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# EFFECT OF NOCTURNAL EXHAUSTION EXERCISE ON THE METABOLISM OF SELECTED ELEMENTS

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*Abstract* - The present study aims to examine how exercise performed until fatigue at night affects element distribution in the serum. The study examined 10 healthy sedentary males who were not actively engaged in any particular sport and whose mean age was 23.00±0.25 years, mean height 177.79±2.25 cm, and mean weight 70.70±1.63 kg. Blood samples were collected from the subjects at midnight twice: during rest before exercise and after exercise. Serum phosphorus, sodium, potassium, sulfur (mmol/L), cobalt, boron, cadmium, chrome, nickel, manganese, molybdenum, copper, iron, zinc and calcium levels (mg/L) were measured using atomic emission spectroscopy (ICP-AES). Exhaustion exercise performed at night brought about a decrease in copper levels only ($p<0.05$), while elevating levels of potassium, sodium, magnesium, calcium, iron, zinc, manganese, nickel, selenium, molybdenum, chrome, cobalt, lead and cadmium ($p<0.05$). The results of the study demonstrate that nighttime exercise until exhaustion significantly alters element metabolism.

*Key words*: Nocturnal exercise, element metabolism, exhaustion

## INTRODUCTION

Regulation of some elements in the blood and tissues is necessary for the metabolic functions of the body to be carried out within their normal range. Strenuous physical activity can cause a multifold increase in the energy conversion rate in the skeletal muscle and this increase can change the levels of elements in the body in various ways (Maughan, 1999). It is assumed that of these elements, potassium, sodium, phosphorus, magnesium and calcium are pivotal for metabolic balance during physical activity and sports, as they are involved in the structure of various bone elements, nucleic acids, cell membranes, proteins and enzymes (Maughan, 1999).

That physical exercise causes redistribution of various trace elements between body stores, blood and tissues and that the increased metabolism results in the deficiency of these elements, are indicators of a possible relation between sportive performance and element metabolism (Bordin et al., 1993). Thus, it is very important to reveal how exercise affects the functions of these elements (König et al., 1998). Elements in the ionized form are involved in heart and muscle contraction, oxidative phosphorylation, and synthesis and activation of enzymatic systems in athletes (Speich et al., 2001). Elements such as zinc, copper and manganese (copper-zinc superoxide dismutase; manganese superoxide dismutase) exert a protective effect against increased free reactive $O_2$ species (Maughan, 1999). Selenium in glutathione peroxidase has a protective effect on the cardiovascular system and muscle (Margaritis et al., 1997). Copper and iron are involved in the events associated with the energy metabolism. They are important in the synthesis of hemoglobin, myoglobin and cytochromes (Speich et al., 2001). Therefore, physical activity may help people exposed to metal contamination (lead, cadmium). However, in many cases, it is not clear which metals are involved in the metabolic changes resulting from physical activity. The purpose of the present study is to explore how nighttime exercise until exhaustion affects element distribution in the serum.

## MATERIALS AND METHODS

### *Subjects*

The study registered 10 healthy sedentary males who were not actively engaged in sports. Mean age of the subjects was 23.00±0.25 years, mean height was 177.70±2.25 cm, and mean weight was 70.10±1.63 kg. The study protocol was approved by the Ethics Committee of Selcuk University School of Physical Education and Sports.

### *Exhaustion Exercise (Bruce protocol)*

Exhaustion was induced according to the Bruce protocol, the most commonly used clinical exercise test where a 10% incline and 2.7 km/h speed of the treadmill is increased at 3-min periods (Cosmed T150 treadmill).

### *Collection of blood samples*

Blood samples of 2.5 ml were drawn from the forearm veins of the subjects at midnight before exhaustion exercise. A second round of samples were collected in the same way immediately after exercise. The samples were centrifuged at 3 000 rpm for 10 min to separate the serum, which was then stored at -80°C until analysis.

### *Serum element analyses*

After the blood samples (2.5 ml) collected from the forearm veins of the subjects to determine serum element levels were centrifuged to separate the serum, the samples were put into plastic-capped tubes that were then kept at -80°C until the day of analysis. Phosphorus, sodium, potassium, sulfur (mmol/L), cobalt, boron, cadmium, chrome, nickel, manganese, molybdenum, copper, iron, zinc and calcium (mg/L) levels in the serum were determined following the atomic emission method.

