TABLE II.

FACTOR ANALYSIS OF THE RELATIONS AMONG INDICES OF BONE DENSITY AND/OR QUALITY, ANTHROPOMETRIC CHARACTERISTICS, AND PARAMETERS OF BONE REMODELING CALCULATED FROM SPERMAN'S CORRELATION MATRIX*

						Factor loadings		
		Variance				Factor 1	Factor 2	Factor 3
Factor no.	Eigenvalue	%	Cumulative %	Variable	Communality	Density, structure, and fragility	Elasticity	Remodeling
1	3.33	46.8	46.8	Number of fractures	0.127	−0.327	−0.110	0.090
2	1.58	22.2	69.0	BMI	0.248	0.474	−0.126	0.085
3	1.21	17.0	86.0	BMD at the hip	0.651	0.793	0.144	−0.023
4	0.42	5.9	91.9	BMD at the spine	0.428	0.623	0.044	0.195
5	0.38	5.3	97.2	BUA	0.800	0.745	0.474	−0.143
6	0.15	2.1	99.3	Stiffness index	0.970	0.448	0.873	−0.089
7	0.05	0.7	100.0	SOS	0.867	0.010	0.931	0.018
8	0.00	0.0	100.0	Serum (ucOC/OC)	0.108	0.015	0.328	0.020
9	0.00	0.0	100.0	Serum ICTP	0.488	0.154	0.106	0.673
10	0.00	0.0	100.0	Urinary DPD (2 h)	0.321	0.008	−0.100	0.558
11	0.00	0.0	100.0	Serum OC	0.501	−0.045	0.077	0.702
12	0.00	0.0	100.0	Years since menopause	0.601	−0.483	−0.281	0.537

* VARIMAX rotation followed factor analysis. Factor loadings represent correlation coefficients between factors and variables, and communality is a measure of the shared variability between the variable and all remaining variables. The number of factors was determined with Kaiser's criterion (only the factors with eigenvalue greater than 1 were included). In this case it means that 86% of the total variability from 12 original variables was explained by only three factors. The boldface text denotes the relevant information in individual statistical parameters.
BMD, bone mineral density; BUA, broadband ultrasound attenuation; DPD, deoxypyridinoline; ICTP, carboxy-terminal telopeptide of type 1 procollagen; OC, osteocalcin; SOS, speed of sound; ucOC, undercarboxylated osteocalcin

problem of cross-reactivity between OC and the molecule fragments also may have limited the interpretation of this parameter.

To summarize, the present study did not confirm associations between serum ucOC and bone density in early postmenopausal women. However, it indicated a moderate relation of ucOC to bone elasticity. Association analyses in women subdivided according to fracture type are needed.

Ivana Žofková, MD, PhD, DSc
Martin Hill, PhD, DSc
Vladimír Palička, MD, PhD
Institute of Endocrinology
Prague, Czech Republic
Institute of Clinical Biochemistry and Diagnostics
Charles University Teaching Hospital
Hradec, Králové, Czech Republic

REFERENCES

1. Sokoll LJ, Booth SL, O'Brien ME, Davidson KW, Tsaioun KI, Sadowski JA. Changes in serum osteocalcin, plasma phylloquinone, and urinary γ-carboxy glutamic acid in response to altered intakes of dietary phylloquinone in human subjects. Am J Clin Nutr 1981;65:779
2. Vergnaud P, Garnero P, Meunier PJ, Bréart G, Kamihagi K, Delmas PD. Undercarboxylated osteocalcin measured with a specific immunoassay predicts hip fracture in elderly women: the EPIDOS study. J Clin Endocrinol Metab 1997;82:719
3. Liu G, Peacock M. Age-related changes in serum undercarboxylated osteocalcin and its relationships with bone density, bone quality, and hip fracture. Calcif Tissue Int 1998;62:286
4. Ferland G. The vitamin K–dependent proteins: an update. Nutr Rev 1998;56:223
5. Szulc P, Arlot M, Chapuy M-C, Duboeuf F, Meunier PJ, Delmas PD. Serum undercarboxylated osteocalcin correlates with hip bone mineral density in elderly women. J Bone Miner Res 1994;9:1591
6. Szulc P, Chapuy M-C, Meunier PJ, Delmas PD. Serum undercarboxylated osteocalcin is a marker of the risk of hip fracture in elderly women. J Clin Invest 1993;91:1769
7. Szulc P, Chapuy M-C, Meunier PJ, Delmas PD. Serum undercarboxylated osteocalcin is a marker of the risk of hip fracture: a three year follow-up study. Bone 1996;18:487
8. Luukinen H, Käkönen S-M, Pettersson K, et al. Strong prediction of fractures among older adults by the ratio of carboxylated to total serum osteocalcin. J Bone Miner Res 2000;15:2473
9. Grundberg CM, Nieman SD, Abrams S, Rosen H. Vitamin K status and bone health: an analysis of methods for determination of undercarboxylated osteocalcin. J Clin Endocrinol Metab 1998;83:3258
10. Ravn P, Fledelius C, Rosenquist C, Overgaard K, Christiansen C. High bone turnover is associated with low bone mass in both pre- and postmenopausal women. Bone 1996;19:291

