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ZINC METABOLISM OF YOUNG COLLEGE
WOMEN ON SELF-SELECTED DIETS

THESIS

Presented to the Graduate Council of the
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By

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INTRODUCTION

The function of zinc in the nutrition of man has been studied by numerous investigators in an attempt to determine the needs of the body for this element. To date no deficiency disease has been produced by a zinc-deficient diet and cured by the addition of this element to the diet. The only criteria thus far presented to establish its essentialness in the dietary of man are the facts that zinc is retained by the body and that its presence is necessary for normal functioning of other nutrients.

That animals must have zinc in their diet has been shown repeatedly. McHague (1926)¹ conducted one of the first studies with animals to determine whether or not growth would take place on a zinc-deficient diet. Though favorable effects on the time of survival due to the addition of twenty-five parts per million of zinc to a zinc-deficient diet were observed, this is not a conclusive study because of the lack of other essential dietary principles in the diet. With a diet adequate in all respects but zinc, Newell and McCollum (1933)² obtained results which

¹J. S. McHague, "Further Evidence that Small Quantities of Copper, Manganese and Zinc Are Factors in the Metabolism of Animals," American Journal of Physiology, Vol. 77 (1933), p. 245.

²John M. Newell and E. V. McCollum, "Studies on the Role of Zinc in Nutrition," Journal of Nutrition, Vol. 6 (1933), p. 289.

caused them to conclude that zinc is probably not an essential nutritional factor in the growth of the rat.

However, growth studies with rats conducted by Stirn et al (1935)³ showed the rate of growth to be markedly retarded on a synthetic diet low in zinc compared to control animals which received the same diet with added zinc. Interference with the development of a normal fur coat was observed on the zinc-low diet. Todd and co-workers (1934)⁴ found retarded growth rate of rats on a zinc low diet to be accelerated by the addition of zinc salts to the diet.

Young mice restricted to a diet deficient in zinc were shown by Day (1942)⁵ to be retarded in growth from the first week with over 50 per cent dying within three to six weeks.

Growth results obtained by Hubbell and Mendel (1927)⁶ caused them to question whether the value of zinc lies in the metal itself or in its association with other metals which are present in small amounts.

³F. E. Stirn, C. A. Elvehjem, and E. B. Hart, "The Indispensability of Zinc in the Nutrition of the Rat," Journal of Biological Chemistry, Vol. 109 (1935), p. 347.

⁴W. R. Todd, C. A. Elvehjem, and E. B. Hart, "Zinc in the Nutrition of the Rat," American Journal of Physiology, Vol. 107 (1934), p. 146.

⁵Harry G. Day, "The Effects of Zinc Deficiency in the Mouse," Federation of American Societies for Experimental Biology Proceedings, Vol. 1 (1942), p. 188.

⁶R. B. Hubbell and L. M. Mendel, "Zinc and Normal Nutrition," Journal of Biological Chemistry, Vol. 75 (1927), p. 567.

Evolving from data showing the essentialness of zinc in animal nutrition were studies to determine how the metal functions. It was noted by Keilin and Mann (1939)⁷ that the red blood cell enzyme, carbonic anhydrase, contains 0.31 to 0.34 per cent of zinc and that all of the zinc within the red blood cell is found in this enzyme. Hove et al (1940)⁸ confirmed this report and also observed decreases in pancreatic trypsin and amylase (1938)⁹ and in intestinal phosphatases (1940).¹⁰ Whereas these studies pointed to a disruption in digestive and absorptive processes in the gastrointestinal tract of zinc-deficient rats, they likewise showed abnormal metabolism of nitrogen. Delayed absorption appears to be greater for nitrogenous products than for carbohydrates (Hove et al, 1937).¹¹

⁷D. Keilin and T. Mann, "Carbonic Anhydrase," Nature, Vol. 144 (1939), p. 442.

⁸E. Hove, C. A. Elvehjem, and E. B. Hart, "The Relation of Zinc to Carbonic Anhydrase," Journal of Biological Chemistry, Vol. 136 (1940), p. 425.

⁹E. Hove, C. A. Elvehjem, and E. B. Hart, "Further Study on Zinc Deficiency in Rats," American Journal of Physiology, Vol. 124 (1938), p. 750.

¹⁰E. Hove, C. A. Elvehjem, and E. B. Hart, "The Effect of Zinc on Alkaline Phosphatases," Journal of Biological Chemistry, Vol. 134 (1940), p. 134.

¹¹E. Hove, C. A. Elvehjem, and E. B. Hart, "Physiology of Zinc in Nutrition," American Journal of Physiology, Vol. 119 (1937), p. 768.

A human study conducted by Scott and Fisher (1938)¹² suggested the possibility that part of the zinc in the pancreas is concerned with the storage of insulin. The amount of zinc contained in the pancreas of diabetics was only one-half that found in the pancreas of normal individuals. Persons suffering from beri-beri were found to have a 50 per cent decrease in the zinc content of skin, hair, and nails by Eggleton (1939),¹³ thus suggesting a relationship between zinc and thiamine metabolism. In this study he likewise noted a correlation between the zinc and vitamin B₁ content of foods.

No requirements for zinc intake in humans have been established. Since the element is almost universally present in animal and plant substance, it would be of interest to know whether or not zinc deficiency could occur in humans. The pattern of zinc metabolism has been studied by only a few workers, producing some interesting findings.

Drinker and co-workers (1927)¹⁴ were among the early observers of zinc metabolism who used human subjects. They

¹²D. A. Scott and A. M. Fisher, "The Insulin and the Zinc Content of Normal and Diabetic Pancreas," Journal of Clinical Investigation, Vol. 17 (1938), p. 725.

¹³W. G. Eggleton, "The Zinc Content of Epidermal Structures in Beri-beri," Biochemical Journal, Vol. 33 (1939), p. 403.

¹⁴K. R. Drinker, J. E. Fehnel, and M. March, "Normal Excretion of Zinc in the Urine and Feces of Man," Journal of Biological Chemistry, Vol. 72 (1927), p. 375.

found urinary excretions of zinc not to increase appreciably with high zinc intake, whereas the fecal elimination more nearly corresponded with intake. Two males and one female subject observed by McCance and Widdowson (1942)¹⁵ excreted an average of 0.3 milligrams of zinc per day in urine. This amount did not vary with intake by mouth or with intravenous injections, thus suggesting that, in normal life, the kidney can take little or no part in regulating the amount of zinc in the body. Within the limits of experimental error these subjects excreted almost the same amount of zinc in their stools as they ingested in their food. This is the only study noted where there was not a definite zinc retention. Three boys between the ages of three and six years studied by Scoular (1939)¹⁶ retained between 2 and 3 milligrams daily on a daily intake of from 4 to 6 milligrams, the highest ingestion being only 41 per cent higher than the lowest in the study. Urinary zinc was from 0.04 to 6 per cent of the intake, this amount not being related to the intake nor constant for a given individual. Forty-two to 163 per cent of the ingested zinc was excreted in the feces. Of thirty-five balances determined, ten were negative.

¹⁵R. A. McCance and E. M. Widdowson, "The Absorption and Excretion of Zinc," Biochemical Journal, Vol. 36 (1942), p. 692.

