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Effect of Zinc Supplements on Preventing Upper Respiratory Infections in Air Force Academy Cadets in Basic Training

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14. ABSTRACT

Zinc plays a vital role in the functioning of the immune system. However, under stress, the presence of zinc is reduced. As a result, the immune system is less effective, and the body is more susceptible to illness. In military training and operations, oftentimes members of the Armed Forces are subjected to high levels of stress which can increase the susceptibility of soldiers to illness. To investigate this effect in healthy young subjects, a two month randomized, double blind, placebo-controlled trial involving 57 cadets was implemented to evaluate zinc's effectiveness in reducing the risk of contracting upper respiratory infections (URIs). The primary objective for the study was to compare incidence of URIs between supplemented and non-supplemented groups. Post study data revealed no significant differences (p=0.53) between groups in terms of physician reported URI cases. However, the data suggest that stress may have played a key role as expected. Both mean plasma zinc levels decreased over time, albeit nonsignificantly, in both groups (p=0.20). However, there was a greater decrease in zinc values (20%) in the placebo group over that seen in the zinc supplemented group (14%). While administration of 20 mg/day of zinc gluconate did not change the incidence of URIs between groups, supplemented participants appeared to have lost less plasma zinc than their nonsupplemented counterparts. Higher levels of zinc may be warranted to confer a protective effect under more challenging immunological conditions.

15. SUBJECT TERMS

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Summary

Background: As a dietary essential, zinc plays a number of important roles within the body. Although known primarily for its antioxidant function, zinc is also an important and integral part of the immune system. Zinc appears to be an important modulator for the production of immune cells as well as ensuring the proper action of various leukocytes such as neutrophils, monocytes, macrophages, and B and T lymphocytes. Early research has shown equivocal results in regards to use of zinc as a palliative treatment for the common cold. However, more recent work provides evidence that use of zinc as a prophylactic regimen may possibly provide better results in preventing colds and other upper respiratory infections. One study in particular found a significant 2/3 reduction in respiratory infections in subjects taking supplemental zinc on a daily basis for one year. Stress is another mitigating factor which can impact zinc nutriture. Additional work with military personnel undergoing sustained psychological and physiological training revealed greater than a 30% decrease in zinc plasma levels in the blood. *Purpose:* The objectives of this study were threefold: 1) to compare URI incidence between supplemented and non-supplemented groups; 2) to assess plasma levels of zinc and copper to evaluate subject compliance and mineral status; 3) to measure biomarkers of stress as a means of quantifying the acute phase response changes in the basic cadets. *Methods:* The research protocol was reviewed and approved by the USAF Academy Institutional Review Board on 13 Dec 2007 (FAC2008015H). Ninety-six participants from the USAFA cadet class of 2012 were recruited for a two month randomized, double blinded, placebo-controlled trial during their initial arrival into basic cadet training. Written informed consent was obtained and confidentiality of data was kept. Cadets were instructed to take one capsule (zinc or placebo), one time per day for sixty days. Pre and post blood samples were taken and recorded along with physician reported URIs. *Results:* 1) Differences in URI incidence between groups were nonsignificant at post study($p=0.21$), 2) Post study plasma levels (in µg/dL) of zinc (supplemented, 95.44 ± 21.9, 95% CI: 86.3 – 104.4 and non-supplemented, 95.1 ± 21.9, 95% CI: 88.8 – 101.5) and copper (supplemented, 123.3 ± 36.4, 95% CI: 108.3 – 138.4, and non-supplemented, 122.3 ± 37.6, 95% CI: 108.6 – 135.8) were found nonsignificant between groups, 3) Differences between groups for stress biomarkers (plasma cortisol, $p=0.20$ and white blood cells (WBCs), $p=0.68$) were also found nonsignificant at end of study. *Conclusions:* Administration of 20 mg/day of zinc (as zinc gluconate) did not appear to result in any change in incidence of respiratory infections (as measured by a medical provider) between groups. As expected cadet cortisol levels were high at onset of study in anticipation of the rigors of military training with a concomitant reduction in WBCs. Both groups saw a decrease in post study plasma zinc levels but these differences were not found significant. Higher levels of zinc may be warranted to confer a protective effect under more challenging immunological conditions.

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Introduction

Zinc is considered one of the essential micronutrients used by the human body. Although zinc fulfills a number of metabolic and physiological roles, interest concerning its action within the immune system has grown considerably in recent years. Specifically, zinc is critical for both aspects of innate as well as acquired immunity for host defense. This includes proper maturation and function of neutrophils, macrophages and natural killer cells requisite for the signaling pathways of nonspecific responses. Additionally, zinc is important for the initiation of both T and B lymphocytes via signaling mediators which incorporate this important mineral as part of specific immune system pathways (Shankar & Prasad, 1998). Some studies (Shankar & Prasad, 1998) have indicated that a deficiency in zinc predisposes individuals to increased risk of infectious diseases. Marginal zinc deficiencies present with problems of impaired taste and smell, night blindness, memory decrement, and immune impairment. Greater degrees of deficiency result in more severe symptoms such as diarrhea, pustular dermatitis, mental disturbances, alopecia, and frequent infections as a result of an even more compromised immune system (Walsh et al., 1994). Studies involving animal models have also reported growth and immune deficiencies as a result of low zinc diets (Pekarek et al., 1977; Coughlan et al., 1988; Kidd et al., 1994).

