. Evidence that the etiology of the syndrome containing type 2 diabetes mellitus results from abnormal magnesium metabolism. Can J Physiol Pharmacol 2008; 86: 16–24.

21. Song Y, Manson JE, Buring JE, Liu S. Dietary magnesium intake in relation to plasma insulin levels and risk of type 2 diabetes in women. Diabetes Care 2004; 27: 59–65.

22. Husain K. Interaction of physical training and chronic nitroglycerin treatment on blood pressure, nitric oxide, and oxidants/antioxidants in the rat heart. Pharmacol Res 2003; 48: 253–261.

23. Zaidi S, Patel A, Mehta N, Patel K, Takiar R, Saiyed H. Early biochemical alterations in manganese toxicity: ameliorating effects of magnesium nitrate and vitamins. Ind Health 2005; 43: 663–668.

24. Harme AR, Ruell PA, McKenna MJ, Chisholm DJ, Hunter SK, Thom JM, Morris NR, Flack JR. Effects of sprint training on extrarenal potassium regulation with intense exercise in Type 1 diabetes. J Appl Physiol 2006; 100: 26–34.

25. Kaptanoğlu B, Turgut G, Genç O, Enli Y, Karabulut I, Zencir M, Turgut S. Effects of acute exercise on the levels of iron, magnesium, and uric acid in liver and spleen tissues. Biol Trace Elem Res 2003; 91: 173–178.

26. Liu YQ, Duan XL, Chang YZ, Wang HT, Qian ZM. Molecular analysis of increased iron status in moderately exercised rats. Mol Cell Biochem 2006; 282: 117–123.

27. Reuter H, Gronke S, Adam C, Ribati M, Brabender J, Zobel C, Frank KF, Wippermann J, Schwinger RH, Brixius K, Muller-Ehmsen J. Sarcoplasmic Ca^{2++} release is prolonged in nonfailing myocardium of diabetic patients. Mol Cell Biochem 2008; 308: 141–149.

28. Fushimi H, Inoue T, Yamada Y, Horie H, Kameyama M, Inoue K, Minami T, Okazaki Y. Zinc deficiency exaggerares diabetik osteoporosis. Diabetes Res Clin Pract 1993; 20: 191–196.

29. Duzguner V, Kaya S. Effect of zinc on the lipid peroxidation and the antioxidant defense systems of the alloxan-induced diabetic rabbits. Free Radic Biol Med 20078; 42: 1481–1486.

30. Mocchegiani E, Bulian D, Santarelli L, Tibaldi A, Muzzioli M, Pierpaoli W, Fabris N. 1994. The immuno-reconstituting effect of melatonin or pineal grafting and its relation to zinc pool in aging mice. J Neuroimmunol 1994; 53: 189–201.

31. Mocchegiani E, Bulian D, Santarelli L, Tibaldi A, Muzzioli M, Lesnikov V, Pierpaoli W, Fabris N. The zinc pool is involved in the immune-reconstituting effect of melatonin in pinealectomized mice. J Pharmacol Exp Ther 1996; 277: 1200–1208.

32. Johnson S. Micronutrient accumulation and depletion in schizophrenia, epilepsy, autism and Parkinson's disease? Med Hypotheses 2001; 56: 625–642.

33. Jana K, Samanta PK, Manna I, Ghosh P, Singh N, Khetan RP, Ray BR. Protective effect of sodium selenite and zinc sulfate on intensive swimming-induced testicular gamatogenic and steroidogenic disorders in mature male rats. Appl Physiol Nutr Metab 2008; 33: 903–914.

Received July 31, 2013.
Accepted May 28, 2014.