¹⁶F. I. Scoular, "A Quantitative Study by Means of Spectrographic Analysis of Zinc in Nutrition," Journal of Nutrition, Vol. 17 (1939), p. 103.

When the average zinc retention for eight children, age eight to twelve years, was studied by Stern et al (1941),¹⁷ the amount ranged from 17 to 52.3 per cent of the 15.6 milligrams of zinc ingested. From 2.6 to 4.2 per cent of the zinc intake was excreted in the urine, with 44.6 to 78.8 per cent being excreted in the feces. This gave an average retention of 4.8 milligrams daily for the children.

From these few human zinc metabolism studies it appears that the gastrointestinal tract is the mode of zinc elimination from the body, with the kidney playing a minor, if any, role in the retention of the element. No definite pattern of retention has been shown to be established.

No studies have been noted that show the intake, retention, and pattern of excretion of zinc for college-age women. Therefore, the present study was made to observe zinc metabolism of young college women on self-selected diets.

¹⁷A. Stern, Margaret Nalder, and Icie G. Macy, "Zinc Metabolism in Childhood," Journal of Nutrition, Vol. 21 (1941), Supplement, p. 8.

PROCEDURE

Subjects for this study were thirteen young college women between the ages of seventeen and twenty-seven who were residents of the Home Management House, North Texas State College, during the time of their participation in the study. All of the subjects were Home Economics majors and participated voluntarily.

The Home Management House is a duplex which was occupied by seven girls on one side and by six on the other. All of the girls residing on one side of the house had a common food supply, thereby the analysis of one day's food supply provided the intake for all the girls on one side. The only exceptions were Co and Ho, during Period I, who had one dinner away from the house. A duplicate of this meal was obtained for analysis and used in determining their zinc intake for that day. This made it necessary to make three zinc determinations in food for that day: one of the breakfast and lunch which were consumed by the entire group and one on each of the dinners.

The study was divided into two five-day metabolism periods. Period I was from October 27 through October 31, with Period II beginning on November 3 and continuing through November 7. Seven girls served as subjects for Period I, with Period II having six subjects. Previous investigations

reported from this laboratory have shown that the college girls ingest relatively small amounts of food. Because of its concentration, Multi-Purpose Food, Formula B, was used as a supplement in the diet during Period II as an economical source of dietary constituents, particularly of protein which has repeatedly been shown to be low. Prior to the balance study, the subjects planned their menus jointly to enable the food consumed to be the same kind and amount during both periods. Therefore the only difference in the food intake for the two periods was the addition of the Multi-Purpose Food to each of the five daily foods during Period II. All collections of food, urine, and feces were made in well-aged bottles to prevent contamination from the glassware.

A duplicate serving of each food was collected in weighed, eclipse, wide-mouth, quart-size fruit jars at the same time the girls were served at the table. Milk was the only variable in the daily food intake of the girls during a given period. A record was kept of the number of cups of milk consumed by each girl each day to make allowance for this variation. Milk was collected in a jar similar to that of the food, with one-fourth cup being added each day to give one homogeneous sample for analysis. After each jar of food had been weighed, the contents were macerated in a Waring Blendor; then the three meals for the day were combined and again macerated. These daily food composites were stored in the refrigerator until they were analyzed.

Carmine was administered according to the method of Scoular and Holt (1948).¹⁸ Fecal collections were made directly into weighed, eclipse, pint-size, wide-mouth fruit jars. After the weight of each girl's wet feces was obtained, the total was macerated in a Waring Blendor and stored in the refrigerator until zinc determinations were made.

Twenty-four hour urine collections were made directly into amber gallon bottles. Aliquots were taken from each day's collection and stored in quart amber bottles in the refrigerator, using toluene as a preservative.

One liter of tap water was evaporated to 50 milliliters, and duplicate tests were run to determine the zinc content of the water supply at the girls' residence. This was done in order to determine how much zinc was being ingested other than from the food source.

Food, water, milk, urine, and fecal zinc determinations were made according to the dithizone method of Hibbard (1937).¹⁹ Three food samples weighing ten grams each were first used. However, one-gram samples proved sufficient and were used for the remaining determinations. After the food was ashed, the extract was taken up in 100 milliliters

¹⁸F. I. Scoular and Flowayne Holt, "Iron and Copper Metabolism of Young College Women on Self-Selected Diets," Journal of Nutrition, Vol. 35 (1949), p. 717.

¹⁹See Appendix for complete chemical procedure.

of 10 per cent hydrochloric acid. In analyzing ten-gram samples, 1, 5, or 10 milliliters of the extraction were used for color development, this depending upon the amount of zinc in the sample. Ten milliliters were used with the one-gram samples.

Ten grams of milk were likewise ashed, diluted to 100 milliliters with 10 per cent hydrochloric acid, and 10 milliliters were taken for color development with dithizone.

Two-gram fecal samples were found to contain enough zinc to be read on the standard curve. After ashing, the sample was made up to 100 milliliters with 10 per cent hydrochloric acid, and 10 milliliters were then taken for dithizone treatment. Ten milliliters of urine were ashed, extracted in 100 milliliters of 10 per cent hydrochloric acid and 10 milliliters taken for some of the color development treatments. When the urine samples produced a more intense color, 5 milliliters were used for the dithizone treatment.

After the color was developed in the food, milk, urine, and fecal samples, readings were made in a Cenco-Sheard-Sanford Photelometer with a blue filter number 435.

For the reference curve used in determining the zinc content of the unknown samples, a standard solution of zinc was made by dissolving 0.2198 milligrams of $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ in 500 milliliters of zinc-free water. Portions of this solution were diluted to contain 0.5, 1, 2, 4, 6, 8, and 10 micrograms of zinc per milliliter. The solutions were treated

with dithizone, and the results were plotted on semi-logarithmic paper (Figure 1).

Following Hibbard's procedure (1937),²⁰ every precaution was taken to prevent contamination of the samples prior to and during the analyses. All reagent bottles and glassware were aged because of previous use. These bottles were rinsed with zinc-free water, and zinc-free water was used in the preparation of the solutions. Furthermore, a reagent blank was read, and this value was subtracted from the readings of the unknown before the zinc value was determined from the reference curve.

The zinc determinations were run in triplicate. Two micrograms of zinc were added to one of the samples to obtain the recoveries shown in Table 1.

²⁰P. L. Hibbard, "A Dithizone Method for Measurement of Small Amounts of Zinc," Industrial and Engineering Chemistry, Analytical Edition, Vol. 9 (1937), p. 127.

Mg/L

CONCENTRATION

12

0.5 1 2 4 6 8 10

10

20

30

40

50

60

70

80

90

100

TRANSMITTANCY

ANALYTICAL CURVE FOR

Zinc

PREPARED BY Tribble

DATE November, 1952

CELL THICKNESS

TYPE INSTRUMENT Photometer

Cenco-Sheard-Sanford

SERIAL NO.

TABLE 1
RECOVERY* OF ZINC IN FOOD, FECES,
AND URINE ALIQUOTS

Sample		Zinc Added	Per Cent	
Kind	Number		Range of Recovery	Average Recovery
		mcg		
Food	12	2	95-109	100.6
Milk	2	2	100-105	102.5
Urine	64	2	88-110	95.5
Feces	13	2	90-110	102.0

*Full data are listed in the Appendix.

