Biol Trace Elem Res
DOI 10.1007/s12011-015-0334-3

Moderately High Dose Zinc Gluconate or Zinc Glycinate: Effects on Plasma Zinc and Erythrocyte Superoxide Dismutase Activities in Young Adult Women

Robert A. DiSilvestro[1] · **Erin Koch**[1] · **Linda Rakes**[1]

Received: 8 January 2015 / Accepted: 6 April 2015

Abstract Some zinc (Zn) research studies have used either Zn gluconate or Zn glycinate, but the two forms have not been compared much. Therefore, a moderately high dose of the two forms (60 mg Zn/day) were compared in a 6-week intervention in young adult women. Plasma Zn, the traditional assessment of Zn status, was increased in all subjects given Zn glycinate (N=10), while no significant change was seen overall for Zn gluconate or placebo (N=10 each). Erythrocyte superoxide dismutase activity, a marker for Zn-induced copper deficiency, was unchanged in all three groups. Thus, for the conditions of this study, Zn glycinate effectively changed Zn status better than Zn gluconate, but neither impacted copper status.

Keywords Zinc · Chelates · Gluconate · Glycinate · Superoxide dismutase · Supplementation

Introduction

Marginal zinc (Zn) deficiency can occur in various circumstances [1, 2]. Correction of this problem can be accomplished by temporary use of a moderately high dose Zn supplement. Even in the absence of such deficiency, moderately high dose Zn supplements might also serve other purposes such as anti-inflammatory actions in certain situations [1, 3], increasing sperm counts in infertile men [4], and treating attention-deficit/hyperactivity disorder in children [5]. In addition, moderate to high dose Zn supplementation has been used in eye supplementation studies [6]. Although each of these effects is considered desirable, on the down side, moderately high dose Zn supplementation can cause a degree of copper deficiency [1].

A question that arises about Zn supplementation is as follows: What form Zn should be employed? At one time, Zn sulfate was often used for Zn supplements, but this use has fallen in recent years. One reason may be that Zn sulfate can produce some GI tract irritation in some people (i.e., 7, 8). In addition, for some consumers, an attitude has arisen that organic mineral complexes are preferred over inorganic forms. For organic Zn supplements, Zn gluconate has emerged as a commonly used version. However, this form has not been extensively evaluated in comparison to other organic Zn complexes. One such organic complex, Zn glycinate, has produced positive results in a number of research studies (i.e., 5, 9, 10). Thus, it can be asked whether either Zn glycinate or Zn gluconate works better than the other. In two acute uptake studies, Zn glycinate has performed better than Zn gluconate [11, 12], but sustained use of Zn glycinate has not been compared to sustained use of Zn gluconate.

A pilot study was carried out to compare moderately high dose supplementation of Zn gluconate to Zn glycinate. Two endpoints were employed. One was plasma Zn, the traditional assessment tool for Zn status [1]. The other was erythrocyte superoxide dismutase activity, a maker of Zn-induced copper deficiency, a possible negative consequence of moderately high dose Zn supplementation [13–15].

Materials and Methods

Subjects

The protocol was approved by The Ohio State University Human Subjects Biomedical Institutional Review Board.

✉ Robert A. DiSilvestro
disilvestro.1@osu.edu

[1] Human Nutrition, Ohio State University, 345 Campbell Hall, 1787 Neil Ave, Columbus, OH 43210, USA

Published online: 17 April 2015

Thirty females, 18–24 years old, were recruited from the student population at Ohio State University. Potential subjects were excluded for the following: major health problems, use of oral contraceptives, taking medications that affect Zn metabolism, or regular use of supplements containing Zn except at 15 mg/day or less as oxide in multi-type supplements (though such supplements were discontinued starting at least a week before study participation). Subjects were drawn from a group of 96 subjects who had given an initial blood draw that was tested for plasma zinc. Subjects with readings in the top 15 % or bottom 10 % of the values obtained were not used in this study.

