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Iodine and/or Selenium Deficiency Alters Tissue Distribution Pattern of Other Trace Elements in Rats

BELMA GIRAY,[1] JACQUELINE RIONDEL,[2] JOSIANNE ARNAUD,[2] VERONIQUE DUCROS,[2] ALAIN FAVIER,[2] AND FILIZ HINCAL*,[1]

[1]Department of Toxicology, Faculty of Pharmacy, Hacettepe University, Ankara, Turkey; and [2]Laboratory of Biology of Oxidative Stress, Faculty of Pharmacy, Joseph Fourier University, Grenoble, France

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ABSTRACT

Tissue distribution of Fe, Mn, Cu, and Zn, the essential trace elements associated with oxidant and/or antioxidant processes, was examined in iodine- and/or selenium-deficient rats (ID, SeD, ISeD). Fe and Mn were the most affected minerals in all types of deficiency states. Mn levels decreased significantly in the liver in all deficiency states (approx 20–30%), in the heart in ID and SeD rats (approx 30–35%) and in the testis in ID rats (approx 15%). Whereas Mn enhancement was noted in kidney (approx 45%) and plasma in SeD and ISeD (approx 20% and 50%, respectively) animals. However, most striking alterations were seen with Fe. Significant elevation of Fe concentrations were observed in all deficiency states in the kidney (approx 90–125%) and heart (approx 20–25%), and in the liver in SeD (approx 35%) and ISeD (approx 75%) rats, whereas significant (approx 20%) Fe enhancement in the testis was observed only in ISeD animals. Lower Cu (approx 10–15%) and higher Zn (approx 10–20%) concentrations in heart tissues in all deficiency states were found; higher Zn (approx 20–35%) in the kidney of SeD and ISeD rats, and lower Cu in the testis of SeD animals were observed. In brain tissue, no alteration was seen in Fe, Mn, and Zn content, however, significantly increased (approx 15–20%) Cu concentrations were noted in all deficiency states. The results of this study indicated that iodine and/or selenium deficiency may modify the distribution and the homeostasis of other minerals.

Index Entries: Selenium; iodine; thyroid hormones; manganese; iron; copper; zinc.

*Author to whom all correspondence and reprint requests should be addressed.

INTRODUCTION

A considerable amount of evidence suggest that a deficiency of a micronutrient influences the body balance of the others; thus, the homeostasis of essential trace elements are closely interrelated in various organs and tissues *(1,2).* There are reports indicating the interaction of selenium (Se) with other minerals in biological systems. Although high Fe intakes in rats do not adversely alter tissue distribution or concentrations of Se and liver glutathione peroxidase (GSH-Px) activity *(3,4),* a deficiency of Fe decreases tissue Se concentrations and GSH-Px activity in the erythrocyte and liver *(5).* The synthesis of Se–GSH-Px protein is reported to be decreased during Fe deficiency, possibly the result of pretranslational regulation *(5).* There are also reports pointing out a possible role for dietary Se in moderating Fe metabolism and showing the occurrence of a modest but constant increase in expression of genes for transferrin, transferrin receptor, and iron-regulatory protein in Se-deficient rat liver *(6).* In addition, there is some evidence to suggest the presence of a specific interaction between tissue Se and Mn *(7,8).* Mn deficiency decreases Se levels in all tissues of pigs *(7),* whereas Se deficiency induces mineral imbalance with a decrease in tissue Mn *(8,9).*

Several findings indicate the involvement of thyroid hormones in tissue mineral metabolism, especially in the liver *(10–12),* whereas normal thyroid status is dependent on the presence of various trace elements for both the synthesis and metabolism of thyroid hormones. In addition to iodine (I_2), the essential component of thyroid hormones thyroxine (T_4) and 3,3′,5-tri-iodothyronine (T_3), Se has an important role for normal thyroid hormone metabolism as an integral component of iodothyronine deiodinases *(13–15)* as well as GSH-Px. The roles of Fe, Zn, and Cu in the thyroid are less well defined, but suboptimal or supraoptimal dietary intakes of all these elements can adversely affect thyroid hormone metabolism *(16).* On the other hand, Cu, Zn, Se, and Mn are well-known essential components of antioxidant defenses, whereas iron acts as an important mediator in cell injury accompanying oxidative stress. Because all minerals have important roles in the regulation of critical cellular processes, modification of transcription factors and receptors, or as cofactors of critical proteins, any alteration in the mineral distribution may lead a pathophysiological state. Inadequate dietary intake of trace elements is responsible for numerous diseases affecting circulatory, respiratory, nervous, endocrine, immune, and reproductive systems *(17).*