DISCUSSION

The daily zinc intake and excretion of thirteen young college women were determined, by chemical analysis, during the fall of 1952. Two five-day periods were used in making this metabolism study, with seven subjects participating during Period I and six during Period II. The subjects were residents of the Home Management House, North Texas State College, where they planned, prepared, and served their menus.

The seven subjects from Period I met with the six subjects from Period II and planned their meals cooperatively so that the menus²¹ would be identical for both periods. Because previous studies from this laboratory²² have shown the residents of the Home Management House to ingest relatively small amounts of food, with resulting low nutrient intake, Multi-Purpose Food was used in the preparation of five of the daily foods during Period II. Because of its concentration, it was hoped that the Multi-Purpose Food would increase the nutritive value of the food intake without a change in the total daily volume. During Period I

²¹The menus for all periods are listed in detail in the Appendix.

²²North Texas State College, School of Home Economics Research, Bulletin No. 228, 1951, p. 33.

Subjects Co and Ho ate the evening meal of Day 5 away from the house. Consequently, a separate analysis of this meal was made in determining the zinc intake of those two subjects for that day. The only variable in the daily food intake, common to all subjects during the two periods, was the amount of milk consumed by each subject; this allowed for individual needs and preferences. Separate analyses of composite food and of milk were made so that this variable could be used in obtaining the total zinc intake of each subject. The zinc content of the daily food composites and of the food composites and milk for each subject are given in Table 2.

The tap water supply at the residence was analyzed and found to contain 0.0072 milligrams of zinc per liter. This amount of zinc from drinking water and beverages was considered negligible, since the tap water used in other cooking processes was analyzed with the food sample.

The zinc values of the food composites showed little variation during the ten days of this study. The smallest value, 9.8 milligrams, was obtained from the food composite for the second day of Period I, whereas the highest value, 14.4 milligrams, was obtained from the food composite for the first day of Period II. These values gave a difference of 4.6 milligrams between the lowest and the highest zinc intake for the two five-day periods. The use of Multi-Purpose Food in the diet in Period II contributed to this difference in zinc intake. Multi-Purpose Food, because of its concentration,

TABLE 2
DAILY ZINC INTAKE OF COLLEGE WOMEN
ON SELF-SELECTED DIETS

Subject	Milligrams of Zinc										Average Daily Intake
	Composite Food							Milk			
	Day 1	Day 2	Day 3	Day 4	Day 5	Total	Av./Day	Total	Av./Day		
Period I											
Wi	12.6	9.8	11.3	12.5	11.3	57.5	11.5	3.0	0.6	12.1	
Co	12.6	9.8	11.3	12.5	13.0*	59.2	11.8	2.3	0.5	12.3	
Yo	12.6	9.8	11.3	12.5	11.3	57.5	11.5	4.5	0.9	12.4	
Ho	12.6	9.8	11.3	12.5	13.0*	59.2	11.8	4.5	0.9	12.7	
St	12.6	9.8	11.3	12.5	11.3	57.5	11.5	1.5	0.3	11.8	
Le	12.6	9.8	11.3	12.5	11.3	57.5	11.5	3.0	0.6	12.1	
Go	12.6	9.8	11.3	12.5	11.3	57.5	11.5	3.8	0.8	12.3	
Average	12.6	9.8	11.3	12.5	11.8	58.0	11.6	3.2	0.7	12.2	
Period II											
Mc	14.4	13.2	12.1	13.7	10.2	63.6	12.7	4.2	0.8	13.6	
Fr	14.4	13.2	12.1	13.7	10.2	63.6	12.7	7.2	1.4	14.2	
Ko	14.4	13.2	12.1	13.7	10.2	63.6	12.7	7.2	1.4	14.2	
Bi	14.4	13.2	12.1	13.7	10.2	63.6	12.7	7.2	1.4	14.2	
As	14.4	13.2	12.1	13.7	10.2	63.6	12.7	6.3	1.3	14.0	
Br	14.4	13.2	12.1	13.7	10.2	63.6	12.7	5.9	1.2	13.9	
Average	14.4	13.2	12.1	13.7	10.2	63.6	12.7	6.5	1.3	14.0	
Mean											
Average	13.5	11.5	11.7	13.1	11.0	60.8	12.2	4.9	1.0	13.0	

*Night meal eaten outside the house and analyzed separately.

gave a larger weight, without a change in volume. This difference in weight, as it affected zinc intake, is evidenced by the values for Day 1. During Period I the composite weighed 968.3 grams and provided 12.6 milligrams, whereas in Period II the same menu and volume weighed 1316.8 grams and provided 14.4 milligrams of zinc. Furthermore, regardless of the use of Multi-Purpose Food, the type of menu served also made a difference in the daily zinc intake. Beef, eggs, yeast bread, and potatoes, which are all good sources of zinc, were served on the days of the highest zinc values.

Multi-Purpose Food, which was used as a substitute for some of the ingredients or as a supplement in each of five foods in the daily menus during Period II, made a decided difference in the zinc intake for this period. Although the size of the servings was the same for corresponding menus for both periods, the total weight of the daily food composites was higher, due to the Multi-Purpose Food, for each day of Period II than for Period I except for Day 5. The lower weight for the fifth day can be accounted for by the manner in which the Multi-Purpose Food was used. As was true for each day, five dishes containing Multi-Purpose Food were served on Day 5, but as a substitute, not as a supplement, for part of the flour and part of the beans. Substitution instead of supplementation of the food consistently gave lower weight and lower zinc values. This is particularly noticeable on Day 2 of both periods when Multi-Purpose Food

was used as a supplement in two dishes of Period II. While the lowest zinc value (9.8 milligrams) was obtained for this day in Period I, the third highest value (13.2 milligrams) was obtained for this day in Period II. The weight of the composite increased by 329.4 grams, whereas the zinc content increased 3.4 milligrams with the supplementation of salmon loaf and macaroni and cheese with Multi-Purpose Food. In general, higher zinc values of food accompanied higher weights of food intake, due to Multi-Purpose Food, although the volume of food consumed was the same. The composite food weights for all periods are given in detail in Table 5 of the Appendix.

During Period II the subjects commented on feeling uncomfortably full after meals and, on Day 2, had to eat part of their breakfast in the middle of the morning in order to consume the same volume of food as the subjects in Period I. Apparently this was associated with the concentration of the food consumed, as the weight for Day 2 of Period II increased 329.4 grams over the same volume of food consumed on Day 2 of Period I. On Day 3 of Period II, when the food composite weight increased only 35.3 grams over Day 3 of Period I, the girls did not have the uncomfortably full feeling experienced on the other days of the second period.

Daily zinc content of the foods ranged from 9.8 to 12.5 and 10.2 to 14.4 milligrams for the two periods, respectively. Day 5 was the only day during Period II when the zinc value

was not higher than it was for the corresponding day and menu of Period I. This can be accredited to a lower weight of food consumption and to the small amount of Multi-Purpose Food substituted in the five daily foods in which it was used. On this day in Period I 1153.5 grams of food were consumed by five subjects, and 1255.4 grams were consumed by two subjects eating one meal out. During Period II the six subjects consumed only 973.7 grams of food for the same day. With this one exception, zinc values were higher during Period II than during Period I, with the daily increase ranging from 0.8 to 3.4 milligrams for Days 3 and 2, respectively.