The inductively coupled plasma emission spectrophotometry (ICP-AES; Varian Australia Pty Ltd., Australia) atomic emission device located in the Soil Sciences Department of Selcuk University Faculty of Agriculture was used for element analyses.

### *Statistical evaluations*

SPSS 16.0 computer software was used for statistical evaluation of the findings. Arithmetic means and standard errors of all parameters were calculated. A normality test was conducted to determine if data had normal distribution. The Wilcoxon test was used to identify any differences between two measurement times. Differences with a p value <0.05 were accepted to be significant.

## RESULTS DISCUSSION

Serum potassium, sodium, magnesium and calcium values after nocturnal exercise were significantly higher in comparison to pre-exercise values ($p<0.05$). Phosphorus and sulfur levels during rest and after exhaustion were not different (Table 1). Serum iron, zinc, manganese and nickel levels in exhaustion were found to be significantly higher than the levels of the same elements measured during rest ($p<0.05$). Boron levels in exhaustion and at rest were not different, but nocturnal exercise until exhaustion led to a decrease

**Table 1.** Serum potassium, sodium, phosphorus, sulfur, magnesium and calcium levels in the subjects.

| Elements | Before Night Exercise | After Night Exercise |
|---|---|---|
| | Rest | Exhaustion |
| Potassium (mmol/L) | 3.80±0.05[b] | 4.40±0.04[a] |
| Sodium (mmol/L) | 43.39±5.50[b] | 66.60±6.60[a] |
| Phosphorus(mmol/L) | 18.3±3.20 | 19.5±3.30 |
| Sulfur (mmol/L) | 1.64±0.50 | 1.70±0.30 |
| Magnesium (mg/L) | 52.89±5.94[b] | 72.16±6.84[a] |
| Calcium (mg/L) | 36.00±4.83[b] | 68.70±9.64[a] |

a,b: Differences between means with different superscripted letters in the same line are statistically significant ($p<0.05$).

**Table 2.** Serum iron, zinc, manganese, boron, copper and nickel levels (mg/L) in the subjects.

| Elements | Before Night Exercise | After Night Exercise |
|---|---|---|
| | Rest | Exhaustion |
| Iron | 5.66±0.530[b] | 8.17±1.73[a] |
| Zinc | 1.73±0.049[b] | 2.10±0.123[a] |
| Manganese | 0.27±0.042[b] | 0.43±0.050[a] |
| Boron | 0.10±0.016 | 0.09±0.010 |
| Copper | 0.05±0.048[a] | 0.01±0.008[b] |
| Nickel | 0.04±0.009[b] | 0.08±0.013[a] |

a,b: Differences between means with different superscripted letters in the same line are statistically significant ($p<0.05$).

**Table 3.** Serum selenium, molybdenum, lead, chrome, cobalt, and cadmium levels (mg/L) in the subjects.

| Elements | Before Night Exercise | After Night Exercise |
|---|---|---|
| | Rest | Exhaustion |
| Selenium | 0.18±0.044[b] | 0.23±0.063[a] |
| Molybdenum | 0.03±0.055[b] | 0.05±0.008[a] |
| Lead | 0.02±0.012[b] | 0.04±0.008[a] |
| Chrome | 0.02±0.003[b] | 0.04±0.005[a] |
| Cobalt | 0.01±0.003[b] | 0.04±0.006[a] |
| Cadmium | 0.01±0.004[b] | 0.03±0.003[a] |

a,b: Differences between means with different superscripted letters in the same line are statistically significant ($p<0.05$).

in copper levels ($p<0.05$, Table 2). Nocturnal exercise until exhaustion significantly elevated serum selenium, molybdenum, lead, chrome, cobalt and nickel levels of the subjects ($p<0.05$, Table 3).