This study was, therefore, undertaken to elucidate the distribution pattern of Mn, Cu, Zn, and Fe levels, the essential minerals associated with oxidant and/or antioxidant processes in different tissues of I_2- and/or Se-deficient rats.

MATERIALS AND METHODS

Animals and Diets

Male Wistar rats supplied by RFFA Credo Animal Breeding Center (Saint Germain, France), and 3-wk-old rats (54 ± 2 g) were used in all experiments. The animals were housed as a group in plastic cages with stainless-steel grid tops and the cages were placed in a room with controlled temperature (23±2°C), humidity (50%), and a 12-h light–dark cycle. Six animals were used for each experimental group. Feeding period was 5 wk and animals received ad libitum diet and water. Body weight was monitored weekly.

The experimental groups consisted of the *control group* (C), fed with regular diet and drinking water, the I_2-deficient and Se-normal group (ID), received regular diet and drinking water containing 1% sodium perchlorate, the I_2-normal and Se-deficient group (SeD), fed with a synthetic diet containing less than 0.005 mg Se/kg and received regular drinking water, and the I_2 and Se-deficient group (ISeD), received both Se-deficient diet and drinking water containing 1% sodium perchlorate.

Decapitation was performed under Nembutal anesthesia at the end of the feeding period. Blood samples were collected into heparinized tubes. Plasma was separated after centrifugation at 800*g* for 15 min. The liver, brain, kidney, heart, and testis were removed and frozen immediately in liquid nitrogen before storage at –80°C.

Thyroid Hormones Analysis

Thyroid hormone status was determined by measuring the plasma TSH, total T_4 (TT_4), total T_3 (TT_3), free T_4 (FT_4), and free T_3 (FT_3) concentrations by radioimmunoassay using commercial kits supplied by Elecsys.

Mineral Analysis

Tissue Se concentrations were determined by a gas chromatographic–mass spectrometric method using the isotopic dilution technique *(18).* Tissue concentrations of Mn, Fe, Cu, and Zn and plasma concentrations of Se, Mn, and Fe were determined by electrothermal atomic absorption spectrometry (Model 3030 [Perkin-Elmer] with a Zeeman background correction, a HGA 600 graphite furnace). A flame atomic absorption spectrophotometer was used for the measurement of Zn and Cu levels in plasma (Model 560; Perkin-Elmer, Überlingen, Germany). Standard Reference Materials [NIST-NBS-1577b (bovine liver) and BCR no. 184 (bovine muscle)] were used for analytical quality control.

Table 1
Thyroid Hormone Parameters in Selenium- and/or Iodine-Deficient Rats

GROUP	TSH (ng/ml)	TT_4 (nmol/L)	TT_3 (nmol/L)
C (n=6)	3.2 ± 0.7 [a]	57.7 ± 3.9 [a]	0.97 ± 0.08 [a]
ID (n=6)	29.3 ± 8.4 [b]	13.3 ± 1.2 [b]	0.57 ± 0.05 [b]
SeD (n=6)	3.7 ± 1.1 [a]	73.2 ± 11.5 [c]	0.85 ± 0.08 [c]
ISeD (n=6)	41.0 ± 2.0 [c]	18.3 ± 5.3 [d]	0.87 ± 0.31[ac]

[a,b,c,d] Values in columns not sharing a common superscript differ significantly, $p<0.05$. Values are given as mean ±SD.

Statistical Analysis

Experimental data were analyzed with one-way analysis of variance (ANOVA) followed by the Student's *t*-test ($p<0.05$). Values are given as mean±SD.