The amount of milk consumed²³ made a noticeable difference in the daily zinc intake of the subjects. The cups of milk consumed daily by the subjects ranged from 0.4 to 1.2 for Period I and from 1 to 1.7 for Period II. The zinc furnished by these amounts of milk was 0.3, 0.9, 0.8, and 1.4 milligrams, respectively. An average of 1.54 cups of milk was consumed daily by the thirteen subjects, with an average of 0.79 milligrams of zinc being furnished per cup. Stern et al (1941),²⁴ in analyzing whole cow's milk, found one liter of milk to contain 2.89 milligrams of zinc. Assuming one liter to equal four cups of milk, the zinc value per cup would be 0.72 milligrams, a value almost equal to that of the present study.

²³Milk consumption by each subject is given in Table 8 of the Appendix.

²⁴Stern et al, op. cit.

The lowest average daily intake of zinc for Period I was obtained for the subject who consumed the smallest amount of milk, 0.4 cups daily. The highest zinc values were obtained for subjects Yo and Ho, who averaged the largest volume of milk daily, namely, 0.9 cups. As shown in Table 2 only one of these subjects had a different night meal which was higher in zinc than the night meal eaten at the house. This parallelling of zinc intake to milk consumption also held for Period II, with Subject Mc consuming the smallest amount of milk, 1 cup, and three subjects the largest amount, or 1.7 cups daily.

Food and milk composites for the subjects gave an average daily intake of zinc that ranged from 11.8 to 12.7 and 13.6 to 14.2 milligrams for the two respective periods. The average daily zinc intake for Period I was 12.2 milligrams and for Period II, 14.0 milligrams. Thus the difference in the averages for the two periods was only 1.8 milligrams. The zinc intake for the present study is somewhat lower than the 15.6 milligrams consumed by the eight children, aged eight to twelve years, studied by Stern et al (1941),²⁵ but higher than the 4 to 6 milligrams consumed by three boys ranging in age from three to six studied by Scoular (1939).²⁶ However, for college women, the subjects consumed relatively

²⁵Ibid.

²⁶Scoular, op. cit.

small amounts of food, as has previously been observed in relation to nitrogen studies.

Fecal and urinary excretions of zinc by the subjects are given in Table 3. The average daily fecal excretion for Period I ranged from 3.2 to 5.2 milligrams, with an average of 4.5 milligrams for the seven subjects studied during this period. This value was higher during Period II. The average for individual subjects ranged from 4.3 to 7.9 milligrams, with an average of 6.5 milligrams for the six subjects. The difference between fecal excretion for the two periods was 2.0 milligrams. As was noted with food composites, there was a definite tendency toward higher fecal excretions with higher weights of feces. Total fecal weights for the subjects are given in Table 7 of the Appendix. The mean average of 5.5 milligrams of fecal zinc for these thirteen subjects was lower than Stern et al's (1941)²⁷ excretion of 7 to 12 milligrams, average 10.4 milligrams, for children ingesting 15.6 milligrams. Furthermore, children studied by Scoular (1939)²⁸ excreted from 2.47 to 6.40 milligrams of zinc, with an average of 4.1 milligrams, in their feces on a lower level of intake than ingested by the college women of the present study. The only adult study was reported by McCance and Widdowson (1942),²⁹ who found fecal excretion to be almost the same as

²⁷Stern et al, op. cit. ²⁸Scoular, op. cit.

²⁹McCance and Widdowson, op. cit.

TABLE 3
TOTAL EXCRETION OF ZINC

Subject	Milligrams of Zinc									
	Urine					Feces		Excretion		
	Day 1	Day 2	Day 3	Day 4	Day 5	Total	Av./Day	Total	Av./Day	
Period I										
Wi	1.9	2.6	0.6	0.8	0.7	6.5	1.3	16.0	3.2	
Co	0.3	0.4	0.9	1.1	0.5	3.1	0.6	20.7	4.1	
Yo	0.6	0.4	0.6	1.3	0.6	3.6	0.7	29.4	5.9	
Ho	1.0	1.0	1.3	0.6	1.4	5.4	1.1	20.7	4.1	
St	1.8	1.7	3.0	0.8	1.8*	9.1	1.9	22.5	4.5	
Le	1.6	0.4	1.5	1.3	2.2	7.0	1.6	26.0	5.2	
Go	0.4	0.2	1.2	0.6	0.5	2.9	0.6	23.3	4.7	
Average	1.1	1.0	1.3	0.9	1.1	5.4	1.1	22.7	4.5	
Period II										
Mc	0.8	0.7	0.7	0.7	1.8	4.2	0.8	38.1	7.6	
Fr	0.6	1.0	0.6	0.9	1.1	4.1	0.8	31.5	6.3	
Ko	0.7	0.8	1.8	0.7	1.0	5.0	1.0	33.4	6.7	
Bi	0.7	0.6	0.8	1.1	0.8	3.9	0.8	39.3	7.9	
As	0.4	0.7	1.1	0.6	1.9	4.7	1.0	32.2	6.4	
Br	0.5	0.3	1.1	0.7	1.2	3.8	0.8	21.7	4.3	
Average	0.6	0.7	1.0	0.8	1.2	4.3	0.9	32.7	6.5	
Mean										
Average	0.9	0.9	1.2	0.9	1.2	4.9	1.0	27.7	5.5	
								34.4	6.5	

*Average for the first four days used, as urine for this day was unobtainable.

the amount ingested. This is not true of the adults of the present study.

Fecal excretion of zinc was not related to the zinc ingested, although there was a higher value of excretion during Period II with the higher zinc intake. These seemed, however, to be somewhat more related to the total weight of the feces than to the total zinc ingestion.

Individual daily urinary zinc excretion ranged from 0.2 to 3.0 and from 0.3 to 1.9 milligrams for Period I and Period II, respectively. These values gave an average daily urinary excretion of 1.1 milligrams for Period I and 0.9 milligrams for Period II. Investigators making children's studies did not find urinary excretion values to reach as high a level as those of the present study. Consuming 4 to 6 milligrams of zinc, the children observed by Scoular (1939)³⁰ excreted an average of 0.7 milligrams of zinc in their urine, whereas Stern et al (1941)³¹ obtained an average urinary excretion value of 0.5 milligrams of zinc with older children on a daily ingestion level of 15.6 milligrams. The fact that the higher ingestion level of the present study did not increase the urinary zinc excretion is in accord with results reported by McCance and Widdowson (1942),³² who observed

³⁰Scoular, op. cit.

³¹Stern et al, op. cit.

³²McCance and Widdowson, op. cit.

urinary excretion not to vary with increased zinc intake by mouth or by intravenous injections.

Five to 15.5 per cent of the zinc ingested by the subjects during Period I was excreted through the urine. This gives an average of 9 per cent for this period. The per cent of urinary excretion for Period II ranged from 5.4 to 7, with an average of 6.7. Whereas the fecal excretions for Period I ranged from 26.4 to 47.3 per cent, with an average of 38 per cent, of the total zinc ingested and from 31 to 56 per cent, with an average of 46.6 per cent, during Period II, these percentage values are low compared with those reported on children of 0.04 to 6.0 per cent urinary excretion and 42 to 163 per cent fecal excretion by Scoular (1939)³³ and the 2.6 to 4.2 and 44.6 to 78.8 per cent urinary and fecal excretions, respectively, observed by Stern et al (1941)³⁴ for older children.