Subjects were randomly assigned to either maltodextrin placebo (N=10) or 60 mg Zn per day as Zn gluconate (N=10) or Zn glycinate (N=10) from Albion Laboratories (Clearfield, UT, USA). The supplementation followed a double blind protocol. Subjects consumed the assigned product for 6 weeks. Supplements were taken with a meal of the subject's choosing. Subjects were given three extra doses and returned unused capsules. Blood samples were taken before and after the supplementation period. Blood samples were collected in the morning, at about the same time of day for each subject, after an overnight fast. Subjects were instructed to maintain their previous dietary and exercise practices during study participation.

Laboratory Analysis

Blood was collected into a tube with heparin. Each tube was centrifuged at 3000×g for 30 min to obtain plasma, which was stored at −10 °C until assayed for Zn by atomic absorbance spectrometry. In addition, erythrocytes were collected and washed with phosphate-buffered saline, extracted with ethanol:chloroform, stored at −10 °C, and later assayed for superoxide dismutase by a modified pyrogallol method as described previously [16].

Statistical Analysis

Significance was set at $p<0.05$. For each assessment, for each treatment group, pre-values were compared to post-values by paired t test using Excel (Microsoft Corp, Redmond, WA). Changes in values for Zn supplementation were compared to changes for placebo by unpaired t test by Excel. Pre-treatment values were compared between placebo and Zn supplementation groups by unpaired t test using Excel.

Results

As seen in Fig. 1, supplementation with Zn glycinate, but not Zn gluconate or placebo, increased plasma Zn (glycinate, 0.9 ±0.3 vs 1.5±0.3; gluconate, 1.1±0.3 vs 0.9±0.3; placebo, 0.9 ±0.2 vs 0.8±0.3; means±SD). As seen in Fig. 2, the change in plasma Zn due to Zn glycinate differed to a highly significant extent from the change due to placebo or Zn gluconate (glycinate, 0.7±0.2; gluconate, −0.2±0.2; placebo, −0.1± 0.1; means±SD). The Zn glycinate effect was very consistent as all subjects showed an increase. In contrast, six of the ten subjects in the gluconate group showed no change or a decrease. Since the latter group did not show a normal distribution, the glycinate and gluconate groups were also compared by a Wilcoxon rank-sum test. Again, the difference showed a high degree of significance.

None of the treatments produced any changes in erythrocyte superoxide dismutase activities (Table 1).

Discussion

Sustained supplementation with Zn glycinate supplementation was more effective at raising plasma Zn than supplementation with Zn gluconate. This result was consistent with two previous acute comparisons [11, 12]. On the other hand, the observation of no statically significant increase in plasma Zn after Zn gluconate supplementation does not resemble certain other studies. In a number of studies, sustained Zn gluconate supplementation has raised serum or plasma Zn readings (i.e., 13, 14, 17, 18). On the other hand, one study examining supplementation of 50 mg Zn/day reports no rise in serum Zn at 6 or 8 weeks [19]. Similarly, an unpublished study from this laboratory found that 6 weeks of supplementation with 30 mg of Zn as gluconate did not raise plasma Zn values in healthy middle-aged people (DiSilvestro RA, unpublished results). Moreover, in an acute study of Zn gluconate supplementation [20], after an initial rise in blood Zn, fecal loses eventually produced an actual negative Zn balance. Thus, some variables in subject traits or background diet may determine the degree to which Zn gluconate supplementation affects Zn status. Although it remains unclear as to why Zn gluconate gives some inconsistent results, a consistent trend has appeared for three comparisons of Zn gluconate versus Zn glycinate. In each case, the results have favored Zn glycinate.

The increases in plasma zinc in response to zinc glycinate were substantial (a mean increase well above a 50 % increase). This large change could point to the especially high efficacy of zinc glycinate versus other forms of zinc. However, the type of subjects could also explain the large reaction. This type of subject may not eat a lot of bioavailable zinc. Also, as noted in the "Materials and Methods," subjects were drawn from 96 potential subjects with those in the highest 15 % for plasma zinc excluded.

