EFFECTS OF EXERCISE TRAINING ON HUMAN COPPER AND ZINC NUTRITURE

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INTRODUCTION

Since 1969, there has been an increasing awareness of the potential value of physical activity as a means of reducing the morbidity and mortality associated with many chronic diseases, such as cardiorespiratory diseases, obesity and diabetes mellitus (Paffenbarger et al., 1986; Garrow, 1986; Holloszy et al., 1986). This awareness has increased participation in a variety of physical activities ranging from recreational pursuits to competitive athletics in children, adolescents and adults. Many participants are concerned with the role that nutrition may play in the development and the maintenance of performance. Unfortunately, authoritative information about the nutritional demands of chronic physical training is not readily available.

Attempts to determine the interaction between physical activity and nutrition can use either of two strategies. One approach is to study the physiological and performance responses to nutritional supplementation. Another strategy is to determine the effects of physical training on nutritional status. The most commonly used research design is to document changes in nutritional status during exercise training.

Information about the nutritional status of athletes during training is limited. Studies of individuals participating in physical conditioning generally focus on changes in energy intake (Short and Short, 1983; Brotherhood, 1984; Grandjean, 1985). There is a paucity of information describing the intake of micronutrients, especially copper and zinc, which are known to be important in energy metabolism (Underwood, 1977), and blood biochemical indices of copper and zinc nutritional status of athletes (Keen and Hackman, 1986; Campbell and Anderson, 1987). There are few reports integrating data on copper and zinc intakes and blood biochemical indices of trace element nutrition of athletes during training.

The purpose of this report is to describe the influence of intense physical training on the copper and zinc status of athletes. Three
studies are discussed that focus on the copper and zinc status, body composition, nutrient intake and blood biochemical indices of nutritional status, of collegiate swimmers. All participants were members of the men's and women's varsity swim teams at the University of North Dakota during the 1985-86 and 1986-87 competitive seasons. These findings are then compared with previously published data on copper and zinc status of athletes.

STUDY I: PRE- AND END-SEASON COMPARISONS

Twenty-one female swimmers aged 19-22 yr underwent body composition assessment, reported 7-day, self-selected dietary intakes and had venous blood samples taken before and at the end of the competitive season (Lukaski et al., 1989). Because nine swimmers either regularly consumed nutritional supplements or used oral contraceptive agents, their data were excluded and only data from 12 swimmers were analyzed. Although body weight changed slightly during the five months of training from 62.1 ± 1.7 (mean ± SE) to 62.7 ± 1.9 kg, fat free mass increased (48.7 ± 1.3 vs 51.1 ± 1.3 kg; p < 0.01) and fat mass decreased (13.4 ± 0.4 vs 11.5 ± 0.3 kg; p < 0.05). Swim training was associated with a 10% (p < 0.05) increase in energy intake (2030 vs 2269 kcal/d) that was attributable to an increase in carbohydrate intake (271 ± 20 vs 334 ± 17 g/d; p < 0.05).

Estimated daily copper (1.1 ± 0.1 and 1.0 ± 0.1 mg/d) and zinc (10.2 ± 0.7 and 9.7 ± 0.8 mg/d) did not change significantly from the start to the end of swim training, but they did show a trend to decrease slightly over time. When these trace element intakes are expressed per 2000 kcal, it appears that nutrient density for copper (1.1 vs 0.9 mg/2000 kcal) and zinc (10 vs 8.6 mg/2000 kcal) declined from pre- to end-season.

To estimate the adequacy of the swimmers' trace element intakes, the average calculated intake can be related to a general guideline representing 67% of the level recommended for the American public (Committee on Dietary Allowances, 1980). Using these terms, zinc intake (10.2 and 9.7 mg/d) approximates the guideline of about 10 mg/d, but copper intake (1.1 and 1.0 mg/d) is low based upon the guideline value of 1.3 mg/d, an estimated safe and adequate intake for copper (Committee on Dietary Allowances, 1980).