Individual total retentions of zinc for Period I ranged from 5.3 to 7.6, with an average of 6.6 milligrams. For Period II, retentions ranged from 5.1 to 8.8, with an average of 6.6 milligrams for the five days. A mean average for retention of 6.6 milligrams of zinc was obtained for the thirteen subjects. This retention is higher than the average retention of 1.2 milligrams for boys three to six years of age

³³Scoular, op. cit.

³⁴Stern et al, op. cit.

TABLE 4
ZINC METABOLISM OF COLLEGE WOMEN
ON SELF-SELECTED DIETS

Subject	Average Per Day							Per Cent Excreted through Urine	Per Cent Excreted through Feces
	Milligrams of Zinc				Retained in Body				
	Food Intake	Excretion		Total					
		Urine	Feces						
Period I									
W1	12.1	1.3	3.2	4.5	7.6	10.7	26.4		
Co	12.3	0.6	4.1	4.8	7.5	5.0	33.7		
Yo	12.4	0.7	5.9	6.6	5.8	5.7	47.3		
Ho	12.7	1.1	4.1	5.2	7.5	8.4	32.4		
St	11.8	1.8	4.5	6.3	6.5	15.5	38.0		
Le	12.1	1.6	5.2	6.8	5.3	12.9	43.0		
Go	12.3	0.6	5.5	6.1	6.1	4.7	45.0		
Average	12.2	1.1	4.6	5.8	6.6	9.0	38.0		
Period II									
Mc	13.6	0.8	7.6	8.4	5.1	9.8	56.0		
Fr	14.2	0.8	6.3	7.1	7.0	5.8	44.5		
Ko	14.2	1.0	6.7	7.7	6.5	7.0	47.0		
Bi	14.2	0.8	7.9	8.6	5.5	5.4	55.0		
As	14.0	0.9	6.4	7.4	6.6	6.6	46.0		
Br	14.0	0.8	4.3	5.1	8.8	5.4	31.0		
Average	14.0	0.9	6.5	7.4	6.6	6.7	46.6		
Mean									
Average	12.8	1.0	5.6	6.6	6.6	7.9	42.3		

on an ingestion level of 4 to 6 milligrams reported by Scoular (1939)³⁵ and of the 4.8 milligrams retention of children eight to twelve years of age ingesting 15.6 milligrams of zinc as reported by Stern et al (1941).³⁶ Whereas ten of the balances observed by Scoular (1939)³⁷ were negative, none of the subjects in the present study excreted more zinc than was ingested.

Apparently adults excrete a larger per cent of ingested zinc in the urine and a smaller per cent in the feces, with retentions higher in proportion to the level of zinc ingested, than do younger subjects.

³⁵Scoular, op. cit.

³⁶Stern et al, op. cit.

³⁷Scoular, op. cit.

SUMMARY

Zinc intake and excretion of thirteen young college women were studied during two five-day Monday-through-Friday periods. Seven subjects participated during Period I, with six participating during Period II. Subjects planned, prepared, and served their menus at the Home Management House where they resided during the study.

Menus were planned by the subjects to enable each person to ingest the same kind and amount of food for the two periods. Therefore, the only variables in the food intake were the substitution or supplementation of Multi-Purpose Food to each of five daily foods during Period II and the amount of milk consumed by each subject during both periods.

An average of 1.54 cups of milk was consumed daily by the thirteen subjects, with an average of 0.79 milligrams of zinc being furnished per cup. Zinc values for food composites ranged from 9.8 to 12.6 milligrams for Period I and from 10.2 to 14.4 milligrams for Period II. The average zinc intake per day was 11.6 and 12.7 milligrams for Periods I and II, respectively. Milk increased the average daily intake for Period I to 12.2 milligrams and to 13.9 milligrams for Period II. Multi-Purpose Food in the diet gave larger weights and higher zinc values for Period II with one exception, this being on a

day when it was substituted rather than supplemented in the preparation of the foods.

The average fecal excretion per day was from 3.2 to 7.9 milligrams for the thirteen subjects. For Period I the daily average was 4.5 milligrams and for Period II, 5.5 milligrams. This represented a fecal excretion of 26.4 to 56 per cent of ingested zinc. Where general intake was greater, as in Period II, the fecal zinc was greater.

The average urinary excretion of zinc for Period I was 1.1 and 0.9 milligrams for Period II. From 4.7 to 15.5 per cent of the ingested zinc was excreted through the urine. The urinary zinc showed no relation to the total zinc intake.

From 5.1 to 8.8 milligrams of zinc were retained by the subjects, with an average of 6.6 milligrams for both five-day periods. No subject was in negative balance during either of the metabolism periods.

By using Multi-Purpose Food, the average zinc intake increased by 1.2 milligrams and the average excretion by 2.3 milligrams, with the retentions remaining the same. On the basis of this limited study, it appears that the zinc intake of college women on self-selected diets is sufficient without supplementation, since retentions were the same for both periods.

APPENDIX

DIRECTIONS FOR PREPARING THE SOLUTIONS

Ten Per Cent Hydrochloric Acid - Ten milliliters of concentrated hydrochloric acid were diluted to one hundred with zinc-free distilled water.

Dithizone (Diphenylthiocarbazone) - Fifteen milligrams were dissolved in 100 milliliters of carbon tetrachloride.

Carbon Tetrachloride - Ordinary U. S. P. grade.

Concentrated Ammonium Hydroxide - Specific gravity 0.9 was diluted 1 to 4 with zinc-free distilled water.

Water, Free of Zinc - Distilled water was re-distilled from a pyrex apparatus.

Ammonium Citrate - Ten grams of citric acid were dissolved in 90 milliliters of zinc-free distilled water and made alkaline with ammonia. This was repeatedly shaken out with dithizone and carbon tetrachloride until no zinc was extracted.

Working Solution of Zinc - 0.2198 milligrams of $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ were dissolved in 500 milliliters of zinc-free distilled water. Two milliliters of this solution were diluted to 100 milliliters to make a solution of 2 micrograms of zinc per milliliter.

0.3 Normal Ammonium Hydroxide - Twenty milliliters of

ammonia were thoroughly mixed with 1 liter of distilled zinc-free water.

CHEMICAL METHOD FOR DETERMINATION OF ZINC

Organic substances were burned in crucibles and the ash extracted with 10 per cent hydrochloric acid. The solution was placed in a conical separatory funnel large enough to hold as much more as the volume of the solution, and ammonium hydroxide was added until the solution was alkaline. If a precipitate was produced, several drops of hydrochloric acid were added until it dissolved, and 1 milliliter of ammonium citrate was added. The solution was again made alkaline with ammonium hydroxide. Several drops of dithizone and 5 milliliters of carbon tetrachloride were added, and the solution was shaken vigorously for a few seconds. After a few more drops of dithizone were added, this process was repeated until the carbon tetrachloride layer was a dark red and the watery layer was yellow, indicating that an excess of dithizone had been added. When it was judged that an excess of dithizone was present, the carbon tetrachloride layer was drawn off into another separatory funnel; more carbon tetrachloride and dithizone were added to the watery mixture in the first separatory funnel, and the mixture was shaken out again. This second chloroform layer was green, indicating that practically all the zinc was obtained in the first extraction. This layer was likewise drawn off into the second separatory funnel. The second separatory funnel

contained the zinc-dithizone complex with an excess of dithizone, which was removed by shaking out two or three times with 25 milliliters of 0.3 normal ammonium hydroxide. Each time after the carbon tetrachloride settled, it was drawn off, more ammonium hydroxide added, and shaken again. When the watery layer was colorless and the carbon tetrachloride some shade of red, the washing was sufficient. The carbon tetrachloride was then drawn off into absorption cells with flat bottoms for reading in Cenco-Sheard-Sanford photelometer with a blue filter, No. 435. The machine was set at 100 with carbon tetrachloride.