Zinc, Copper, and Iron in Plasma and Tissues After Intestinal Ischemia and Reperfusion in the Rat

INTRODUCTION

The plasma and tissue content of trace minerals such as zinc, copper, and iron are significantly affected by stress,[1] burns,[2] and

The research reported in this article was supported by the Human Nutrition Research Funds of Texas.

Correspondence to: V. Bruce Grossie, Jr., PhD, Department of Nutrition and Food Sciences, Texas Woman's University, PO Box 425888, Denton, TX 76204-5888, USA. E-mail: bgrossie@mailtwu.edu

infection.[3] An inflammatory response has been reported to decrease the serum concentration of zinc and iron and increase the blood levels of copper.[4] A combined insult of burn and infection[2] results in an exacerbation of the zinc response of the host.

Intestinal ischemia is a common event in traumatic injury or major surgery. Whereas extended periods of ischemia impair organ function, restoration of blood flow may incite an inflammatory response. The function of the gut[5] and of remote tissues such as liver, lung,[6] and bone marrow[7] are adversely affected by intestinal ischemia followed by reperfusion (I/R). The decreased bone marrow function was also associated with a decrease in the immune function of the host.[7]

MATERIALS AND METHODS

This study was designed to measure the temporal change in tissue zinc, copper, and iron content after 45 min of intestinal I/R. Sprague Dawley male rats ($n = 40$, 125–150 g) were housed in acid-washed, stainless steel metabolic cages and received the AIN 93-G diet (Dyets Inc., Bethlehem, PA, USA) and deionized water ad libitum until the ischemic phase of the study. The experiments were performed in accordance with the National Research Council, Institute for Laboratory Animal Resources, Guide for the Care and Use of Laboratory Animals and were approved by the Animal Use Committee of Texas Woman's University.

Rats were anesthetized (2% isoflurane at 2 L/min in 100% oxygen); intestinal ischemia was induced by superior mesenteric artery (SMA) occlusion, as previously described.[8] At 0, 0.5, 2, 5, and 24 h after the start of intestinal reperfusion, rats were anesthetized and exsanguinated through the abdominal aorta, with the blood collected into a heparinized vacutainer tube. The intestine, liver, thymus, and lung were carefully removed using stainless steel instruments. Tissue and plasma zinc, copper, and iron contents were determined with a Varian Spectr AA-40 atomic absorption spectrometer (Varian, Inc., Palo Alto, CA, USA).

Statistical analysis was done with SPSS 8.0 for Windows (1998). Means for each organ were compared by two-way analysis of variance. If the analysis for the group was significant ($P < 0.05$), the individual means were compared with Fisher's protected least significant difference (PLSD) post hoc analysis.

RESULTS

The changes in plasma concentration (μg/mL) and liver and spleen zinc content (μg/g wet tissue) are shown in Table I. The plasma zinc concentration at 5 h after I/R or sham surgery was significantly decreased as compared with the concentration before, or at 0.5 (data not shown) and 2 h after reperfusion. The plasma concentration was increasing but had not reached baseline levels after 24 h of reperfusion. The zinc content of liver and spleen was similar for the first 5 h but was significantly higher at 24 h after reperfusion. There was no significant temporal effect on the zinc content of the small intestine, thymus, or lung of sham or I/R rats.

The copper content of the liver was significantly increased at 24 h after I/R as compared with the sham group (data not shown); no other temporal effect was observed. I/R, however, did not affect the copper content of the plasma, spleen, small intestine, thymus, and lung. There was no temporal or I/R effect on the iron content of plasma and organs (data not shown).

TABLE I.