## DISCUSSION

The presented findings show that exercise performed at night did not cause any change in phosphorus and sulfur parameters in comparison to pre-exercise values of these elements, but increased levels of potassium, sodium, magnesium and calcium. Phosphorus is involved in several events during physical activity, including maintenance of metabolic balance, energy metabolism and cellular respiration (Speich et al., 2001). However, interestingly, there are only a few studies about the relation between phosphorus and sulfur and exercise. In fact, night exercise did not change the phosphorus and sulfur levels. It was reported that long-running exercise in a high-temperature and humid environment led to an increase in sodium and potassium levels and significantly inhibited magnesium levels (Singh and Sirisinghe, 1999). The increased sodium and potassium values in nocturnal exercise are consistent with the results of Singh and Sirisinghe (1999), while magnesium levels are not (Buchman et al., 1998). As reported by Shephard and Shek (1998), maximal exercise could cause an increase in serum magnesium by reducing the amount of magnesium that can be filtered by the kidneys. Likewise, reports of women from different sports branches having higher serum magnesium levels than their controls (Nuviala et al., 1999) are in agreement with the elevated magnesium levels we found in nighttime exercise. Concerning the relation between exercise and calcium, it was argued that very high-intensity exercise could increase calcium loss (Maughan, 1999). In this study, we found increased serum calcium values with nighttime exercise. This result of ours seems to be inconsistent with Maughin's report (Maughan, 1999). However, increased serum calcium in our study resulted from an exhaustion exercise. The study by Sivrikaya et al. (2012), who reported that acute swimming exercise could cause an increase in body calcium, can be considered to support our results.

When potassium, sodium, phosphorus, sulfur, magnesium and calcium parameters were assessed together in our study, it was seen that nocturnal exercise until exhaustion elevated all these parameters. These results may be important. However, our medline review did not show any study with which we could directly compare these results. The subjects in this study were individuals who were not performing any sports activity and these individuals experienced late night exercise (24 h) for the first time in their lives. The stress caused by the event itself might have contributed to these results. Consequently, our results indicate that nocturnal exercise significantly elevated potassium, sodium, phosphorus, sulfur, magnesium and calcium levels.

Nighttime exhaustion exercise in this study did not cause any change in boron relative to pre-exercise values, but brought about an increase in iron, zinc, manganese, copper and nickel levels. Deficiency of iron, which is an $O_2$-binding molecule of cytochromes and a cofactor of a number of enzymes, directly causes hemoglobin deficiency and, acting through the heart and the muscle system, leads to decreased exercise performance (Maughan 1999; Speich et al., 2001). Therefore, there is a critical relationship between physical performance and iron. The study reporting higher serum iron levels in rats subjected to acute swimming exercise than those in the control animals (Baltaci et al., 2009) is consistent with the elevated iron levels we found after nocturnal exercise in our study. Zinc, which is involved in nucleic acid synthesis, protein synthesis, growth and development, antioxidant activity, testosterone secretion and brain functions, is related to more than 300 enzymes in the body. Rodriguez Tuya et al. (1996) reported higher plasma zinc levels in individuals performing judo and fencing sports. Similarly, female athletes, of whom 16 were karate players and 23 were medium- and long-distance runners, were shown to have higher serum zinc (Nuviala et al., 1999). The results of both of these studies are supportive of the elevated serum zinc we obtained with nighttime exercise. Too little is known about the relation between sports and physical activity on one the hand, and manganese concentrations and the changes in them, on the other (Speich et al., 2001). However, given the exercise stress and antioxidant status, it is safe to assume that manganese, which is involved in the structure of superoxide dismutase (SOD), may be important in exercise. In that respect, the elevated manganese levels we obtained after nighttime exercise may prove to be a noteworthy result. Studies into the relation between boron and exercise usually focus on the effects of this element on muscle development and bone metabolism (Naghii and Samman, 1993). Although it was noted that boron could be important in physical performance, the nighttime exercise in our study did not change the levels of this element. In fact, studies about the relation between boron and exercise generally address boron deficiency and supplementation (Yazici et al., 2008; Yazici et al., 2011). A critical element in energy metabolism, copper is necessary for the synthesis of hemoglobin, myoglobin, cytochromes and some peptide hormones (Maughan, 1999). Although copper plays an important role during physical activity, there are different results about the copper status of athletes (Resina et al., 1991). Due to the changes in the ceruloplasmin and copper-binding enzymes, blood copper concentrations in athletes can be higher, lower, or not different from those in the control group (Resina et al., 1991). Male ice hockey players were found to have lower serum copper levels than the controls (Rankinen et al., 1995). Other researchers also reported significant decreases in serum copper levels after physical activity (Marrella et al., 1993; Bicer et al., 2011). The reduced serum copper concentrations we found after nighttime exercise are similar to the results of the researchers cited above. We did not find any study about the relationship between serum nickel levels and exercise in our medline review. However, nickel has already been shown to cause cell damage when it accumulates in the tissues (Gupta et al., 2007). Elevated serum nickel levels following nighttime exercise in our study may be associated with cellular damage and possibly represent the first result regarding the relation between nighttime exercise and nickel.

