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ABSORPTION OF ZINC SULFATE, METHIONINE, AND POLYASCORBATE IN THE PRESENCE AND ABSENCE OF A PLANT-BASED RURAL MEXICAN DIET[1]

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ABSTRACT

Using the zinc tolerance test, we compared the absorption of zinc from zinc methionine with that from zinc sulfate or zinc polyascorbate. Nine adults received 25 mg of elemental zinc in the three different supplements, either alone in a water solution or added to a standard meal (SM). All subjects were also studied with water alone and the SM alone, as controls. The SM contained plant foods that are habitually consumed in rural Mexico. When the supplements were given with water the area under the curve (AUC) for the 4-h observation was: 262 ± 30 μ/dl for zinc methionine, 225 ± 9 μg/dL for zinc polyascorbate, 210 ± 33 μg/dL for zinc sulfate and 1 ± 6 μg/dL for water. Plasma zinc increased more after methionine than after zinc sulfate ($p<0.05$). The ingestion of SM alone produced a significant reduction in plasma zinc compared to fasting levels (AUC= -50 ± 9 μg/dL) and the simultaneous addition of any of the three supplements failed to prevent this decline. The presence of food probably affected the utilization of absorbed zinc by peripheral tissues so that plasma zinc response was not useful for measuring the effect of food on zinc absorption.

Key Words: Zinc, Mexico, Absorption, Supplements

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INTRODUCTION

Clinical manifestations of zinc deficiency have been reported in several regions of the world (1-3). Zinc deficiency has been associated mainly with the presence of inhibitors of zinc absorption in the diets habitually consumed by these populations. More recently, the existence of growth retardation in Mexican-American children living in the United States has been attributed to zinc deficiency (4, 5).

In Mexico, growth stunting is prevalent in children living in the less-developed rural areas (6). The habitual diets of these children are based on plant foods, high in dietary fiber and phytic acid which are known to be potential inhibitors of zinc absorption (7, 8). In a recent study we demonstrated that a typical rural Mexican diet significantly inhibits zinc bioavailability (9). Others have also demonstrated a negative effect of plant foods on the absorption and utilization of dietary zinc (10, 11). Because of the high prevalence of growth stunting and poor zinc bioavailability, we initiated an investigation of the potential benefit of zinc supplementation on the growth and health of rural Mexican children. In order to choose the form of zinc that would be the most effective supplement we compared, using the zinc tolerance test, the absorption of zinc from a zinc-methionine complex with absorption from conventional inorganic zinc sulfate and zinc polyascorbate. Absorption of zinc from these supplements was compared after their administration in water. In addition, because the rural children might sometimes consume the zinc supplement close to a meal, we also compared absorption from the supplements given with a meal consisting of plant foods habitually consumed in the rural areas of Mexico.

MATERIALS AND METHODS

Subjects

Nine apparently healthy adult volunteers were involved in the study. They were 6 women and 3 men ranging in age from 24 to 48 years (average 37.5). These subjects were selected from students and staff of the National Institute of Nutrition (INN) in Mexico City, where the study was conducted. All subjects signed consent forms after the nature, purposes and potential risks of the study were explained to them. The research protocol was approved by the Committee of Biomedical Research on Humans at INN.

Zinc Absorption Evaluations

Each subject was studied during eight different treatments as follows: a) 63 mg of zinc sulfate (25 mg of elemental zinc); b) 125 mg of zinc polyascorbate (25 mg of elemental zinc) (Ester-C Zinc, Intercal Corp., AZ); c) 125 mg of zinc methionine (25 mg of elemental zinc) (OptiZinc, InterHealth Corp., CA); d) 100 ml deionized water; e) a standard test meal plus 25 mg of zinc as zinc sulfate; f) the standard test meal plus 25 mg of zinc as zinc polyascorbate; g) the standard test meal plus 25 mg of zinc as zinc methionine; and h) the standard test meal alone. In all treatments that included a zinc supplement, the zinc dose was diluted with 100 ml deionized water. The standard test meal consisted of foods regularly included in a typical rural Mexican diet as previously reported (9, 12), and described in Table 1.

TABLE 1

Foods Included in the Standard Rural Mexican Meal

Food	Amount (g)
Corn "tortillas"	69.3
Black beans	33.8
Vegetable oil	4.5
Lettuce	18.0
Tomato	45.0
Onions	< 1.0
"Chile"	< 1.0
Orange	135.0
Corn beverage "atole"	
Corn flour	9.0
Sugar	8.0
Water	200.0

Serum zinc response curves after each treatment were used as the measure of relative zinc absorption. A 5 ml sample of venous blood was drawn early in the morning after an overnight fast. The subjects then consumed one of the treatments and blood samples were collected at 1-h intervals for 4-h (13). Participants were ambulatory but consumed only deionized water during the duration of the evaluation. The eight different treatments were administered to all subjects in a Latin square design, with one week between each test.