RESULTS

Thyroid Hormones

In ID rats, I_2 deficiency was confirmed by a significant increase in plasma TSH. Se deficiency alone had no significant effect on plasma TSH. The highest TSH concentrations occurred in rats deficient in both Se and I_2. Plasma TT_4 concentrations significantly increased in Se deficiency and decreased in I_2 deficiency compared to the values of control rats. A slight but significant enhancement was noted in ISeD compared to ID. TT_3 concentrations decreased in ID and SeD (*see* Table 1). (Because the same trend was observed for FT_4 and FT_3, only the total values are included in the text.)

Mineral Status

As expected, Se levels were found to be significantly lower in plasma and all tissues examined in SeD and ISeD, but the decrease was only approx 35% in the brain and approx 45% in the testis (*see* Table 2).

There was no significant alteration in plasma and brain Fe concentrations. However, hepatic Fe levels significantly increased in SeD (approx 35%) and ISeD (approx 75%); and kidney Fe levels increased in all deficiency states (approx 90% in ID and SeD, approx 125% in ISeD). The Fe content of the heart was also increased (approx 20–25%) in all deficiency states, whereas significant (approx 20%) Fe enhancement was seen only in the testis of ISeD animals (*see* Fig 1).

Table 2
Tissue Distribution of Minerals in Se- and/or I_2- Deficient Rats

TISSUE	GROUP	LIVER	BRAIN	KIDNEY	HEART	TESTIS	PLASMA*
Se (μg/g tissue)	C (n=6)	0.6 ± 0.10 [a]	0.13 ± 0.008 [a]	0.8 ± 0.08 [a]	0.27 ± 0.004 [a]	0.72 ± 0.05 [a]	3.91 ± 0.45 [a]
	ID (n=6)	0.8 ± 0.07 [a]	0.14 ± 0.009 [a]	0.8 ± 0.04 [a]	0.26 ± 0.008 [a]	0.74 ± 0.02 [a]	3.74 ± 0.22 [a]
	SeD (n=6)	0.02 ± 0.01 [b]	0.09 ± 0.005 [b]	0.13 ± 0.02 [b]	0.05 ± 0.007 [b]	0.38 ± 0.03 [b]	0.11 ± 0.03 [b]
	ISeD (n=6)	0.03 ± 0.01 [b]	0.10 ± 0.022 [b]	0.16 ± 0.05 [b]	0.05 ± 0.01 [b]	0.38 ± 0.06 [b]	0.16 ± 0.02 [b]
Cu (μg/g tissue)	C (n=6)	4.1 ± 0.2 [a]	1.5 ± 0.06 [a]	5.4 ± 0.9 [a]	4.74 ± 0.07 [a]	1.40 ± 0.04 [a]	14.1 ± 1.7 [a]
	ID (n=6)	4.1 ± 0.2 [a]	1.7 ± 0.11 [bc]	5.3 ± 0.6 [a]	4.32 ± 0.25 [b]	1.45 ± 0.07 [a]	16.8 ± 1.6 [a]
	SeD (n=6)	4.1 ± 0.2 [a]	1.7 ± 0.09 [b]	5.1 ± 0.6 [a]	4.35 ± 0.35 [b]	1.28 ± 0.05 [b]	16.0 ± 2.0 [a]
	ISeD (n=6)	4.2 ± 0.2 [a]	1.8 ± 0.08 [c]	4.4 ± 0.3 [a]	4.12 ± 0.26 [b]	1.39 ± 0.06 [a]	15.3 ± 2.2 [a]
Zn (μg/g tissue	C (n=6)	27.5 ± 1.3 [a]	12.6 ± 2.3 [a]	17.8 ± 1.3 [a]	20.7 ± 1.1 [a]	27.6 ± 0.5 [a]	19.0 ± 1.2 [a]
	ID (n=6)	28.7 ± 2.0 [a]	12.3 ± 1.2 [a]	20.0 ± 2.3 [ab]	22.8 ± 1.6 [bc]	23.2 ± 0.9 [a]	18.0 ± 1.0 [a]
	SeD (n=6)	26.0 ± 1.4 [a]	12.5 ± 1.0 [a]	21.6 ± 2.0 [b]	21.7 ± 1.9 [ac]	23.3 ± 0.9 [a]	19.8 ± 2.6 [a]
	ISeD (n=6)	26.5 ± 2.2 [a]	12.4 ± 0.6 [a]	23.8 ± 1.3 [c]	25.1 ± 2.6 [b]	23.7 ± 2.4 [a]	19.9 ± 1.8 [a]