Plasma copper (95 ± 11 vs 94 ± 10 µg/dl) and zinc (81 ± 2 vs 77 ± 3 µg/dl) were unchanged with training and were within the ranges of normal values (copper: 70-140 µg/dl; zinc: 70-120 µg/dl) for females. The measured plasma concentrations of enzymatic (419 ± 37 vs 397 ± 36 mg/L) and immunoreactive (283 ± 20 vs 258 ± 29 mg/L) ceruloplasmin, and the ratio of enzymatic to immunoreactive (1.48 vs 1.54) ceruloplasmin, a proposed index of human copper status (Lukaski et al., 1983), were also unchanged. However, the activity of superoxide dismutase (SOD) a copper-containing enzyme, in red blood cells was elevated (4193 ± 170 vs 5139 ± 183 Units/g Hgb; p < 0.004) after training.

In retrospect, each of the 12 participating swimmers gained first, second or third place in either an individual or a relay event at the North Central Conference Women's Swimming Meet that was won by their team. Also, six of these swimmers earned All-American status at the Division II National Championships.

Therefore, the findings of this preliminary study suggest that physical training does not adversely affect copper and zinc status when intakes approximate recommended intakes. Also erythrocyte superoxide dismutase activity apparently increases in response to intense physical conditioning.
Study II: Comparisons Among Swimmers and Controls

A second study (Lukaski et al., in review) was undertaken among female (n=16) and male (n=13) swimmers and among age-matched, nontraining female (n=13) and male (n=15) controls. The purpose of this investigation was to determine the reproducibility of our preliminary results.

Body composition variables did not change significantly over time in either the swimmers or the controls. Among the swimmers, however, the females gained about 1.6 kg of fat-free mass and the males lost about 1.5 kg of fat mass.

Swim training was associated with increased energy intake. The female (2193 ± 121 vs 2329 ± 167 kcal/d; p <0.01) and male swimmers (3342 ± 176 vs 3717 ± 200 kcal/d; p <0.01) increased their daily caloric intake over time. This change was accomplished by increasing (p <0.05) daily protein (74 ± 3 vs 86 ± 4 g and 110 ± 4 vs 132 ± 4 g) and carbohydrate (285 ± 20 vs 320 ± 18 g and 429 ± 24 vs 480 ± 25 g) intakes in the female and male swimmers, respectively. Among the controls, daily energy intake was unchanged for the women (2077 ± 160 vs 2053 ± 163 kcal) and the men (3231 ± 165 vs 3194 ± 134 kcal).

Copper intake was similar before the start and at the end of the season among female swimmers (1.3 ± 0.1 and 1.4 ± 0.1 mg/d) and controls (1.2 ± 0.1 and 1.1 ± 0.1 mg/d), it did not change over time. Similarly, zinc intake was not different between female swimmers (10.4 ± 0.8 and 10.4 ± 0.9 µg/d) and controls (9.8 ± 0.9 and 9.6 ± 0.8 µg/d), and it was unchanged over time. In contrast, copper (1.8 ± 0.2 and 1.6 ± 0.1 mg/d) and zinc (15.2 ± 1.0 and 13.6 ± 0.7 mg/d) intakes decreased (p <0.05) in male controls and increased (p <0.05) in male swimmers (copper: 1.6 ± 0.1 and 1.9 ± 0.1 mg/d; zinc: 15.6 ± 0.8 and 17.9 ± 1.0 mg/d) over time. When expressed per 2000 kcal, the nutrient density of the self-selected diets was similar among all groups and over time; it ranged from 1.0-1.2 mg/2000 kcal for copper and 8.2-9.6 mg zinc/2000 kcal.

Relative to the guideline of 67% of the proposed intake for the American public (Committee on Dietary Allowances, 1980), the female swimmers met or exceeded the target intake value for copper (1.3 mg/d) and zinc (10 mg/d), but the female controls did not attain these standards for copper and zinc. Both the male swimmers and controls exceeded the guidelines for copper and for zinc.

Plasma copper concentrations were similar between female controls and swimmers before (94 ± 6 vs 100 ± 9 µg/dl) and at the end of the season (88 ± 5 vs 102 ± 5 µg/dl), and between male controls and swimmers before (91 ± 3 vs 84 ± 3 µg/dl) and after six months (90 ± 3 vs 85 ± 3 µg/dl). There was no change in plasma copper concentration either over time or after training.