A blank test was made frequently on all materials used and a correction made for zinc found in the blank test.

Determinations of zinc were made against a standard curve representing 0.5 micrograms to 10 micrograms of zinc per milliliter. These concentrations were prepared by dissolving 0.2198 milligrams of $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ in 500 milliliters of zinc-free water. One-half milliliter was diluted to 100 milliliters for the 0.5 concentration, one milliliter diluted to 100 milliliters for the 0.5 concentration, one milliliter diluted to 100 milliliters for 1 microgram, 2 for 2 micrograms, and thus continued to 10 milliliters diluted to 100 for the 10 microgram concentration. Blank corrections for zinc were made in setting up the standard curve, as ammonia and hydrochloric acid were tested and found to contain some zinc.

TABLE 5
ORIGINAL READINGS AND DETERMINATIONS
OF ZINC IN FOOD AND MILK

Day	Total Weight of Food	Sample I				Sample II			
		Ali- quot	Dilu- tion	Zinc		Ali- quot	Dilu- tion	Zinc	
				Reading on Curve	Amount in Sample*			Reading on Curve	Amount in Sample*
	gm	gm	ml	mcg/ml	mcg	gm	ml	mcg/ml	mcg
I**									
1	968.3	9.99	100	1.200	120.00	10.00	100	1.400	140.00
2	872.0	1.00	100	0.979	9.79	1.00	100	1.276	12.76
3	829.8	10.00	100	6.400	127.90	10.00	100	6.200	123.90
4	1023.8	9.99	100	1.200	120.03	9.99	100	1.250	125.00
5	657.4	1.00	100	1.250	12.50	1.00	100	1.240	12.40
6	496.1	1.00	100	0.820	8.20	0.99	100	0.800	8.00
7	598.0	1.00	100	0.640	6.39	1.00	100	0.640	6.37
Milk	605.0	10.00	100	3.000	30.50	10.00	100	3.100	31.00
II**									
1	1316.8	1.00	100	1.100	10.91	1.00	100	1.100	10.97
2	1201.4	1.00	100	1.100	11.00	1.00	100	1.100	10.98
3	865.1	1.00	100	1.400	13.94	1.00	100	1.400	13.94
4	1101.6	1.00	100	1.300	12.91	1.00	100	1.200	11.98
5	973.7	1.01	100	1.100	10.85	1.00	100	1.100	10.95
Milk	613.6	10.00	100	3.400	34.00	10.00	100	3.500	35.00

*Denotes amount in 1 and 10-gram samples.

**Denotes period of study.

TABLE 5--Continued

Zinc	Recovery Sample				Per Cent Recovery	Total Zinc in Food
Average Amount in Sample	Ali-quot	Dilu-tion	Zinc			
			Reading on Curve	Amount in Sample*		
mcg	gm	ml	mcg/ml	mcg		mg
130.00	9.99	100	1.319	131.90	95	12.58
11.27	1.00	100	1.327	13.27	100	9.81
126.00	10.00	100	6.400	128.00	105	11.28
122.52	9.99	100	1.247	124.71	109	12.54
12.41	1.00	100	1.450	14.50	103	8.16
8.09	1.00	100	1.000	10.00	95	4.79
6.38	1.00	100	0.850	8.49	106	3.17
30.70	10.00	100	3.260	32.60	105	1.86
10.94	1.00	100	1.300	12.90	98	14.42
10.99	1.00	100	1.300	12.96	98.5	13.21
13.94	1.00	100	1.600	15.97	102	12.07
12.45	1.00	100	1.450	14.41	98	13.71
10.90	1.01	100	1.300	12.86	98	10.22
34.50	10.00	100	3.650	36.50	100	2.12

TABLE 6
ORIGINAL READINGS AND DETERMINATIONS
OF ZINC IN URINE

Name	Day	Urine Ex- creted	Sample I				Sample II			
			Ali- quot	Dilu- tion	Zinc		Ali- quot	Dilu- tion	Zinc	
					Reading on Curve	Amount in Sample			Reading on Curve	Amount in Sample
		ml	ml	ml	mcg/ml	mcg	ml	ml	mcg/ml	mcg
I**										
St	1	1040	10	100	1.4	14.0	10	100	2.1	21.0
St	2	980	10	100	1.97	19.7	10	100	1.52	15.2
St	3	880	10	100	3.8	38.0	10	100	2.9	29.0
St	4	1105	10	100	1.2	12.0	10	100	0.6	6.0
Le	1	950	10	100	1.3	13.0	10	100	2.0	20.0
Le	2	670	10	100	0.5	5.0	10	100	0.6	6.0
Le	3	1270	10	100	1.4	14.0	10	100	1.0	10.0
Le	4	1060	10	100	1.1	11.0	10	100	1.3	13.0
Le	5	1095	10	100	0.5	10.0	10	100	0.51	10.2
W1	1	1200	10	100	1.7	17.0	10	100	1.5	15.0
W1	2	2040	10	100	1.4	14.0	10	100	1.0	10.0
W1	3	860	10	100	0.6	6.0	10	100	0.8	8.0
W1	4	1075	10	100	0.6	6.0	10	100	0.8	8.0
W1	5	1255	10	100	0.52	5.2	10	100	0.54	5.4
Co	1	475	10	100	0.52	5.2	10	100	0.54	5.4
Co	2	705.5	10	100	0.5	5.0	10	100	0.5	5.0
Co	3	1260	10	100	0.6	6.0	10	100	0.8	8.0
Co	4	885	10	100	1.4	14.0	10	100	1.0	10.0
Co	5	790	10	100	0.7	7.0	10	100	0.7	7.0
Ho	1	1460	10	100	0.5	5.0	10	100	0.9	9.0
Ho	2	2075	10	100	0.5	5.0	10	100	0.5	5.0
Ho	3	1900	10	100	0.6	6.0	10	100	0.8	8.0
Ho	4	1100	10	100	0.52	5.2	10	100	0.54	5.4
Ho	5	875	10	100	1.7	17.0	10	100	1.5	15.0
Go	1	350	10	100	1.0	10.0	10	100	1.4	14.0
Go	2	335	10	100	0.8	8.0	10	100	0.6	6.0
Go	3	740	10	100	1.7	17.0	10	100	1.5	15.0
Go	4	550	10	100	1.0	10.0	10	100	1.0	10.0
Go	5	520	10	100	1.0	10.0	10	100	1.0	10.0