More recently research has focused on the use of zinc supplements in reducing the risk or ameliorating the effects of upper respiratory infections (URIs) such as the common cold or flu and according to the Merck Manual 18th ed., 2006, "The common cold is an acute, usually afebrile, self-reported viral infection involving upper respiratory symptoms, such as rhinorrhea, cough, and sore throat." (p. 1595). Observing the results of trials from zinc deficient diets (and the concomitant improvements in immunocompetence when fed zinc replete diets), medical researchers are interested in determining appropriate zinc levels to preclude the infectious state. Mossad et al. (1996) found the provision of zinc (in the form of zinc gluconate lozenges) to subjects experiencing the onset of cold symptoms significantly reduced the duration of illness by nearly half. A more recent meta-analysis of studies involving zinc supplementation and colds did not, however, find any significant reductions in cold symptom duration (Jackson et al., 2000). The discrepancy in findings may be in the manner in which supplementation is used. Zinc may have its best efficacy when used in a preventive fashion as opposed to using it to ameliorate cold and flu symptoms. According to Prasad et al. (2007) it may be more important to establish a higher level of circulating plasma zinc prior to an infectious state for best results. Prasad's group used elderly subjects who typically have lower circulating plasma zinc levels and divided them into placebo and test groups over a specified period of time. After one year, the test group had significantly lowered incidence of infectious upper respiratory cases over the placebo group. Interestingly, the test group also had significantly lower oxidative stress and inflammatory cytokine levels; both are known as key contributors to a number of degenerative disorders such as cardiovascular disease, cancer, immunological compromises and atherosclerosis. Among zinc's myriad of important roles is its function as an antioxidant. It may be plausible that zinc is working in a number of ways to exert its protective effects in the body.

Although aging results in lowered zinc intakes through the diet, it is possible that physical or psychological stress can also lower plasma zinc levels. Perceived mental stress or incurred physical stressors usually result in an acute phase response which predictably drive physiological changes in the body. According to some, these changes are nothing more than bodily compensations driving adaptations which allow for cellular repair and immune response[11].

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Yet the results of these stressors are sufficient to cause lowered zinc in the circulating plasma. Singh et al., 1991 reported significantly lowered zinc concentrations in men training as part of Special Forces unit. Researchers examined blood samples before and after a particular extreme bout of mental and physical training and observed a 33% reduction in zinc levels. Although some of the loss was attributed to sweat and urinary routes, investigators suggested that a considerable amount of zinc was sequestered by organs and tissues for use in mounting an immune response. To quantify stress levels in the basic cadets, plasma cortisol was measured at baseline and end of study. The glucocorticoids negatively influence immune function by disturbing the balance in T and B lymphocyte production (Khardori et al., 2007). It appears that certain T helper cells (Th_1) which up-regulate the cell mediated portion of the acquired response are suppressed. This greatly compromises host defense, especially against viral infection. It is important to measure the level of stress as a means of quantifying the appropriate amount of zinc. Inordinate levels of stress may result in greater demands for zinc intake and use by the body for fighting infection (Shankar et al., 1998).

Taking the aforementioned in consideration, it may be helpful for individuals to take zinc supplements in advance of anticipated circumstances which cause an inordinate lowering of zinc levels during critical immunological challenges. Here at USAFA, cadets are exposed to a number of stressful situations which could probably deplete zinc stores to less than optimal levels--from the basic cadets, experiencing the stresses of in processing and Jack's Valley training to cadets adapting to the academic and military rigors of academy life. Although a cause and effect relationship has not been established, it is interesting that cadets present with more than the usual number of cases of URIs during these stressful episodes. According to data provided by the 10th Medical Group, Population Health (SGM), there appears to be an increased incidence of URIs during certain times of the academic year. Specifically, there is a double or triple increase in cases diagnosed starting in the beginning of the cold and flu season (fall through spring) and more importantly, another several fold increase in diagnosed cases during Basic Cadet Training in the summer.

Based on earlier work (Prasad et al., 2007) that showed that zinc may be used in a prophylactic fashion to prevent infection, the purpose of this new study was to look at a different critical time phase (summer basic cadet training) to further evaluate the potential protective effects of zinc. Based on the limited data regarding the aforementioned and the need to understand and develop potential public health countermeasures during times of increased risk for URIs, this study investigated whether or not the zinc supplemented condition results in measurable decreases in reported URI cases. Using appropriate biomarkers (plasma zinc, copper, cortisol, WBCs) that were adequately detected and measured, this study provided insight into the role of zinc supplementation as a nutraceutical (as opposed to a dietary essential) towards maximizing unit readiness (deploying warfighters) through individual health.

Methods

We incorporated a randomized, double blind, placebo controlled design for this study. This study was approved by the United States Air Force Academy Institutional Review Board before subject recruitment began. All participants were briefed on the important elements of the protocol and read the informed consent document (ICD), which was approved by the USAFA Institutional Review Board on 13 Dec 07 (FAC2008015H). After each participant read the ICD, had all questions answered and agreed to all the conditions, the participant signed the ICD.

Statistical Analyses.