Similarly, no differences were observed in plasma zinc concentration between female controls and swimmers before (86 ± 4 vs 83 ± 3 µg/dl) and at the end of the season (84 ± 3 vs 82 ± 3 µg/dl), and between male controls and swimmers before (87 ± 3 vs 90 ± 3 µg/dl) and after training (85 ± 3 vs 94 ± 3 µg/dl). Plasma zinc concentrations did not change over time in either group from the start to the end of the season.

All plasma copper and zinc concentrations were within the range of normal values. For Grand Forks, North Dakota residents, the range of normal plasma copper concentrations is 70-140 µg/dl and for plasma zinc concentrations is 70-120 µg/dl.
Copper status was also assessed using ceruloplasmin concentrations. For females, enzymatic ceruloplasmin concentrations were similar between controls and swimmers in preseason \((39 \pm 44 \text{ vs } 457 \pm 49 \text{ mg/L})\) and at the end of the season \((46 \pm 45 \text{ vs } 42 \pm 32 \text{ mg/L})\). A similar pattern was noted for immunoreactive ceruloplasmin in the female controls and swimmers before the start of the season \((266 \pm 14 \text{ vs } 280 \pm 30 \text{ mg/L})\) and at the end of the season \((257 \pm 13 \text{ vs } 291 \pm 26 \text{ mg/L})\). For the male groups, enzymatic ceruloplasmin was not different for controls and swimmers in preseason \((382 \pm 21 \text{ vs } 425 \pm 19 \text{ mg/L})\) and at the end of season \((367 \pm 29 \text{ vs } 446 \pm 17 \text{ mg/L})\). Also, immunoreactive ceruloplasmin was similar before \((245 \pm 13 \text{ vs } 259 \pm 11 \text{ mg/L})\) and at the end of season \((244 \pm 10 \text{ vs } 245 \pm 10 \text{ mg/L})\) for controls and swimmers. The ratio of enzymatic to immunoreactive ceruloplasmin was unaffected by group and by time; it ranged from 1.7–1.8 in the females and from 1.5–1.6 in the males.

Erythrocyte superoxide dismutase activity was significantly \((p < 0.001)\) affected by an interaction between group and time. Both female and male controls had values less \((p < 0.01)\) than swimmers of the same sex. The important factor influencing this difference was the large increase \((p < 0.01)\) in the red blood cell superoxide dismutase activity from preseason to the end of training in the female \((2627 \pm 140 \text{ vs } 4193 \pm 124 \text{ Units/g Hgb})\) and the male \((2541 \pm 150 \text{ vs } 3336 \pm 113 \text{ Units/g Hgb})\) swimmers in comparison to the female \((2479 \pm 128 \text{ vs } 2282 \pm 120 \text{ Units/g Hgb})\) and the male \((2522 \pm 137 \text{ vs } 2377 \pm 102 \text{ Units/g Hgb})\) controls.

The athletic success of the swimmers participating in this project is noteworthy. Twelve of the 16 female and 10 of the 13 male swimmers scored points in the conference team competition that was won by the University of North Dakota's men's and women's teams. In addition, six of the women and three of the men were awarded All-American status at the national championship meet.

The results of this controlled study confirm that erythrocyte superoxide dismutase activity increases after physical training in the presence of apparently constant and adequate copper intake.

**Study III: Effects of Oral Contraceptive Agents on Copper and Zinc Status During Swim Training**

During Studies I and II, it was apparent that a number of the female controls and swimmers were using oral contraceptives. It is well recognized that estrogen-containing oral contraceptive preparations influence blood biochemical indices of copper status (Prema et al., 1980; Vir and Love, 1981). We sought to determine the interaction between oral contraceptive usage, physical training and copper and zinc status.

Five female controls and six swimmers who reported using an estrogen-progesterone combination preparation at least six months prior to the start of Study II and who planned to continue its use throughout the next six months were studied. Body composition, nutrient intakes, and blood biochemical indicators of copper and zinc status were determined and compared to similar observations in female nonswimmers \((n=13)\) and swimmers \((n=16)\) not using oral contraceptives.