[a,b,c,d] Values in columns not sharing a common superscript differ significantly, $p<0.05$. Values are given as mean ± SD.
* Mineral concentrations in plasma are given as micrograms per liter.

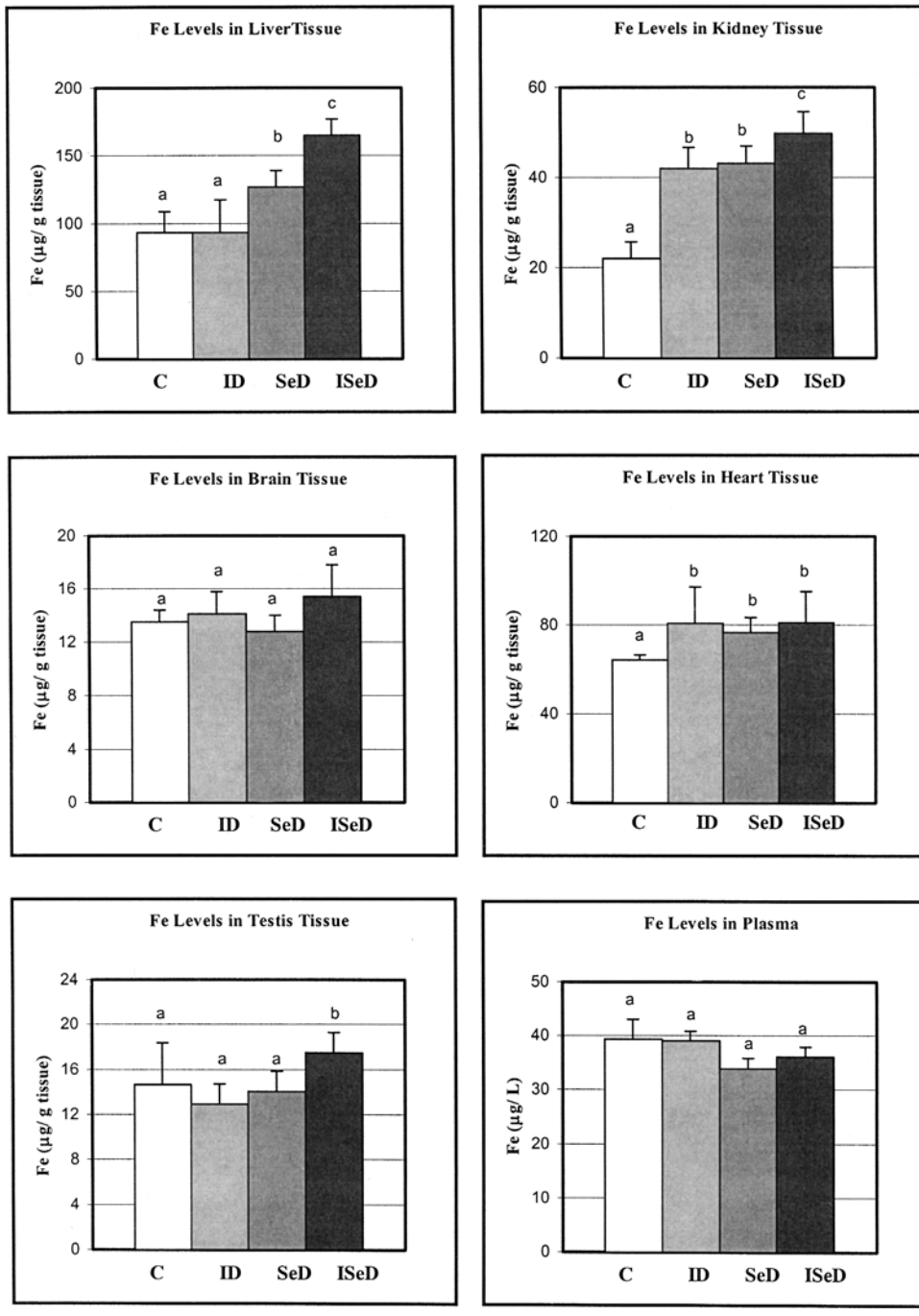

Fig. 1. Iron concentrations in various tissues of Se- and/or iodine-deficient rats. Superscripts of different letters differ significantly ($p<0.05$) from each other.