The lack of effect of Zn gluconate on plasma Zn readings does not necessarily mean that no impact on Zn status occurred. Plasma Zn does not respond to changes in Zn status as easily as some other markers [1, 2]. However, the plasma

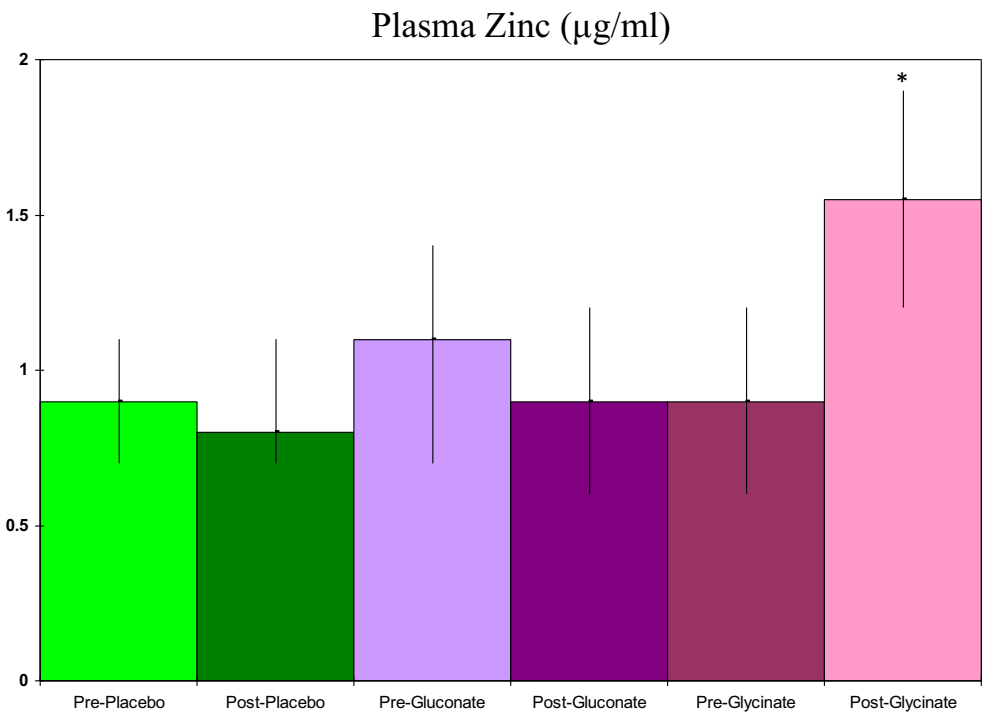

Fig. 1 Plasma zinc (µg/ml) before and after supplementation with placebo, zinc gluconate, or zinc glycinate. Values are means± SD. *$p<0.001$, paired t test

Zn results in the present study indicate a stronger response to Zn glycinate, at least for the conditions of this study.

The current study's lack of a Zn effect on copper status differs from some previous studies that employed 50 mg zinc/day and used erythrocyte superoxide dismutase activities for a marker [13–15]. One possible explanation is that the present study subjects' copper status was so good as to render them resistant to zinc-induced changes in copper status. However, the erythrocyte superoxide dismutase activities do not support this proposition. The mean values were similar or less than those in previous studies from this research group that show increases with copper supplementation [21–23]. In fact, within two of these previous studies [22, 23], starting activities predicted whether copper supplementation would have an effect. The activities in the present study would predict that the subjects tended to be in sub-maximal copper