Blood was collected using zinc-free plastic syringes and stainless steel needles, and transferred to plastic tubes containing 0.05 ml of 20% potassium oxalate as an anticoagulant. After centrifugation, the plasma was frozen and stored until analyzed. Samples were thawed and diluted 1:5 (vol:vol) with deionized water. Plasma zinc determination was measured by atomic absorption spectrophotometry using zinc reference standards in 5% glycerol (vol/vol). Recovery of added zinc in seven tests was 98 ± 0.4% (mean ± SD).

Data Analysis

Changes in the mean plasma zinc concentration for a given time interval following each treatment were compared by Student's t-test (14). The area under the discontinuous curves that represent changes in plasma zinc level was determined by triangulation and compared among treatments by the least significant difference method (14).

RESULTS

Fasting plasma zinc

Average plasma zinc at baseline for each treatment ranged from 63 ± 4 to 81 ± 5 μg/dL (mean ± sem). When subjects remained in the fasted state for the 4-h observation, zinc levels were remarkably stable with a very low variability among subjects (Figure 1, lower curve).

Plasma zinc response to zinc supplements administered in water

There was a substantial elevation of plasma zinc above fasting levels during the 4-h following the ingestion of all three supplements when they were administered in water (Figure 1). The peak height above baseline at 2-h post ingestion was 90.0 μg/dL with zinc methionine, 76.4 μg/dL with zinc polyascorbate, 69.4 μg/dL with zinc sulfate and 0.4 μg/dL with water alone. The increase after all three supplements was significantly higher than for the control ($p<0.05$). The only other significant difference in peak height was for zinc methionine compared with zinc sulfate ($p<0.05$). The area under the curve (AUC) for 4-h after zinc methionine was 262 ± 30 μg/dL, for zinc polyascorbate 225 ± 19 μg/dL, for zinc sulfate 210 ± 33 μg/dL and for water, 1 ± 6 μg/dL. Only the zinc methionine area was significantly higher than that for zinc sulfate ($p <0.05$).

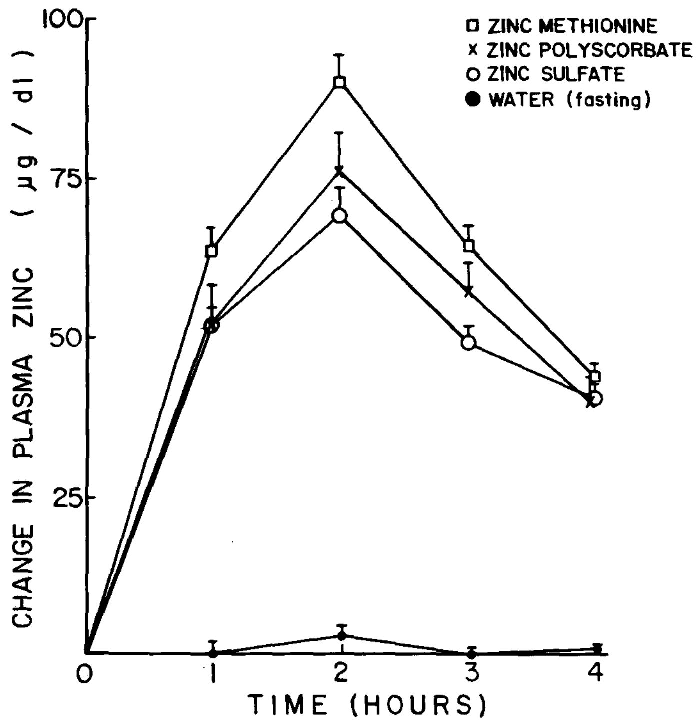

FIG.1. Change in plasma zinc concentration (mean ± sem) at 60 minute intervals following an oral dose of zinc supplements or water.

Effect of a rural Mexican meal on plasma zinc response

Figure 2 compares the change in plasma zinc concentration during fasting (lower curve of Fig. 1) with that after ingestion of the standard test meal. There was a significant decline in the level of zinc in plasma at 2, 3 and 4 hours postprandial ($p < 0.05$). The AUC for the fasting test was 1 ± 6 μg/dL and that for the test meal was -50 ± 9 μg/dL.