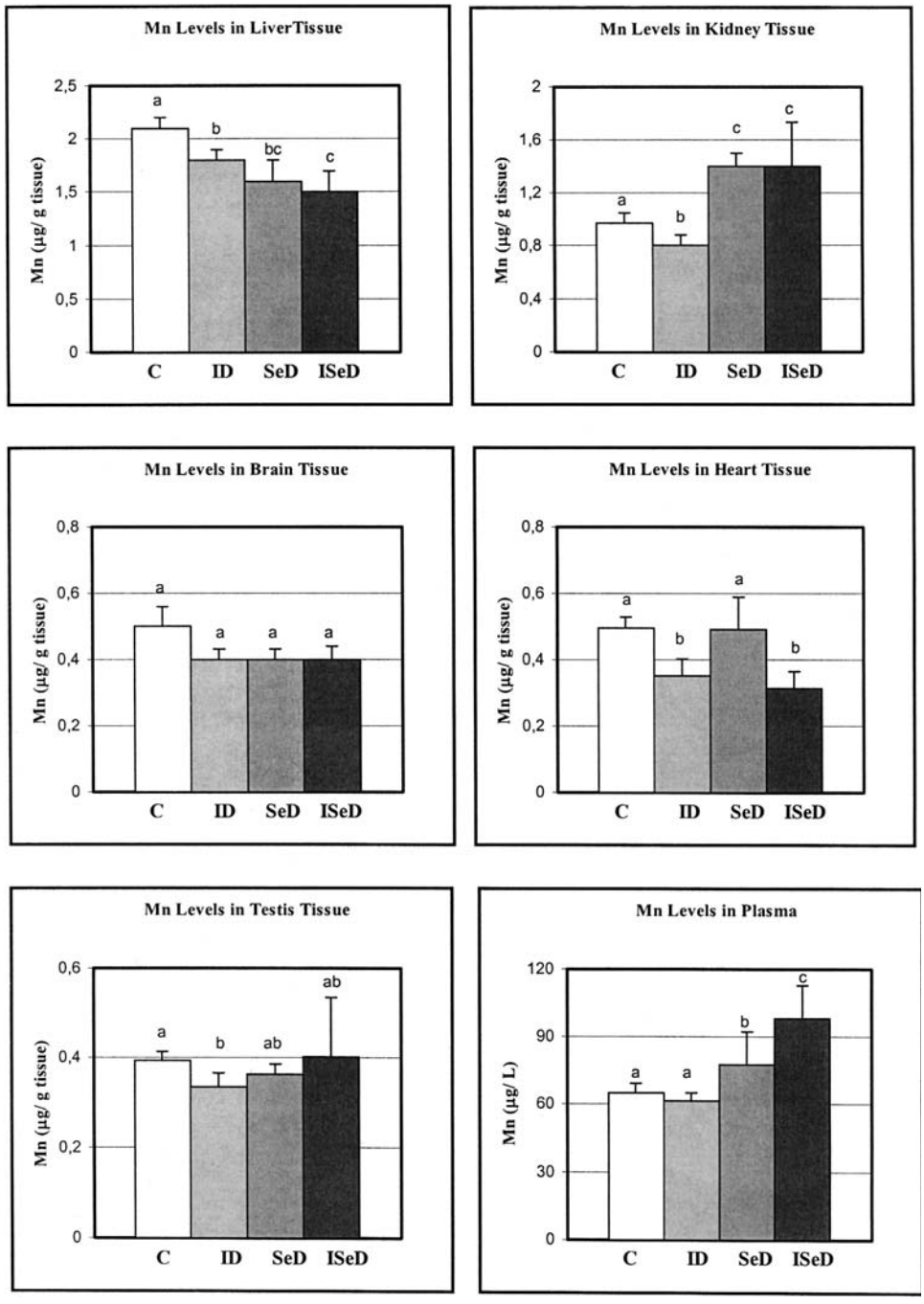

Fig. 2. Manganese concentrations in various tissues of Se- and/or iodine-deficient rats. Superscripts of different letters differ significantly ($p<0.05$) from each other.

Manganese concentrations decreased significantly in the liver in all deficiency states (approx 20–30%), in the heart in ID and SeD rats (approx 30–35%), and in the testis in ID rats (approx 15%). Whereas Mn enhancements were noted in the kidney (approx 45%) and plasma in SeD and ISeD (approx 20% and 50%, respectively) animals (*see* Fig 2).

There was no significant alteration in Cu and Zn levels in the liver and plasma (*see* Table 2). Lower Cu (approx 10–15%) and higher Zn (approx 10–20%) concentrations in the heart in all deficiency states; higher Zn (approx 20–35%) in the kidney in SeD and ISeD rats and lower Cu in the testis in SeD rats were observed. In brain tissue, no alteration was seen in Fe, Mn, and Zn content; however, significantly increased (approx 15–20%) Cu concentrations were noted in all deficiency states.

DISCUSSION

The results of the present study showed that in I_2- and/or Se-deficient rats, alterations in tissue distribution of essential trace elements occur; thus, I_2 and/or Se deficiency may modify the homeostasis of other minerals.

In our experimental conditions, I_2 deficiency produced typical effects of hypothyroidism, such as significantly lower plasma T_4 and T_3 concentrations and higher TSH levels. SeD rats also showed expected alterations of thyroid parameters: A significant increase in T_4 and a decrease in T_3 were observed. Combined deficiency caused highest alterations in TSH and T_4, but the decrease in T_3 was not different than for SeD rats, and these changes were in agreement with others findings *(19).