The purpose of this study was to compare URI infection rates in 45 zinc supplemented versus 45 non-supplemented cadets. A total sample size of 90 was derived based on an assumption of a 50% reduction in URI incidence rates detected in the zinc supplemented group with a 2 sided p value of 0.05 and an approximate power of $\geq 95\%$ (calculation based on previous work and available power analysis software (Lenth 2007, Prasad et al., 2007). However, in consideration of drop-out rates as experienced in previous studies similar to this work, we requested approval to recruit 100 subjects in order to retain power for detecting differences. Demographic characteristics between treatment and control groups were evaluated by t test and chi square test (gender, supplement use, pre-existing condition, medical care and medication use). For other variables, t tests were used to compare group differences. Chi square tests were used to compare incidence of URIs in both groups. Measures over time were examined with a multivariate ANOVA repeated measures analyses. Statistical analyses were accomplished with JMP software (version 7; SAS Institute Inc) run on a Fujitsu Lifebook tablet computer (Fujitsu Computer Systems, Sunnyvale, CA). Any mention of statistical significance refers to an alpha level of .05.

Experimental Procedures.

This study included both male and female cadets. We excluded volunteers currently under medical supervision, having a history of chronic illness (Crohn's Disease, Irritable Bowel Syndrome, Acrodermatitis Enteropathica or Congenital Zinc Deficiency), or currently self-supplementing with zinc or a multivitamin. During the recruitment phase, potential subjects were screened for study inclusion using a survey. We randomly assigned subjects to groups which either received a zinc supplement or placebo in a blinded fashion. Practitioners or individuals involved in drawing blood or running laboratory analyses were also blinded to the assignment. In the summer of 2008, at the beginning of basic cadet training, subjects ingested one capsule (either zinc or placebo) each day in the morning, taken before eating breakfast for a period of two months. Both zinc (20 mg per capsule) and placebo capsules (same capsule type as used for the zinc except filled with cornstarch or gelatin) were provided in a 30 day supply container through an FDA licensed clinical specialty pharmacy (Axium Healthcare Pharmacy, Lake Mary, FL). The zinc supplement also underwent an independent lab analysis (DynaLabs, St Louis, MO) to certify the content and quantity of zinc within the supplement capsules. While there is a minor risk associated with taking a zinc supplement over and above dietary intake, the risk was considered minimal because total zinc intake (supplement and dietary intake) per day was

estimated to be below recommended upper limit thresholds. According to the Institutes of Medicine (IOM) 2001, Dietary References Intake Report, the upper limit (UL) threshold for zinc intake should not exceed 40 mg/day for both men and women. The Recommended Dietary Allowance (RDA) for zinc is 8 mg/day for women and 11 mg/day for men. Median zinc intake from food in the United States is approximately 9 mg/day for females and for males, it is 14 mg/day. The zinc supplement capsules provided 15 mg of zinc per day which resulted in daily zinc levels beyond RDA levels but below the tolerable upper limits as directed by the IOM. Zinc is relatively nontoxic and only causes problems in terms of copper deficiency at levels exceeding 50 mg/day for periods of time exceeding 3 months (King & Keen, 1999). In fact, one recent study fed subjects 45 mg zinc per day for a one year period without any ill effects reported (Prasad et al., 2007).

All subjects provided blood samples obtained pre and post study by trained laboratory personnel in the 10th Medical Group Cadet Clinic. Samples were collected at baseline (26 June) and end of study (25 August) time points for analyses of plasma zinc, copper, cortisol and WBC count. Plasma biomarkers are required to validate subject compliance as well as to ensure that zinc does not interfere with copper absorption. Tracking cadet cortisol and WBC count provided insight to the extent that stress impacted basic cadets as they experienced basic cadet training. Subjects observed regular procedures whenever ill in seeking medical intervention and had their diagnosed URI cases tracked by a medical provider data base.

Laboratory Analysis

Blood analyses were accomplished at the United States Air Force School of Aerospace Medicine, Chemistry Division at Brooks City Base, Texas. Plasma zinc was assayed by methods that used inductively coupled plasma (ICP)-emission spectrometry (PERKIN ELMER Optima 3000 DV, Dual View ICP Optical Emission Spectrometer); samples were diluted with zinc-free water before analysis. Reference standards included High Purity Standards, SM-12-92-009 and UTAK Serum TDM/Toxicology Control, 66816. Plasma copper was also analyzed by inductively coupled plasma (ICP)-emission spectrometry.

Results

Although subjects were given zinc supplements well within recommended upper level dietary allowances, each subject was instructed to immediately advise investigators if they experienced any unusual symptoms not associated with a diagnosed illness (loss of appetite or headache). While we did not have any adverse events reported, we experienced a high drop out for this study due to disenrollments and lack of compliance issues. Of 96 subjects at onset of study, 39 dropped from the study through self-elimination (27 from the supplemented group and 12 from the non-supplemented group). In the text below, a general description of the results for key variables are provided, followed by detailed discussion and graphs of selected measures.

Table 1: Demographics

Variable	Zinc Group	Placebo Group
Mean Age (years±SD)†	18.5±1.27	18.4±0.94
Sex Male	33	33
Female	16	14
Use of Supplements	0	0
Use of Medications	0	0
Under Medical Care	0	0
Any medical history§	0	0

† p=0.41 (for comparison of mean age)

§ any current or family history of digestive disorders

Limited demographic/characteristics information was collected for age and sex of participant during subject enrollment and can be found in Table 1. The mean age for all participants was 18.4 years (SD = ±1.12). The zinc supplemented group initially began with 49 subjects and the non-supplemented group started with 47 subjects with approximately equal numbers of males and females in each group. None of the subjects reported using any medications or supplements at the time of enrollment. Furthermore, no subjects claimed to have a significant medical history or to be under the care of a health care provider for a health concern.