Body weight was stable in all groups of women during the six months of this study. Among the nontraining women, fat free mass was unchanged from pre- to end-season in both the nonusers \((46.9 \pm 2.1 \text{ and } 46.2 \pm 2.3 \text{ kg})\) and contraceptive users \((44.5 \pm 2.3 \text{ and } 44.2 \pm 2.7 \text{ kg})\). After
training, however, fat free mass increased by 1.5 kg (4.2 ± 1.1 and 1.5 ± 1.3 kg) in swimmers not using contraceptives, and was unchanged (4.1 ± 1.4 and 4.6 ± 1.8 kg) in the swimmers using oral contraceptives. Body fat mass decreased slightly, about 0.5 kg, only in both groups of swimmers.

Energy intake increased only in each group of swimmers. Whereas daily energy intake did not change over time for the control women not using oral contraceptives (2077 ± 160 and 2053 ± 163 kcal) and those nontraining oral contraceptive users (2284 ± 160 and 2247 ± 170 kcal), it increased (p < 0.05) during training among the swimmers not using (2197 ± 123 and 2329 ± 167 kcal) and using (2118 ± 192 and 2465 ± 213 kcal) oral contraceptives. This increased energy intake was associated with an increase (p < 0.05) in protein (74 ± 3 vs 86 ± 4, and 76 ± 2 vs 84 ± 2 g/d) and carbohydrate (285 ± 20 vs 320 ± 18 g/d and 292 ± 20 vs 350 ± 21 g/d) in both groups of swimmers.

Copper intakes were similar among all groups of women, and ranged from 1.1–1.5 mg/d. There was a slight trend of a decreased daily copper intake in the controls using oral contraceptives (1.4–1.1 mg/d) and swimmers (1.3–1.2 mg/d). Zinc intake averaged about 10 mg/d for all groups throughout the study. These estimated intakes of copper and zinc approached or met the dietary guidelines (Committee on Dietary Allowances, 1980) of 1.3 mg copper/d and 10 mg zinc/d.

As expected, plasma copper concentrations were about 30% greater in the oral steroid contraceptive users. Among the female controls and swimmers who did not use oral contraceptive agents, copper concentrations were 94 ± 6 and 100 ± 9 μg/dl before the season and 88 ± 5 and 102 ± 8 μg/dl after the season. In contrast, control and swimmer contraceptive users had plasma copper concentrations of 149 ± 6 and 151 ± 7 μg/dl before and 147 ± 5 and 148 ± 6 μg/dl at the end of the season.

Both chemical forms of ceruloplasmin were also elevated in the oral contraceptive users. On the average, enzymatic and immunoreactive ceruloplasmin values were 20–25% and 30–35% greater, respectively, in the control and swimmer contraceptive user groups. All values were within the range of normal values for women (200–400 mg/L).

Plasma zinc concentrations were remarkably similar in each group of volunteers. Values among controls not using contraceptives (87 ± 6 and 80 ± 7 μg/dl) and swimmers (82 ± 8 and 82 ± 6 μg/dl) did not change during the study. Similarly, plasma zinc was unchanged among controls using oral contraceptives (83 ± 5 and 86 ± μg/dl) and swimmers (81 ± 6 and 85 ± 4 μg/dl).

Despite the apparent differences in blood biochemical indices of copper status among the nonusers and users of oral contraceptives, red blood cell superoxide dismutase activity increased (p < 0.05) only in the swimmers after training. For the nonuser swimmers, it changed from 2455 ± 210 to 4150 ± 160 Units/g Hgb, and for the swimmers using oral contraceptives it also increased (2820 ± 220 vs 3990 ± 170 Units/g Hgb).

Among the six female swimmers using oral contraceptive agents, four secured first, second or third place at the conference meet and three earned All-American status at the national championship. Thus, oral contraceptive use per se apparently neither impairs athletic performance nor interferes with training-induced changes in erythrocyte superoxide dismutase activity.
Superoxide Dismutase Activity, Free Radicals and Exercise

Superoxide dismutase is a copper- and zinc-containing enzyme located in the cytosol of mammalian cells; its activity is dependent upon copper and its conformation requires zinc (McCord and Fridovich, 1969). A physiological function of superoxide dismutase is to scavenge and eliminate free oxygen radicals. Free radicals such as the superoxide anion are produced as a result of normal cellular oxidative metabolism. Increased amounts of superoxide radicals are associated with increased lipid peroxidation and accumulation of metabolites that can lead to cellular injury and death (Leibovitz and Siegel, 1980). An important mechanism that controls cellular damage caused by free oxygen radicals is superoxide dismutase that reduces the superoxide anion to peroxide which can be removed by catalase (Marklund, 1986).