* Imbalance of mineral distribution was seen both in I_2- and Se-deficiency states, but the extent of imbalance was more obvious in combined deficiency. Interactions of Se with other minerals in animals were observed by several researchers, but results were somewhat conflicting *(8,9,20,21).* There are a few studies on mineral metabolism in I_2 deficiency, but no studies exist dealing with mineral metabolism in combined I_2 and Se deficiency either in animals or in humans.

In the present study, the most striking alterations were observed in the tissue Fe distribution pattern. Except for the brain and plasma, the Fe concentrations increased in all tissues examined. Highest alterations observed in the liver and kidney, and all deficiency states caused enhancement of Fe in kidney and heart, whereas only combined deficiency caused an elevation in the testis. The data reported by Chaeronpous-Kawamoto et al. *(20)* show 1.1–2.5 times higher concentrations of Fe in the liver, kidney, and spleen tissues, and a 50% increase in Fe and in transferrin saturation in serum of SeD rats after a feeding period of 24 wk. The same researchers identified subcellular sites of Fe deposition in the liver and kidney and observed a dramatic increase in iron deposition in those tissues of rats on a Se-deficient diet for 8–82 wk *(21).* In full agreement with our results, Zhu et al. *(8)* observed no alteration in plasma Fe content, but they observed an approx 40% increase in the liver of SeD rats fed for 8 wk. However, in contrast to