Laboratory Analysis

A comparison of both baseline and post-study lab data between groups is shown in Table 2. Prior to treatment, the mean zinc blood levels for both the zinc group 111.24±20.2 (103.3 – 119.1) and placebo group 119.5±29.8 (109.2 – 129.9) did not significantly differ (p=0.21). Similarly, post study mean zinc values between groups did not differ significantly (p=0.95). Copper values also did not differ significantly between groups at baseline (p=0.45) nor post-study (p=0.90). None of the subjects' results were outside normal parameters for zinc at the beginning of the study. Normal range for zinc is 50 – 150 µg/dL and 64-156 µg/dL for copper (King, 1999 and Turnland, 1999). Within group analyses showed the placebo group experienced a more than 20 percent significant decrease in plasma zinc levels over the course of their training (p=0.00002). We, however, also unexpectedly observed a similar significant decrease (14 percent) in our zinc supplemented group from pre to post study (p=0.0005). Within group analyses for the remaining biomarkers (copper, Cortisol and WBCs) did not reveal any significant differences from pre to post study (copper: supplemented (p=0.59), non-supplemented (p=0.19), cortisol: supplemented (p=0.08), non-supplemented (p=0.18) except for WBCs which saw a significant drop in both supplemented and non-supplemented groups and (WBCs: supplemented (p=0.04) and non-supplemented (p=0.001)

Table 2:
Laboratory Analysis – Comparison of Biomarker Concentrations in the Plasma

Variable (µg/dL)	Zinc Group N=25	Placebo Group N=32	*p*‡
Zinc†			
Pre-study	111.24±20.2 (103.3 – 119.1)	119.5±29.8 (109.2 – 129.9)	0.21
Post-study	95.44±21.9 (86.3 – 104.4)	95.16±17.6 (88.8 – 101.5)	0.95
Copper†			
Pre-study	117.61±39.5 (102.1 – 133.1)	109.75±38.1 (96.5 – 122.9)	0.45
Post-study	123.40±36.4 (108.3 – 138.4)	122.2±37.6 (108.6 – 135.8)	0.90
Cortisol†			
Pre-study	15.91±5.6 (13.6 – 18.1)	17.43±6.3 (15.2 – 19.6)	0.33
Post-study	12.75±6.9 (10.05 – 15.4)	15.09±7.4 (12.4 – 17.7)	0.22
WBCs (x 10^3)†			
Pre-study	8.48±2.14 (7.6 – 9.4)	8.58±1.55 (8.04 – 9.12)	0.96
Post-study	7.62±2.2 (6.7 – 8.5)	7.13±1.8 (6.5 – 7.7)	0.51

† = all values reported as mean±SD (95% CI)

‡ = t-test analyses, two tail comparison, assuming ≠ variances

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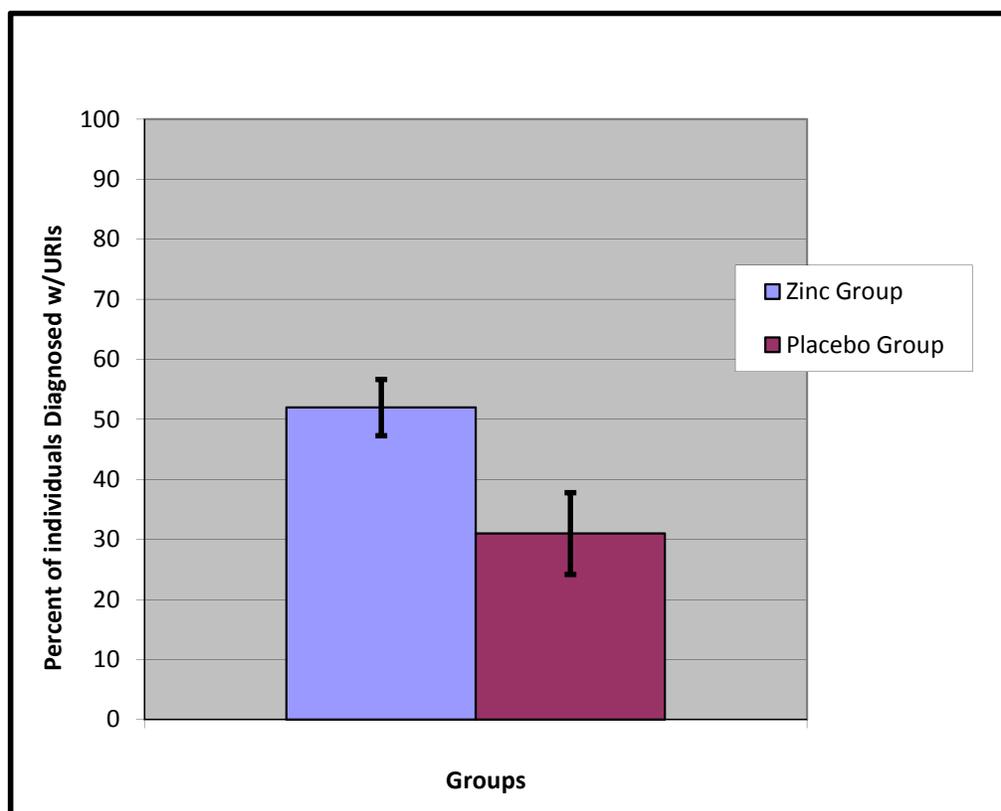
Figure 1: Incidence of physician reported URI cases by group

(X^2 test of proportions, nonsignificant at p=0.12)

Disease assessment

Based on physician diagnosis, the supplemented group had higher reported URI cases 13/25, 52%) than was recorded in the placebo group (10/32, 31%). Although the zinc group had higher reported cases, between group differences in terms of reported URIs were not significant (p=0.12).