TABLE 6--Continued

Zinc	Recovery Sample				Per Cent Recovery*	Total Zinc in Urine
Average Amount in Sample	Ali-quot	Dilu-tion	Zinc			
			Reading on Curve	Amount in Sample		
mcg	ml	ml	mcg/ml	mcg		mg
17.5	10	100	3.55	35.5	90	1.82
17.5	10	100	3.77	37.7	101	1.71
33.5	10	100	5.2	52.0	93	2.95
9.0	10	100	2.75	27.5	93	0.83
16.5	10	100	3.4	34.0	88	1.57
5.5	10	100	2.5	25.0	97	0.37
12.0	10	100	3.1	31.0	95	1.53
12.0	10	100	1.8	18.0	90	1.27
10.1	10	100	2.19	21.9	109	2.21
16.0	10	100	3.52	35.2	91	1.92
12.0	10	100	3.1	31.0	95	2.56
7.0	10	100	2.5	25.0	90	0.60
7.0	10	100	2.6	26.0	95	0.75
5.3	10	100	2.72	27.2	109	0.67
5.3	10	100	2.70	27.0	108	0.25
5.0	10	100	2.70	27.0	110	0.35
7.0	10	100	2.60	26.0	95	0.88
12.0	10	100	3.0	30.0	90	1.06
7.0	10	100	2.6	26.0	95	0.53
7.0	10	100	2.6	26.0	95	1.02
5.0	10	100	2.5	25.0	100	1.04
7.0	10	100	2.5	25.0	90	1.32
5.3	10	100	2.7	27.0	108	0.58
16.0	10	100	3.52	35.2	96	1.40
12.0	10	100	3.2	32.0	90	0.42
7.0	10	100	2.7	27.0	100	0.235
16.0	10	100	3.52	35.2	96	1.18
10.0	10	100	2.9	29.0	95	0.55
10.0	10	100	3.0	30.0	100	0.52

TABLE 6--Continued

Name	Day	Urine Ex- creted	Sample I				Sample II			
			Ali- quot	Dilu- tion	Zinc		Ali- quot	Dilu- tion	Zinc	
					Reading on Curve	Amount in Sample			Reading on Curve	Amount in Sample
		ml	ml	ml	mcg/ml	mcg	ml	ml	mcg/ml	mcg
Yo	1	500	10	100	1.0	10.0	10	100	1.4	14.0
Yo	2	550	10	100	0.7	7.0	10	100	0.7	7.0
Yo	3	640	10	100	1.0	10.0	10	100	1.0	10.0
Yo	4	585	10	100	1.0	10.0	10	100	1.2	12.0
Yo	5	900	10	100	0.7	7.0	10	100	0.7	7.0
II**										
Mc	1	1440	10	100	0.52	5.2	10	100	0.54	5.4
Mc	2	1455	10	100	0.5	5.0	10	100	0.5	5.0
Mc	3	1250	10	100	0.52	5.2	10	100	0.54	5.4
Mc	4	1040	10	100	0.6	6.0	10	100	0.8	8.0
Mc	5	795.5	10	100	1.5	15.0	10	100	1.7	17.0
Fr	1	1180	10	100	0.5	5.0	10	100	0.5	5.0
Fr	2	1365	10	100	0.6	6.0	10	100	0.8	8.0
Fr	3	1090	10	100	0.52	5.2	10	100	0.54	5.4
Fr	4	1350	10	100	0.5	5.0	10	100	0.7	7.0
Fr	5	1075	10	100	1.0	10.0	10	100	1.0	10.0
Ko	1	1200	10	100	0.6	6.0	10	100	0.6	6.0
Ko	2	1365	10	100	0.5	5.0	10	100	0.7	7.0
Ko	3	1745	10	100	1.0	10.0	10	100	1.0	10.0
Ko	4	1090	10	100	0.6	6.0	10	100	0.7	7.0
Ko	5	1550	10	100	0.6	6.0	10	100	0.8	8.0
Br	1	980	10	100	0.5	5.0	10	100	0.5	5.0
Br	2	645	10	100	0.7	7.0	10	100	0.5	5.0
Br	3	1020	10	100	1.0	10.0	10	100	1.2	12.0
Br	4	1210	10	100	0.5	5.0	10	100	0.6	6.0
Br	5	1040	10	100	1.0	10.0	10	100	1.2	12.0
Bi	1	1250	10	100	0.5	5.0	10	100	0.6	6.0
Bi	2	1090	10	100	0.5	5.0	10	100	0.6	6.0
Bi	3	1155	10	100	0.6	6.0	10	100	0.7	7.0
Bi	4	1150	10	100	0.8	8.0	10	100	1.0	10.0
Bi	5	750	10	100	1.0	10.0	10	100	1.2	12.0

TABLE 6--Continued

Zinc	Recovery Sample				Per Cent Recovery*	Total Zinc in Urine
Average Amount in Sample	Aliquot	Dilution	Zinc			
			Reading on Curve	Amount in Sample		
mcg	ml	ml	mcg/ml	mcg		mg
12.0	10	100	3.2	32.0	90	0.60
7.0	10	100	2.6	26.0	95	0.39
10.0	10	100	2.8	28.0	90	0.64
11.0	10	100	3.3	33.0	110	1.29
7.0	10	100	2.5	25.0	90	0.63
5.3	10	100	2.7	27.0	108	0.76
5.0	10	100	2.5	25.0	100	0.73
5.3	10	100	2.7	27.0	108	0.66
7.0	10	100	2.5	25.0	90	0.73
16.0	10	100	3.52	35.2	96	1.27
5.0	10	100	2.5	25.0	100	0.59
7.0	10	100	2.5	25.0	90	0.96
5.3	10	100	2.7	27.0	108	0.58
6.0	10	100	2.5	25.0	95	0.88
10.0	10	100	2.9	29.0	95	1.08
6.0	10	100	2.5	25.0	95	0.72
6.0	10	100	2.5	25.0	90	0.82
10.0	10	100	3.0	30.0	100	1.75
6.5	10	100	2.6	26.0	98	0.71
7.0	10	100	2.6	26.0	95	1.01
5.0	10	100	2.3	23.0	90	0.49
6.0	10	100	2.4	24.0	90	0.32
11.0	10	100	3.0	30.0	95	1.12
5.5	10	100	2.4	24.0	93	0.67
11.0	10	100	2.9	29.0	90	1.20
5.5	10	100	2.3	23.0	89	0.66
5.5	10	100	2.4	24.0	93	0.59
6.5	10	100	2.4	24.0	87	0.75
9.0	10	100	2.8	28.0	95	1.05
11.0	10	100	3.0	30.0	95	0.83

TABLE 6--Continued

Name	Day	Urine Ex- creted	Sample I				Sample II			
			Ali- quot	Dilu- tion	Zinc		Ali- quot	Dilu- tion	Zinc	
					Reading on Curve	Amount in Sample			Reading on Curve	Amount in Sample
		ml	ml	ml	mcg/ml	mcg	ml	ml	mcg/ml	mcg
As	1	820	10	100	0.5	5.0	10	100	0.5	5.0
As	2	1065	10	100	0.8	8.0	10	100	0.5	5.0
As	3	1115	10	100	1.0	10.0	10	100	1.0	10.0
As	4	1110	10	100	0.6	6.0	10	100	0.5	5.0
As	5	1275	10	100	1.0	10.0	10	100	2.0	20.0

*Recovery added to milliliters taken for color development.