EFFECT OF ISCHEMIA AND REPERFUSION ON THE ZINC CONTENT OF RAT PLASMA, LIVER, AND SPLEEN

Hours after release of SMA occlusion	Status of intestinal reperfusion*	Plasma (μg/mL)†	Liver (μg/g)†	Spleen (μg/g)†
0	Sham	7.9 ± 1.1	144 ± 26	831 ± 208
	I/R	6.0 ± 4.2	87 ± 27	902 ± 181
2.0	Sham	7.2 ± 1.8	134 ± 61	831 ± 157
	I/R	6.7 ± 1.1	135 ± 66	782 ± 314
5.0	Sham	3.3 ± 0.6‡	142 ± 45	686 ± 208
	I/R	3.2 ± 0.5‡	185 ± 102	914 ± 139
24	Sham	6.4 ± 2.2	312 ± 52‡	1354 ± 433‡
	I/R	5.7 ± 2.9	319 ± 63‡	1298 ± 367‡

* Each group had four animals except the 24-h sham group, which had six.
† Data are mean ± standard deviation.
‡ Means differ significantly ($P < 0.05$) compared with all previous time points.
I/R, ischemia then reperfusion; SMA, superior mesenteric artery.

DISCUSSION

The effect of intestinal I/R on the plasma and tissue zinc, copper, and iron contents have not been determined. Inflammation in response to many factors[1–4] has been shown to alter the plasma concentration of trace elements. The results of the present study suggest that the major effect of intestinal I/R is on the plasma and tissue contents of zinc. Lindman et al.[8] reported that major surgery and other tissue injuries such as acute myocardial infarction and acute *Klebsiella pneumonia* result in a decrease in plasma zinc that returns to normal after healing or clearing of the infection. Brambila et al.[9] demonstrated that a 4-cm abdominal incision results in a decrease in serum levels between 15 and 20 h after surgery, which is accompanied by an increase in the liver zinc content. This differed from our study in that the surgical incision was greater in our study. In addition, the incision in our study, although covered with saline-soaked gauze and a plastic sheet, was not closed until 45 min after the incision to allow access to remove the SMA clamp. The recovery of serum zinc in the study by Brambila et al.[9] was evident by 24 h after surgery. These differences between the results of our study and those of Brambila et al. suggest that the greater the stress, the greater the depression of plasma zinc and the slower the recovery of zinc content. Our data, however, will not allow us to make any further conclusions as to the kinetics of this recovery.

The results confirm that tissue injury such as that from major surgery will have a significant impact on the plasma and tissue contents of zinc. The results suggest that zinc is more susceptible to injury than is copper or iron in plasma and tissues. The significance of these alterations is currently being investigated.

ACKNOWLEDGMENTS

The authors thank Krista Cohlmia, PhD, for her advice concerning the statistical analysis of the data.

V. Bruce Grossie, Jr, PhD
W. Russ Kennedy, PhD
Dorice Narins, PhD
Department of Nutrition and Food Sciences
Texas Woman's University
Denton, Texas, USA

REFERENCES

1. Singh A, Smoak BL, Patterson KY, LeMay LG, Veillon C, Deuster PA. Biochemical indices of selected trace minerals in man: effect of stress. Am J Clin Nutr 1991;53:126

2. Powanda MC, Villarreal Y, Rodriguez E Jr, Braxton G III, Kennedy CR. Redistribution of zinc in burned and burned infected rats. Proc Soc Exp Biol Med 1980;163:296
3. Shanbhogue LKR, Paterson N. Effect of sepsis and surgery on trace minerals. JPEN 1990;14:287
4. Galloway P, McMillan DC, Sattar N. Effect of the inflammatory response on trace element and vitamin status. Ann Clin Biochem 2000;37:289
5. Grossie VB Jr, Weisbrodt NW, Moore FA, Moody F. Ischemia/reperfusion-induced disruption of rat small intestine transit is reversed by total enteral nutrition. Nutrition 2001;17:939
6. Poggetti RS, Moore FA, Moore EE, Koeike K, Banerjee A. Simultaneous liver and lung injury following gut ischemia is mediated by xanthine oxidase. J Trauma 1992;32:723
7. Fontes B, Moore FA, Moore EE, et al. Gut ischemia induces bone marrow failure and increases risk of infection. J Surg Res 1994;57:505
8. Lindeman RD, Bottomley DG, Cornelson RL Jr, Jacobs LA. Influence of acute tissue injury on zinc metabolism in man. J Lab Clin Med 1972;79:452
9. Brambila E, Munoz-Sanchez JL, Albores A, Waalkes M. Early effects of surgery on zinc and metallothionein levels in female rats. Biol Trace Elem Res 1999;70:173