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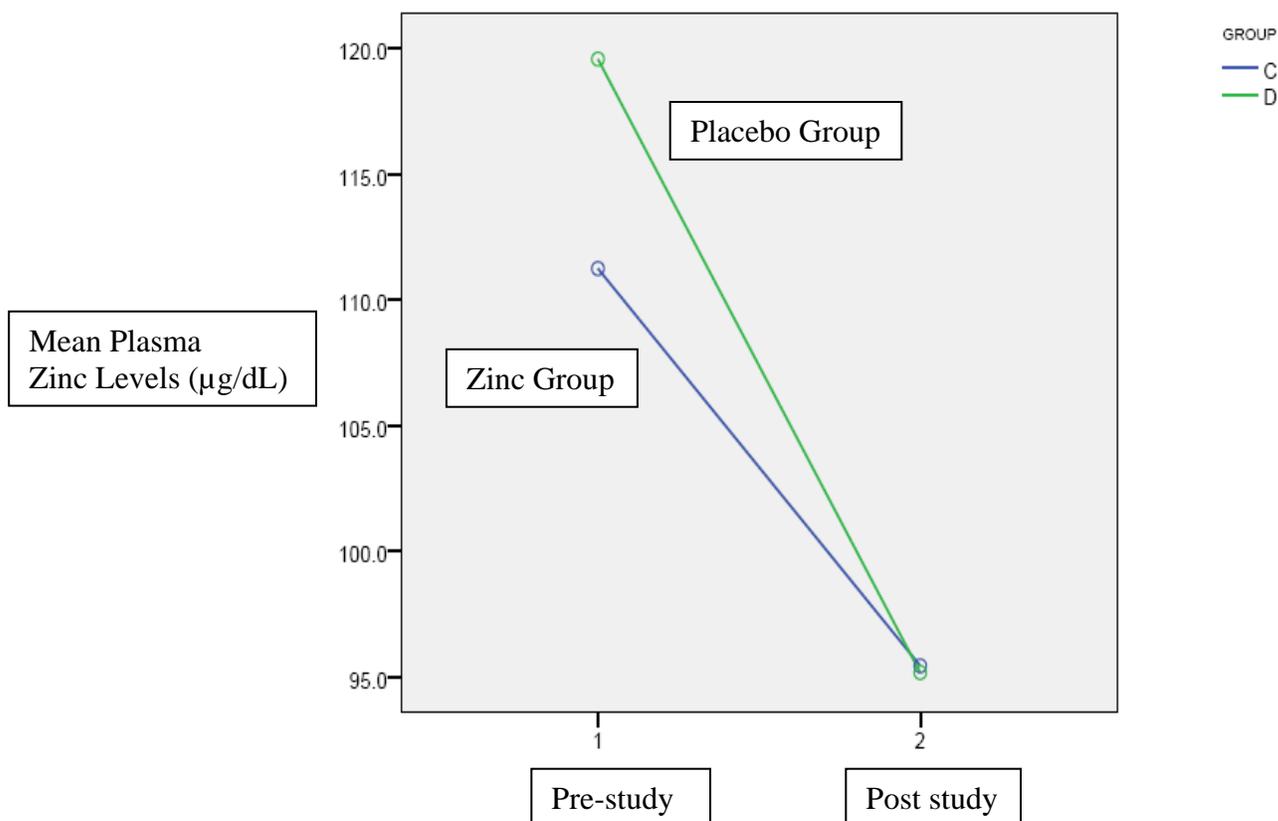
Figure 2: Effect of Zinc Supplementation on Plasma Zinc Concentrations
Mutivariate Repeated Measures Analyses for change in groups over time (p=0.20)