**Denotes period of study.

TABLE 6--Continued

Zinc	Recovery Sample				Per Cent Recovery*	Total Zinc in Urine
Average Amount in Sample	Aliquot	Dilution	Zinc			
			Reading on Curve	Amount in Sample		
mcg	ml	ml	mcg/ml	mcg		mg
5.0	10	100	2.4	24.0	95	0.41
6.5	10	100	2.4	24.0	88	0.69
10.0	10	100	2.8	28.0	90	1.115
5.5	10	100	2.4	24.0	93	0.61
15.0	10	100	3.52	35.2	101	1.91

TABLE 7
ORIGINAL READINGS AND DETERMINATIONS
OF ZINC IN FECES

Name	Total Weight of Feces	Sample I				Sample II			
		Ali- quot	Dilu- tion	Zinc		Ali- quot	Dilu- tion	Zinc	
				Reading on Curve	Amount in Sample			Reading on Curve	Amount in Sample
	gm	gm	ml	mcg/ml	mcg	gm	ml	mcg/ml	mcg
I*									
W1	272.3	2.0	100	11.71	117.1	2.0	100	11.73	117.3
Co	356.7	2.0	100	11.8	118.0	2.0	100	11.4	114.0
Ho	267.2	2.0	100	11.38	113.8	2.0	100	11.58	115.8
Le	446.65	2.0	100	11.56	115.6	2.0	100	11.6	116.0
St	384.1	2.0	100	11.8	118.0	2.0	100	11.6	116.0
Go	474.2	2.0	100	11.8	118.0	2.0	100	11.6	116.0
Yo	501.7	2.0	100	11.8	118.0	2.0	100	11.6	116.0
II*									
Mc	650.6	2.0	100	11.8	118.0	2.0	100	11.6	116.0
Br	373.7	2.0	100	11.8	118.0	2.0	100	11.6	116.0
Fr	553.4	2.0	100	11.38	113.8	2.0	100	11.4	114.0
Bi	670.3	2.0	100	11.71	117.1	2.0	100	11.73	117.3
Ko	571.6	2.0	100	11.8	118.0	2.0	100	11.6	116.0
As	555.1	2.0	100	11.8	118.0	2.0	100	11.4	114.0

*Denotes period of study.

TABLE 7--Continued

Zinc	Recovery Sample				Per Cent Recovery	Total Zinc in Feces
Average Amount in Sample	Ali-quot	Dilu-tion	Zinc			
			Reading on Curve	Amount in Sample		
mcg	gm	ml	mcg/ml	mcg		mg
117.2	2.0	100	11.9	119.0	90	16.01
116.0	2.0	100	11.8	118.0	100	20.69
113.9	2.0	100	11.58	115.8	95	20.66
115.3	2.0	100	11.76	117.6	105	26.04
117.0	2.0	100	11.91	119.1	105	22.47
117.0	2.0	100	11.92	119.2	110	27.69
117.0	2.0	100	11.91	119.1	105	29.35
117.0	2.0	100	11.92	119.2	110	38.09
117.0	2.0	100	11.91	119.1	105	21.66
113.9	2.0	100	11.58	115.8	95	31.52
117.2	2.0	100	11.9	119.0	90	39.28
117.0	2.0	100	11.92	119.2	110	33.44
116.0	2.0	100	11.8	118.0	100	32.20

TABLE 8
MILK CONSUMED BY EACH SUBJECT DURING
EACH OF THE BALANCE PERIODS

Balance	Days	Total Cups	Average Cups/Day
Period I			
Co	5	3	0.6
Wi	5	4	0.8
Ho	5	6	1.2
St	5	2	0.4
Le	5	4	0.8
Yo	5	6	1.2
Go	5	5	1.0
Period II			
As	5	7.5	1.5
Bi	5	8.5	1.7
Br	5	7.0	1.4
Fr	5	8.5	1.7
Ko	5	8.5	1.7
Mc	5	5.0	1.0

TABLE 9

MENUS USED DURING THE TWO FIVE-DAY PERIODS

Meal	Monday 10/27 11/3	Tuesday 10/28 11/4	Wednesday 10/29 11/5	Thursday 10/30 11/6	Friday 10/31 11/7
Break- fast	Orange Juice Apple Pan- cakes ¹ Butter Maple Syrup	Orange Juice Waffles ³ Sausage Maple Syrup Butter Milk	Scrambled Eggs Bacon Biscuits ¹ Butter Jelly Milk	Scrambled Eggs Bacon Muffins ¹ Butter Jelly Milk	Scrambled Eggs Ham Bran Muffins ¹ Butter Jelly Milk
Lunch	Deviled Eggs Lima Beans Corn Biscuits ¹ Butter Banana Pudding ² Iced Tea	Salmon Loaf ² Spinach and Eggs Pineapple- Cheese Salad Rolls Butter Oatmeal Cookies ⁴ Green Grapes Iced Tea	Baked Ham Mashed Potatoes Asparagus Carrot- Pineapple- Jello-Nut Salad Rolls Butter Oatmeal Cookies ⁴ Orange Sherbet Iced Tea	Spanish Rice ⁵ Green Beans Carrot, Cel- ery Strips Cornbread ⁷ Butter Pineapple Upside Down Cake ¹ Iced Tea	Baked Beans, Weiners ⁶ Pear, Cheese Salad Bread Applesauce Bars ¹

Dinner	Meat Patties ² Potato Salad Harvard Beets Rolls Butter Peach Halves Peanut Butter Cookies ¹ Iced Tea	Macaroni ¹ Cheese ⁸ Tomatoes English Peas Muffins ⁴ Butter Butterscotch Pudding Iced Tea	Spam Potato Patties ¹⁰ Tomatoes Raisin Muffins ⁴ Butter Peanut Butter Cookies ¹ Iced Tea	Meat Loaf ² Baked Potatoes, Butter Lettuce, Tomato Salad Rolls Baked Apples Iced Tea	Creamed Ham on Toast ⁹ Cottage Cheese Lime Jello Plain Cake ¹ Strawberry Ice Cream Milk
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¹One cup of Multi-Purpose Food, Formula B, was substituted for one cup of flour in Period II.

²Supplemented with two cups cooked Multi-Purpose Food, Formula B, during Period II.

³One and one-half cups of Multi-Purpose Food, Formula B, were substituted for one and one-half cups of flour in Period II.

⁴One-half cup of Multi-Purpose Food, Formula B, was substituted for one-half cup of flour in Period II.

⁵One cup of cooked Multi-Purpose Food, Formula B, was substituted for one cup of cooked rice in Period II.

⁶One cup of Multi-Purpose Food, Formula B, was substituted for one cup of beans in Period II.

⁷One cup of Multi-Purpose Food, Formula B, was substituted for one cup of cornmeal in Period II.

⁸Supplemented with one and one-half cups of Multi-Purpose Food, Formula B, in Period II.

⁹One tablespoon of Multi-Purpose Food, Formula B, was substituted for one tablespoon of flour in Period II.

¹⁰Two cups of cooked Multi-Purpose Food, Formula B, were substituted for two cups of shredded raw potato in Period II.

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