Intense exercise has been reported to increase free radicals in rodent muscle and liver (Davies et al., 1982). Recent evidence indicates that free radical production in skeletal muscle of rats is directly related to exercise intensity (Alessio et al., 1988). Data on lipid peroxidation by free radicals in human muscle are limited, but indicate that stressful and prolonged exercise is associated with increased blood thiobarbituric acid reactive substance, an indirect measure of lipid peroxidation (Kanter et al., 1986). In addition, superoxide dismutase activity from human skeletal muscle was significantly correlated with peak oxygen uptake (Jenkins et al., 1984). Thus, increased superoxide dismutase activity may be an enzymatic adaptation to aerobic training that is protective against oxidative damage to tissue.

Exercise and Copper and Zinc Status

Attempts to determine the influence of exercise training on human copper and zinc status either describe serum or plasma trace element concentrations before and after training or compare circulating trace element concentrations in athletes to nontraining controls.

There are few reports of copper status among athletes. Dowdy and Burt (1980) studied male, collegiate swimmers during a six month training period and reported a significant decrease in ceruloplasmin (370 to 250 mg/L). Serum copper also declined from 64 to 50 µg/dl.

In contrast, other studies report no differences in blood biochemical measurements of copper status. Haralambie and Keul (1970) observed no difference in serum copper between male athletes and sedentary controls (94 vs 91 µg/dl). Similarly, Lukaski et al. (1983) reported no difference in plasma copper concentrations between male athletes and age-matched controls (90 vs 81 µg/dl). These reports are consistent with observations in male runners (Dressendorfer and Sookolov, 1980).

There are also conflicting data relating zinc status to physical activity. Dressendorfer and Sookolov (1980) reported hypozincemia in male long-distance runners in comparison to sedentary men (76 vs 94 µg/dl). Haralambie (1981) evaluated serum zinc concentrations in 57 female and 103 male athletes and observed that 43% of the women and 23% of the men had values less than 75 µg/dl. Deuster et al. (1986) reported that 25% of 51 female runners had serum zinc concentrations less than 80 µg/dl.

Anderson and his colleagues (1984) measured serum zinc and copper concentrations in male runners before and after a six mile run. Serum zinc did not change from pre- to post-run (81 vs 85 µg/dl), but decreased
(p <0.05) to 75 μg/d, two hours after the run. In contrast, serum copper was unchanged at these same samplings (93, 95 and 94 μg/dl). Comparisons between the urinary excretion of trace elements on the day of running and the day of no running indicated an increase (p <0.05) in daily urinary zinc loss after running.

Any interpretation of blood biochemical indices of trace element nutritional status requires information about intake of these nutrients. Unfortunately, the majority of the studies described above did not provide data on estimates of copper and zinc intake. Thus, it is difficult to make conclusions as to the physiological significance of the observed changes or differences in circulating trace element concentrations because it is unknown if the observations can be attributed to dietary intake or adaptation to exercise training.

SUMMARY AND CONCLUSION

The findings of our three studies indicate no significant decreases in blood biochemical indices of copper and zinc nutritional status among female and male swimmers during physical training when dietary intakes of these trace elements are adequate (≥ 67% recommended safe and adequate intake). At these intakes, erythrocyte superoxide dismutase activity increased after physical training indicating a biochemical adaptation of human copper metabolism associated with exercise training. It is noteworthy that this increase in superoxide dismutase activity occurred without an apparent increase in daily copper intake. The calculated copper intakes were 1.3–1.4 and 1.6–1.9 mg/d in the female and male swimmers, respectively. These intakes are less than the suggested value of 2.0 mg/d (Committee on Dietary Allowances, 1980), but they are similar to the reported intakes of other groups in the United States (Klevay et al., 1979).

In conclusion, these findings do not appear to support the belief that physical training per se produces adverse effects on copper and zinc nutrure.

REFERENCES