our results, they did not observe any alteration in kidney Fe. These results along with ours indicate that an imbalance of Fe does occur in Se deficiency, but the distribution pattern can vary depending on the duration and severity of the deficiency. The pathological phenomena of Se deficiency are considered to be very similar to those caused by Fe overload *(22).* However, an inverse relation also exists, and Fe deficiency causes a decrease in GSH-Px activity *(5).* Fe constitutes the active center of cytochrome P-450 (P-450), and Matsumoto et al. *(9)* recently found a 100% increase in the activities of P-450s in liver microsomes of second-generation SeD rats. They concluded that the increase in Fe content of microsomal fraction was the result, at least in part, to the induction of P-450s. As discussed by the same researchers, Se deficiency may cause an iron overload in intracellular binding pools, and as a result of the redox stress, iron may have been released. Iron is vital for almost living organisms by participating in a wide variety of metabolic processes, including oxygen transport, DNA synthesis, and electron transport *(23).* However, body iron in excess offers no health benefit, but leads to tissue damage, as a result of formation of free radicals leading to the peroxidation of lipid membrane *(23).* In fact, as reported by the others *(24,25),* thiobarbituric acid reactive substances (TBARS) and other indices of lipid peroxidation increased in tissues, including liver, in our SeD animals (data will be reported elsewhere). Furthermore, in a previous study, we observed lower serum Se levels in children with Fe-deficiency anemia, but Se concentrations returned to normal levels after Fe supplementation *(26).*

Manganese was the second most affected mineral by all types of deficiency state in the present experimental conditions. The decrease we observed in liver Mn in SeD rats is in agreement with the results of Matsumoto et al. *(9),* which showed a decrease of Mn content in mitochondrial, microsomal, and cytosolic fractions of liver homogenates in SeD rats. Zhu et al. *(8)* observed significant alterations in Mn concentrations in SeD rats and reported that the decrease of Mn levels was related to the degree of Se deficiency. Alteration of thyroid hormone metabolism was also observed with Mn deficiency *(27).* In ID rats, Liu et al. *(28)* did not find any significant alteration in erythrocyte Mn, Cu, and Zn contents. However, I_2 supplementation decreased Mn and Zn concentrations, and I_2 plus Se supplementation reduced Mn levels significantly. Aihara et al. *(29)* reported abnormal Zn, Cu, Mn, and Se metabolism in adults with thyroid diseases and noted that the causes of these changes are far from explained. Mn functions as a constituent of metalloenzymes and as an enzyme activator. The liver is the primary organ involved in Mn homeostasis. It has several chemical and biochemical properties similar to iron, and there is evidence of metabolic interaction between the two minerals, particularly at the level of absorption from the intestine *(16,30–32).* Fe homeostasis may play an important role in the regulation of Mn transport, and competition between Mn and Fe for intestinal absorption is well established in human and rats *(31–33).* Our results may, thus, provide an explanation, at least in part, for the alteration of Mn distribution in rat tissues.

As a component of antioxidant metalloenzymes and metallothioneins, Zn has an important role in the defense system against oxidative stress *(34).* It protects SH groups from oxidation and in vitro might slow down oxidative damage to SH groups by competition with other transition *(34).* Zinc also has an important role in thyroid hormone metabolism. In addition to its participation in protein synthesis, it is involved in T3 binding to its nuclear receptor *(35).* Its roles are complex and may include effects on both the synthesis and mode of action of the hormones *(16).* Although there are some reports about the effects of Zn deficiency on thyroid hormone status, limited studies exist with regard to the alteration of Zn level in I_2 and/or Se deficiency *(36–38).* In the present study, Zn concentrations significantly increased only in kidney tissues of SeD and ISeD rats and in heart tissues of ID and ISeD animals.

Copper acts both as an antioxidant and a pro-oxidant in the body *(34).* Cu status has been linked to decreased plasma T_3 levels in animals and man *(39).* However, the only significant alteration of Cu found in the present study was decreased levels (approx 10–15%) in heart tissues and increased concentrations (approx 15–20%) in the brain. It is noteworthy that in brain tissue, no alterations were seen in Fe, Mn, and Zn content in any deficiency states, but significantly increased (approx 15–20%) Cu concentrations were encountered in all deficiency states.

CONCLUSION

The results of this study demonstrated that I_2 and/or Se deficiency in rats modifies the distribution pattern and homeostasis of other minerals, particularly Fe and Mn. We report that imbalance of mineral distribution and Fe overload occur not only in I_2- or Se-deficient animals, but also, more dramatically, in combined I_2 and Se deficiency.

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