With time (two months), within group plasma zinc concentrations for each group decreased significantly (zinc, p=0.0004, placebo, p=0.00001) but no significant change was found between groups (p=0.95) nor any time by group interaction (p=0.20), figure 2.   Similar analyses (figures 3-5) of plasma copper, cortosol concentrations and WBC numbers by group revealed no significant differences, between groups (p=0.90), (p=0.22), (p=0.51) or time by group interaction (p=0.33), (p=0.57), (p=0.20) respectively.
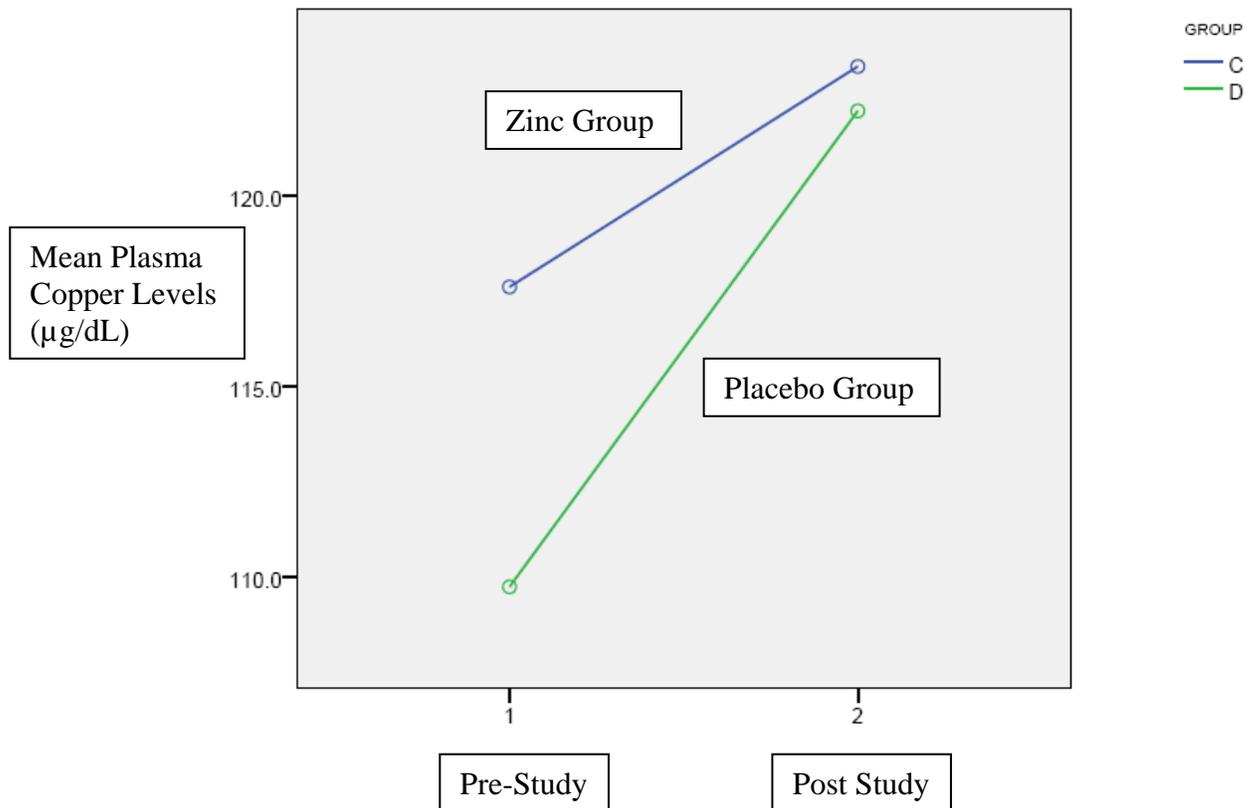
Figure 3: Effect of Zinc Supplementation on Plasma Copper Concentrations
Mutivariate Repeated Measures Analyses for change in groups over time (p=0.33)

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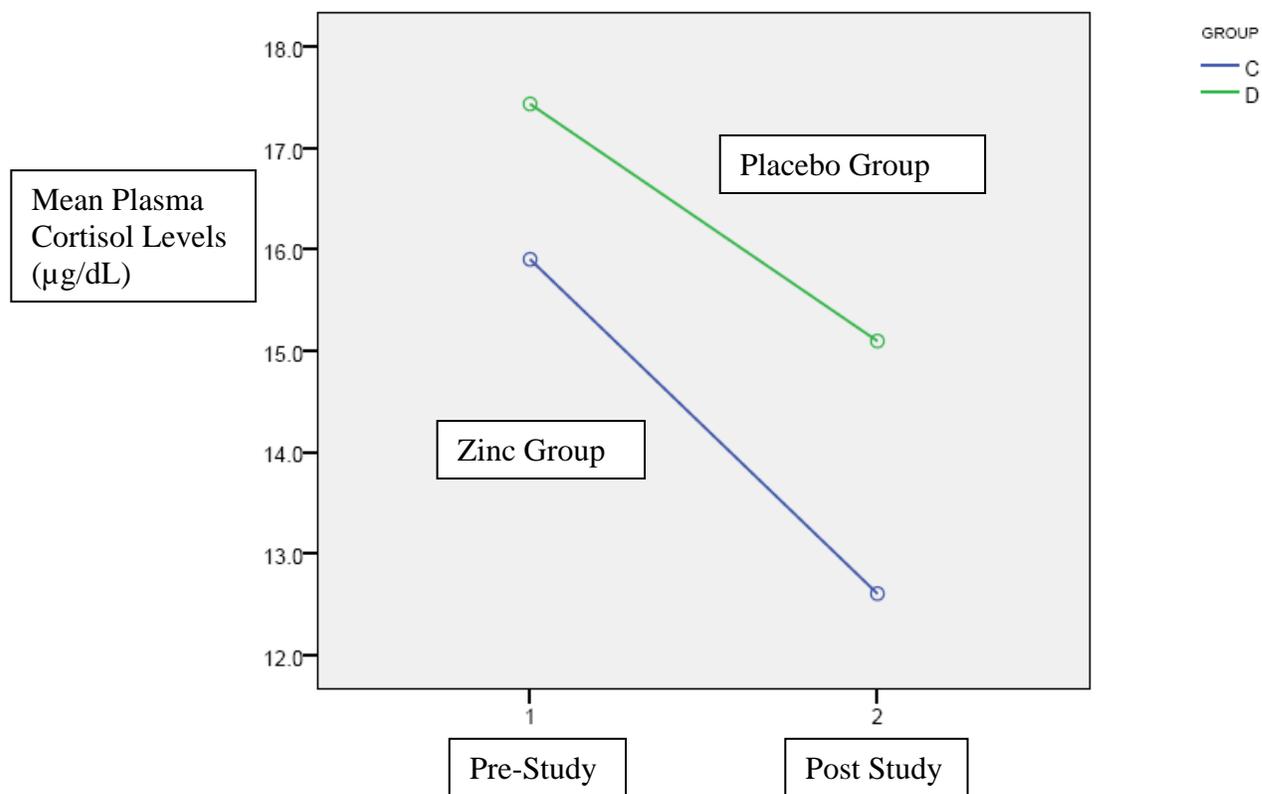
Figure 4: Changes in Plasma Cortisol Concentrations
Mutivariate Repeated Measures Analyses for change in groups over time (p=0.57)