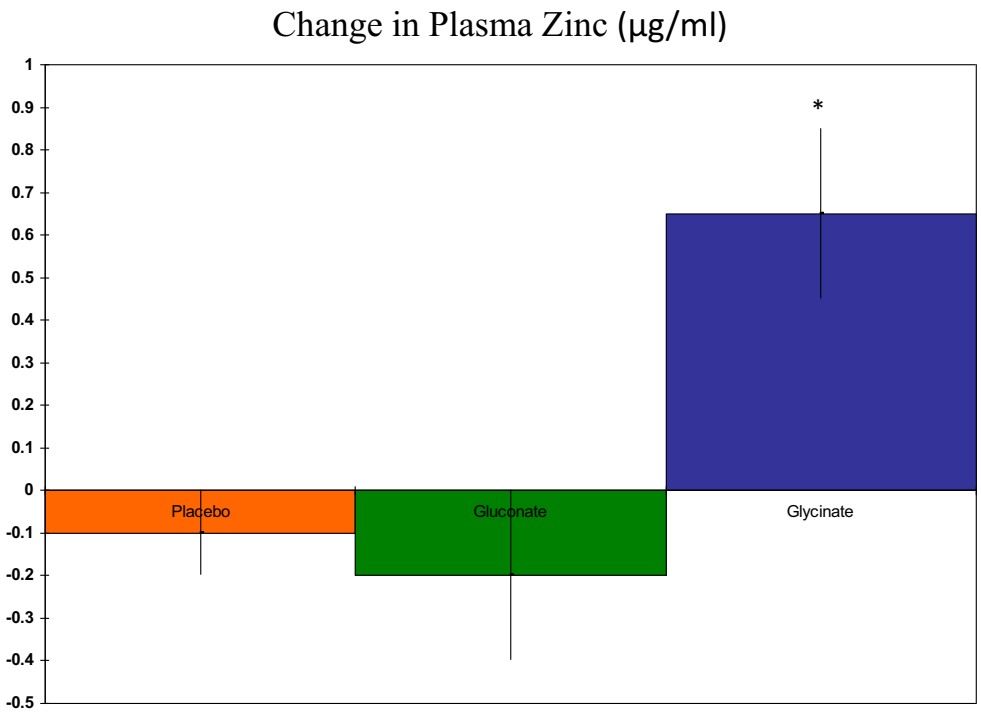

Fig. 2 Change in plasma zinc (µg/ml) pre- minus post-supplementation with placebo, zinc gluconate, or zinc glycinate. Values are means±SD. *$p<0.001$ versus placebo or gluconate, unpaired t test and Wilcoxon rank-sum test

Table 1 Superoxide dismutase activities

Group	Pre-treatment	Post-treatment
Placebo	3363±462	3389±450
Zinc gluconate	3348±581	3301±499
Zinc glycinate	3425±602	3408±622

Means in units per milliliter packed cells±SD. No significant changes in any group between pre- and post-treatment (paired *t* test, $p>0.05$)

functional status (based on the ability of copper supplementation to increase erythrocyte superoxide dismutase activities).

Two of the studies showing a zinc supplement-induced depression in erythrocyte superoxide dismutase activities [13, 14] have conditions that overlap the current study in terms of supplementation length (6 weeks) and the use of Zn gluconate. However, in both of these previous studies, Zn supplementation raises plasma or serum Zn, which was not the case in the present study. Thus, the amount of Zn that actually enters the body may affect whether copper status is affected. Still, this may not be the only factor since in the present study, Zn glycinate raised plasma Zn without affecting erythrocyte superoxide dismutase activities. Possibly, in the small intestine, amino acid chelated Zn absorbs with protein digestion products in a way that does not compete with copper absorption. Such competition is thought to provide the basis for Zn-induced copper deficiency [1]. At one time, it was thought that the basis was Zn-induced elevation of intestinal cell metallothionein, but high Zn intake can produce copper deficiency in metallothionein knockout mice [24].

In summary, under the conditions of this study, a moderately high dose of Zn as glycinate can raise plasma Zn without affecting an indicator of copper status. Under the current study conditions, Zn gluconate affected neither plasma Zn nor the indicator of copper status.

Conflict of Interest R. DiSilvestro is currently using zinc glycinate in products affiliated with his startup company, but that was not the case at the time the study was done (which was over 12 years ago).