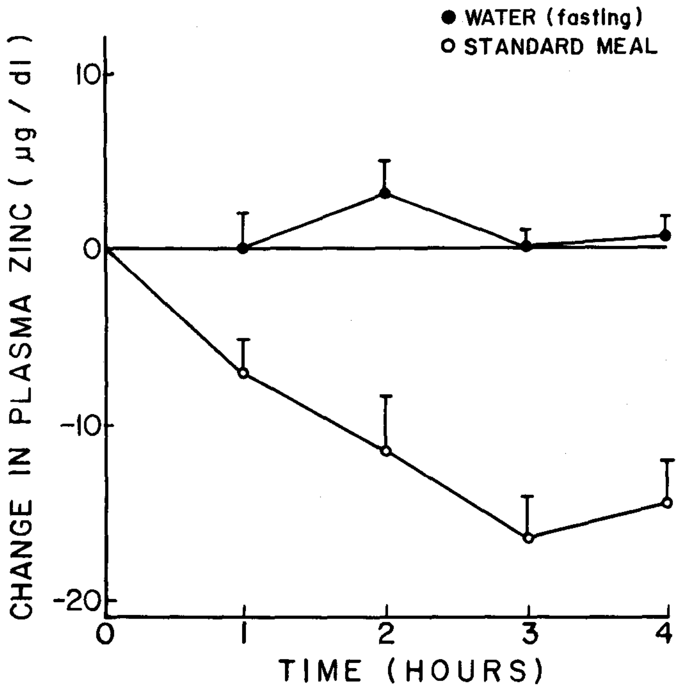

FIG.2. Change in plasma zinc concentration (mean ± sem) at 60 minute intervals following intake of the standard meals or water.

Plasma zinc response to zinc supplements given with the standard meal

The zinc tolerance curves in response to the ingestion of zinc supplements consumed with a rural Mexican meal are shown in Figure 3. When the supplements were administered with the standard meal there was a decline in plasma zinc during the 4-h observation. The AUC under the curve with each treatment was -19 ± 9 μg/dL for the standard meal (SM) + zinc sulfate, -40 ± 21 μg/dL for SM + zinc methionine, -43 ± 23 μg/dL for SM + zinc polyascorbate and -50 ± 9 μg/dL for the SM alone. There was no significant difference among treatments at any interval except that the slight increase in plasma zinc observed for SM + zinc sulfate at 1-h (2.1 ± 2.5 μg/dL) was significantly higher than for the SM alone (-4.7 ± 1.6 μg/dL) ($p<0.05$). The change in plasma zinc levels after any of the supplements was added to the standard meal was significantly ($p<0.001$) less than when the supplements were given in a water solution, at all intervals.

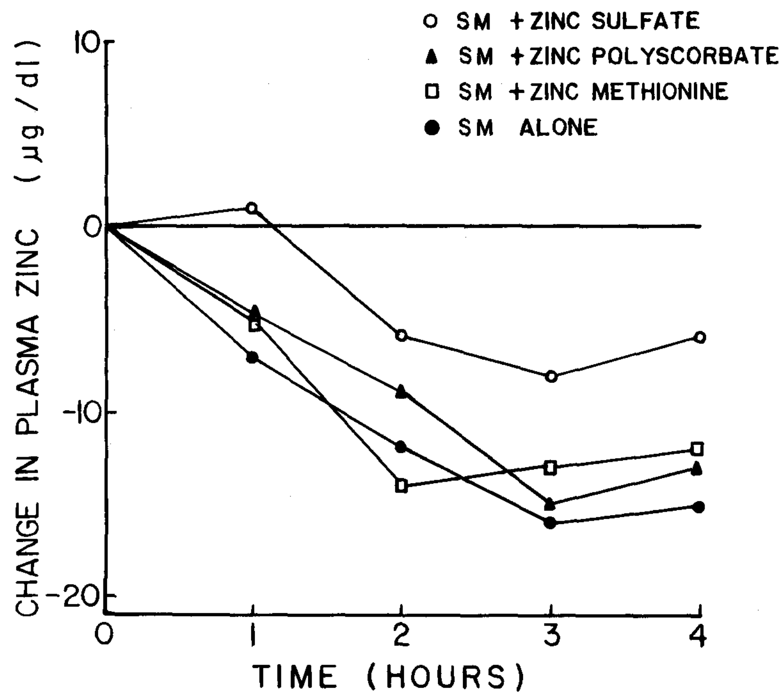

FIG.3. Change in plasma zinc concentration (mean ± sem) at 60 minute intervals following intake of the standard meal (SM) and the SM added with zinc supplements.