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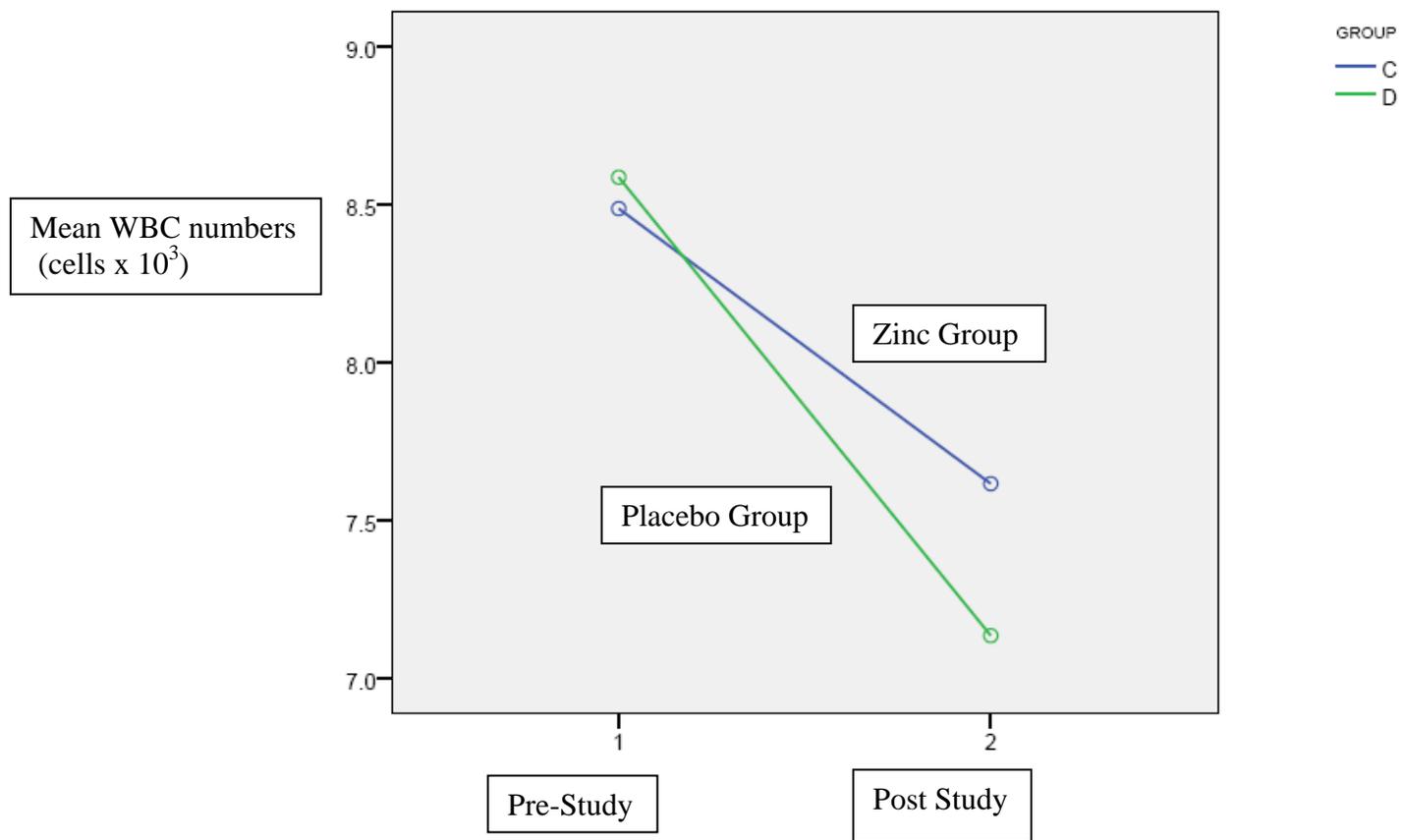
Figure 5: Changes in WBC numbers
Mutivariate Repeated Measures Analyses for change in groups over time (p=0.20)

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Discussion

The value of zinc to treat the duration and severity of colds has an ambiguous record. Generally zinc has been used during the onset of illness with equivocal results. We investigated whether zinc could be used in a prophylactic manner in anticipation of an impending immunological challenge (URIs) under stressful conditions. Our study did not find a prophylactic effect of zinc at 20 mg/day levels. Both groups had significant decreases in plasma zinc by end of study and although we observed higher numbers of physician reported URI cases within our supplemented group, the difference was not significant. While these results were not predicted, we did see some trends within the data that suggest that stress may have played an important role as expected. A previous study by Prasad et al., 2007 showed advance zinc ingestion can reduce risk for infections in elderly audiences and this spurred our efforts to work with younger populations. Although our study population presumably had a much higher degree of immunological competence than the elderly, the cadets experienced a much greater degree of stress than other populations (military training and academic demands). Stress appears to have an adverse effect on plasma zinc and can result in considerable loss under circumstances involving high physical and mental demands (Singh et al., 1991). We felt that although we were working with young healthy subjects, the mental and physical stress of basic cadet training would take its toll and result in subjects having less than desirable zinc nutriture for immunocompetence. Stress and the concomitant release of extended cortisol levels over time appear to have an immunomodulatory effect which may impact the manner in which an immune response is mounted (Khardori et al., 2007).