References

1. DiSilvestro RA (2005) Handbook of minerals as nutritional supplements. CRC Press, Boca Raton
2. Prasad A (2013) Discovery of human zinc deficiency: its impact on human health and disease. Adv Nutr 4:176–190
3. Bagchi D, Vuchetich PJ, Stohs SJ et al (1998) Protective effects of zinc salts on TPA-induced hepatic and brain lipid peroxidation, glutathione depletion, DNA damage and peritoneal macrophage activation in mice. Gen Pharmacol 30:43–50
4. Hadwan M, Almashhedy L, Alsalman A (2012) Oral zinc supplementation restores high molecular weight seminal zinc binding protein to normal value in Iraqi infertile men. BMC Urol 12:32
5. Arnold L, Disilvestro R, Joseph E et al (2011) Zinc for attention-deficit/hyperactivity disorder: placebo-controlled double-blind pilot trial alone and combined with amphetamine. J Child Adolesc Psychopharmacol 21:1–19
6. Vishwanathan R, Chung M, Johnson E (2013) A systematic review on zinc for the prevention and treatment of age-related macular degeneration. Invest Ophthalmol Vis Sci 54:3985–3998
7. Samman S, Roberts D (1987) The effect of zinc supplements on plasma zinc and copper levels and the reported symptoms in healthy volunteers. Med J Aust 146:246–249
8. Wiernicka A, Jańczyk W, Socha P et al (2013) Gastrointestinal side effects in children with Wilson's disease treated with zinc sulphate. World J Gastroenterol 19:4356–4362
9. Rerksuppaphol S, Rerksuppaphol L (2013) A randomized controlled trial of chelated zinc for prevention of the common cold in Thai school children. Paediatr Int Child Health 33:145–150
10. Blostein-Fujii A, DiSilvestro RA, Malarkey W et al (1997) Short term zinc supplementation in type II diabetic women: effects on plasma 5′-nucleotidase activities, insulin-like growth factor levels and lipoprotein oxidation rates in vitro. Am J Clin Nutr 66:639–642
11. Gandia P, Bour D, Houin G et al (2007) A bioavailability study comparing two oral formulations containing zinc (Zn bisglycinate vs. Zn gluconate) after a single administration to twelve healthy female volunteers. Int J Vitam Nutr Res 77:243–248
12. DiSilvestro RA, Swan M (2008) Comparison of four commercially available zinc supplements for performance in a zinc tolerance test. FASEB J 22:693.3
13. Fischer P, Giroux A, L'Abbé M (1984) Effect of zinc supplementation on copper status in adult man. Am J Clin Nutr 40:743–746
14. Yadrick M, Kenney M, Winterfeldt E (1989) Iron, copper, and zinc status: response to supplementation with zinc or zinc and iron in adult females. Am J Clin Nutr 49:145–150
15. Abdallah S, Samman S (1993) The effect of increasing dietary zinc on the activity of superoxide dismutase and zinc concentration in erythrocytes of healthy female subjects. Eur J Clin Nutr 47:327–332
16. DiSilvestro RA, Marten JT (1990) Effects of inflammation and copper intake on rat liver and erythrocyte Cu-Zn superoxide dismutase activity levels. J Nutr 120:1223–1227
17. Donangelo C, Woodhouse L, King S, Viteri F, King J (2002) Supplemental zinc lowers measures of iron status in young women with low iron reserves. J Nutr 132:1860–1864
18. Shah U, Abu-Shaheen A, Al-Tannir M et al (2013) The efficacy of zinc supplementation in young children with acute lower respiratory infections: a randomized double-blind controlled trial. Clin Nutr 32:193–199
19. Black M, Medeiros D, Brunett E, Welke R (1988) Zinc supplements and serum lipids in young adult white males. Am J Clin Nutr 47:970–975
20. Tompkins T, Renard N, Kiuchi A (2007) Clinical evaluation of the bioavailability of zinc-enriched yeast and zinc gluconate in healthy volunteers. Biol Trace Elem Res 120:28–35
21. DiSilvestro RA, Joseph E, Raimo A, Kim YM (2012) Copper supplementation effects on blood copper enzymes and plasma cardiovascular health markers in middle aged people. Metabolism 61:1242–1246
22. Jones AA, DiSilvestro RA, Coleman M, Wagner TL (1997) Copper supplementation of adult men: effects on blood copper enzyme activities and indicators of cardiovascular disease risk. Metabolism 46:1380–1383
23. DiSilvestro RA, Marten JT, Skehan M (1992) Effects of copper supplementation on ceruloplasmin and copper-zinc superoxide dismutase activities in free-living rheumatoid arthritis patients. J Amer Coll Nutr 11:177–180
24. Reeves PG (1998) Copper metabolism in metallothionein-null mice fed a high-zinc diet. J Nutr Biochem 9:598–601