DISCUSSION

The overall purpose of this study was to evaluate whether zinc would be better absorbed from a zinc methionine complex or zinc polyascorbate than from zinc sulfate, especially when consumed in the presence of a typical rural Mexican meal in which the high phytate content is known to impair zinc absorption. The approach used was a comparison of plasma zinc response to ingestion of each form of supplement, in the presence and absence of a meal. In the absence of a meal there was a greater plasma AUC with zinc-methionine than with zinc sulfate, suggesting a better absorption of the former under those circumstances. Absorption from polyascorbate was intermediate and the AUC was not statistically different from that for zinc methionine or sulfate. We had expected that meal consumption would lower plasma zinc, but still anticipated that the level of decline might differ among the supplements. For example, Solomons et al. (13) evaluated zinc absorption by the change in plasma zinc response following a standard Guatemalan meal somewhat similar to the SM used in our study. As in our Mexican results, the Guatemalan diet decreased plasma zinc below fasting levels. However, the AUC during the 6-h of observation in the Guatemalan study was -18 ± 4 μg/dL compared to -50 μg/dL found in our study. Also, the addition of 25 mg of zinc as zinc sulfate to the Guatemalan diet obliterated the depression in plasma zinc concentration whereas in our study the same amount of zinc had no effect. The difference in composition of the diets may have been responsible for the difference in results between the two studies. The average fasting plasma zinc levels of the Mexican subjects in this study are similar to those of the urban Guatemalan subjects, but lower than those in other populations (15).

Several other investigators have used the oral zinc tolerance test to assess the effect of different foods and meals on zinc absorption (16-18), and have found that the response depends more on the composition of the food than on the zinc dose or diet (19,20). Often the results have been ascribed to the effect of differences in the diet on zinc absorption even though it is acknowledged that the method has its limitations (20,21). In our study, the ingestion of a standard meal alone produced a significant reduction in plasma zinc compared to fasting levels and the addition of zinc in three different forms did not prevent this decline. The explanation is that the decline in plasma zinc in the presence of a meal may be attributed, at least in part, to an effect of the meal on plasma zinc utilization or clearance. Valberg et al. (21) compared changes in plasma zinc concentration with estimates of zinc absorption obtained by administering radioactively labeled zinc, after giving zinc chloride alone or with a turkey meal. The AUC obtained from the plasma zinc response was 47 and 28 μmol/L (5-h test) for the zinc chloride alone and with the food respectively, suggesting that the turkey meal actually decreased zinc absorption. However, the absorption of labeled zinc, assessed by disappearance of label from feces and whole body counting, was the same whether the test dose was given with water or with turkey. They reached a similar conclusion concerning the limitations of the zinc tolerance test to measure zinc absorption in the presence of other foods.

Zinc status also influences the effects of a meal on the postprandial plasma zinc response. Hambidge et al. (22) showed that after human subjects were fed a zinc-restricted diet for 8 days their maximal post-prandial decline in plasma zinc after a low zinc breakfast was 19 μg/dL, significantly more than the 12 μg/dL drop that occurred in zinc-adequate subjects. This is further evidence that the post-prandial decline is affected more by post-absorptive events than by the efficiency of absorption per se.

Work in animal models suggests that the form of zinc supplement does affect how much zinc will be absorbed in the presence of foods that adversely affect absorption of the mineral. In 1963, Scott and Zeigler (23) postulated, based on observations in poultry, that a chelate of zinc with proteins or amino acids might enhance zinc absorption under some conditions. Later research demonstrated a strong potentiating effect of a zinc histidine complex on zinc uptake by the rat ileum (24). Recently, Wedekind et al. (25) compared the bioavailability of zinc methionine to that of zinc sulfate using three different diets fed to chicks: crystalline amino acids, soy isolate and a corn-soybean mixture. Bioavailability of the zinc from methionine relative to zinc from sulfate was 208, 177 and 117% for the corn-soybean meal, soy isolate and amino acid diets respectively. Based on these data we expected that more zinc would be absorbed from zinc methionine than from zinc sulfate in the presence of the rural meal, but no difference was detected using the zinc tolerance test.

A more reliable approach to comparing zinc absorption is to label the zinc in the supplements with a radioactive or stable isotope, and measure unabsorbed zinc in feces (26) or urinary excretion of the label (27). However, these approaches also have their limitations and more importantly are relatively expensive and impractical for studies on human subjects, especially in a developing country. This is unfortunate given the need to know which forms of supplemental zinc are most efficiently absorbed from specific local diets that contain constituents inhibitory to zinc absorption.

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