Although mean plasma zinc levels decreased over time in both groups (p=0.20), zinc values in the placebo group trended lower (-20%) than those in the zinc group (-14%) but not significantly. Zinc supplementation may have precluded a greater drop in the treatment group than that observed in the placebo group. We examined WBC numbers as the integral portion of the body's immune system for differences between groups at post study and did not find any significant differences. As with the zinc plasma values between groups, WBC numbers in the placebo group trended lower (-17%) than those in the zinc group (-10%) but not significantly. Keeping in account that both groups were exposed to the same stressful events (cortisol levels were not significantly different at both pre and post study), it is interesting to observe that while zinc and WBC numbers were not significantly different between groups, the trend to lose more zinc and WBCs occurred in the placebo group. The data suggest that the supplemented group lost fewer WBCs to stress than was observed with the non-supplemented group because more zinc was available in the supplemented group to build new WBCs. The blood acts as the most dynamic indicator for zinc nutriture status because of its accessibility to the white blood cells of the immune system. A highly exchangeable form of zinc exists between the plasma and the cellular component. Studies involving animal models fed increasing levels of zinc have reported varying levels of immune activity (Prasad, 1991). There may be certain discrete thresholds of zinc supplementation (above RDAs but below toxicity effects) required to reach heightened immune response. By extension, it may be possible we did not administer sufficient zinc to reduce URI incidence within this studied population.

In one of our previous zinc studies involving cadets, we also provided a self report instrument which allowed participants to submit their weekly health status anonymously over the web (the current study did not have sufficient time for participants to complete another web survey). Our group decided to include a self report instrument for that study in order to capture

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subjects with more moderate symptoms. Oftentimes individuals will not seek medical intervention until symptoms becomes more serious or uncomfortable. Using a previously validated disease assessment instrument for the common cold (Takkouche et al., 2001, 2002), our subjects reported a myriad of symptoms on a rated scale. Subjects also had the option of reporting zero symptoms at each weekly reporting period. At post study, we found that zinc supplemented individuals reported fewer URI symptoms throughout the trial than those in the placebo group. This was intriguing from the standpoint that although medically diagnosed cases were not different between groups, those self reporting their symptoms on a recurring basis appeared to have experienced some protective effect from the zinc. It may be that depending on the severity of symptoms, a varying level of zinc may be needed. As the immune system detects a threat, a graduated response is elicited in terms of leukocyte development. Zinc becomes very important in terms of meeting the maturation of lymphocytes, especially B and T lymphocytes (Shankar & Prasad, 1998). Greater numbers of developing cells translates into greater amounts of zinc required for growth and activation. For more virulent threats, higher levels of zinc may be warranted. In the earlier mentioned study by Prasad in 2007, 45 mg/d were provided to elderly subjects. Greater threats, even in younger populations, may still require higher than normal zinc intakes, especially those exceeding recommended upper limits (combining both supplement and dietary sources totaling > 40 mg/day). Limitations for this study included a higher than anticipated cadet disenrollment rate and some subject noncompliance.

Conclusions

Higher levels of zinc supplementation may need to be studied. Although the RDA remains at levels below current studied levels, there could be circumstances which justify supplementation on a limited basis. Clinically, there could be medical conditions which may compromise a patient's immunocompetence (cortisol injections, immunosuppressive regimens, old age, stress, burn victims, etc.). From a military standpoint, there appears to be considerable interest in using zinc supplementation prophylactically. A greater portion of our military personnel now deploy to various locations all over the world and face a myriad of diseases/illnesses, especially diarrhea, respiratory disorders, malaria and leishmaniasis. Both animal and human trials have reported the success and efficacy of using zinc supplements in controlling the aforementioned diseases (McClung & Scrimgeour, 2005). Of critical importance is determining the appropriate amount of zinc without causing deleterious effects at the immune or other physiological level.

Recommendations

We intend to pursue an additional investigation which will examine a higher level of zinc (30 mg/day) while continuing to quantify stress (with plasma cortisol, along with other established biomarkers such as C reactive protein) and leukocyte development. This higher level of ingested zinc may offer us a better insight whether zinc works in a prophylactic fashion or not. Eventually, this will aid us in advancing to other studies that will help derive the appropriate nutritional response for an immunological challenging condition.

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Appendix A
List of Symbols, Abbreviations and Acronyms

USAFA	United States Air Force Academy
URIs	Upper Respiratory Infections
ICD	Informed Consent Document
UL	Upper Limit
RDA	Recommended Dietary Allowance
±SD	Standard Deviation
X^2	Chi Square Test
WBCs	White Blood Cells

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