Serum selenium, molybdenum, lead, chrome, cobalt and cadmium levels in the study displayed a significant increase after nocturnal exhaustion exercise compared to the pre-exercise levels of these elements. Selenium, which is pivotal for human health, is required in various metabolic processes, including thyroid hormone metabolism, protection against oxidative stress and immune functions (Akil et al., 2011). Selenium activates glutathione peroxidase, and therefore it is involved in the antioxidant mechanisms that prevent oxidant damage (Akil et al., 2011). Exhaustive physical exercise is known to cause oxidant damage, possibly by triggering free radical production in many tissues such as muscle, liver, heart and lungs in animals (Akil et al., 2011). Given that exercise increases oxidative stress and selenium stimulates antioxidant activity, there seems to be an inevitable relation between selenium and exercise (Akil et al., 2011). Data about how selenium is affected by exercise are not consistent. Akil et al. (2011) showed that acute swimming exercise reduced serum selenium levels. It was noted that distribution of selenium in the body could change during exercise (Grant et al., 2002) and that this change might be important for physical performance (Thomson, 2004). In our study, the increased serum selenium levels observed after nocturnal exercise may have stemmed from the stress caused by late-night exercise on the subjects and the altered selenium distribution between blood and tissues due to acute and exhausting exercise. When in excess, molybdenum, an essential trace element in both humans and animals, is known to be able to cause inadequate copper absorption (Thomson, 2004). In our study however, molybdenum levels increased significantly after nighttime exercise. There seems to be an inverse relation between molybdenum and copper (Thomson, 2004). The elevated molybdenum levels we found may be attributed to the decreased serum copper mentioned above. To say the least, this result is consistent with the report of a possible relation between molybdenum and copper (Thomson, 2004). Chrome metabolism is known to change under stress (Speich et al., 2001). It was suggested that endurance training could increase urinary excretion of chrome through the kidneys and negatively influence the chrome metabolism (Frentsos and Baer, 1997). It was reported that chrome supplementation did not have any effect on the performance of young male and female athletes (Lukashi, 2000), and the same results were obtained in football players (Clancy et al., 1994). The elevated serum chrome we found after nighttime exercise in this study may have resulted from stress or exhaustion caused by exercise and/or the hemoconcentration mechanism. There are no scientific data suggesting that cobalt is beneficial to performance and/or that the need for cobalt increases during physical activity (Maughan, 1999). However, it was recommended that cobalt should be supplemented in a biologically active form in the diet, as it is involved in the structure of vitamin B12, and that cobalt supplementation may be of use to athletes (Maughan, 1999; Speich et al., 2001). Considering these data, the increased serum cobalt we found in nighttime exercise may be an interesting result. Cadmium intake to the organism takes place through the lungs (industrial wastes, smoking), food, and drinking water (Speich et al., 2001). Copper, which blocks the biocatalytic effect of zinc and cobalt, causes a toxic effect in the erythrocytes, kidneys and liver (Speich et al., 2001). These findings apply to lead, which is toxic for the organism as well (Speich et al., 2001). Nonetheless, cadmium and lead values in athletes are open to interpretation (Rodriguez Tuya et al., 1996). The results of our study demonstrate that nighttime exercise elevated both serum cadmium and lead levels when compared to pre-exercise levels. As with the previous findings, increased cadmium and lead levels may have resulted from factors like the exhaustion and stress caused in the organism by nighttime exercise, because the subjects in our study were sedentary individuals who did not exercise actively and who performed late-night exercise for the first time.

## CONCLUSION

Nocturnal exhaustion exercise was found to significantly elevate serum potassium, sodium, magnesium, calcium, iron, zinc, manganese, nickel, selenium, molybdenum, chrome, cobalt, lead and cadmium levels in comparison to pre-exercise levels of these elements. Late-night exercise reduced only serum copper. The results we obtained regarding potassium, sodium, magnesium, calcium, iron, zinc, manganese, nickel, selenium, molybdenum, chrome, cobalt, lead and cadmium levels in nocturnal exercise are the first results regarding the relation between nighttime exercise and trace element metabolism.

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