Effects of Parathyroid Hormone on Plasma Zinc Concentration in Rat with Chronic Renal Failure

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Abstract

Although both secondary hyperparathyroidism (HPT) and hypozincemia are commonly observed in humans and animals with chronic renal failure (CRF), the relationship between secondary HPT and hypozincemia is little delineated. The present study was designed to examine whether the elevated plasma parathyroid hormones (PTH) levels do affect the disposition of extrarenal zinc and decrease plasma zinc level in CRF rats. The experiment was performed in normal and CRF rats with intact parathyroid gland and parathyroidectomized (PTX), using an acute zinc load alone or in combination with PTH infusion in five groups of rats: normal control, CRF control, CRF + PTH, CRF + PTX and CRF + PTX + PTH. Five sixths nephrectomy was used to produce CRF. All rats were infused with 0.05 mg/kg/min $ZnSO_4$ alone or in combination with 10 μg/kg/min PTH through intravenous infusion for 90 min with serial monitoring of plasma zinc levels every 30 min. The alteration of plasma interleukin-6 (IL-6) levels and the effect of zinc levels in red blood cells (RBCs), as well as the output of bile juice zinc and urinary zinc excretion during the 90-min infusion were also examined. After 90-min infusion, liver tissue was harvested to determine its contents of zinc and metallothionein (MT). During zinc sulfate infusion, the responses of plasma zinc concentration in PTH-combined infusion groups markedly decreased as compared with those of the non-PTH-combined infusion groups, especially in the CRF rats with PTX. However, when zinc sulfate alone was infused, the response of plasma zinc concentration was found to increase in CRF rats with PTX as compared with that of the CRF control rats. PTH infusion groups significantly increased the levels of plasma IL-6 ($P < 0.05$), but it did not alter the levels of RBC zinc and the secretion of bile zinc during the 90-min infusion. After 90-min zinc sulfate infusion, higher liver zinc and MT contents were found in CRF control, CRF + PTH and CRF + PTX + PTH rats, but was not found in the CRF + PTX rats. Zinc sulfate infused alone was found to increase the excretion of basal zinc in bile juice and urine, in both normal and CRF rats. The percentage of zinc load translocated out from the plasma during 90-min zinc sulfate infusion significantly rises in CRF rats and CRF rats with PTH-combined infusion as compared with normal control rats. However, in CRF rats with PTX, the percentage of zinc load translocated out from plasma during 90-min zinc sulfate infusion was similar to that in the normal control rats. Therefore, we suggested that in CRF rats, the excessive secretion of PTH may play a role in the pathogenesis of hypozincemia because PTH enhanced extrarenal zinc disposal.

Key Words: parathyroid hormone, hypozincemia, chronic renal failure, metallothionein, secondary hyperparathyroidism

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Received: September 9, 2004; Revised (Finally): November 18, 2004; Accepted: January 14, 2005.

Introduction

Zinc is an essential nutrient because it is a critical component of numerous metalloenzymes and zinc-dependent transcription factors (3). Zinc also appears to be a neuromodulator of certain postsynaptic neurons in the brain (9). Because of these important functions, nutritional zinc deficiency can have devastating consequences for human and animal health, including growth retardation, immune system dysfunction, and mental disorder. Some diseases have been suggested to induce zinc deficiency, such as chronic uremia (1, 29, 30), sickle cell disease (44), chronic alcoholism (55), Crohn's disease (31), and the genetic disorder acrodermatitis enteropathica (17). Hypozincemia is a main feature of zinc deficiency. Hypozincemia is commonly found in patients with renal insufficiency (10, 57) or uremic patients on hemodialysis (32, 33). Recent studies suggest that in patients with chronic uremia, both of the increased urinary zinc excretion and decreased intestinal zinc absorption (27, 34) may be the main cause of hypozincemia, because the kidney and intestines play an important roles in the maintenance of plasma zinc homeostasis (46). However, other studies have suggested that in patients with chronic uremia, hypozincemia might partially be related to a redistribution of total body zinc (10, 48), because it has been found to increase the content of zinc in autopsy liver tissue.

Acute zinc ($^{65}ZnCl_2$) load *via* venous injection has been found to rapidly increase accumulation in liver, kidney, skeletal muscle, and small intestine tissues in normal rat treated with parathyroid extract (11), especially in liver tissues, because the liver plays an important role in plasma zinc balance (39, 54). The mechanism of liver zinc uptake has been characterized as consisting of two distinct phases (54). The first is a fast-uptake phase, which represents the transport step into the hepatocyte, and the second is a slower exchange phase, which represents intracellular zinc binding or distribution (47). However, some hormones have been shown to stimulate the slow exchange phase and increase the uptake of liver zinc, such as glucocorticoids hormones (45).

A chronic serial subcutaneous injection of PTH has been found to reduce plasma zinc level and increase liver zinc accumulation in normal and uremic rats (24). Hypersecretion of PTH is a well-established complication of renal insufficiency in animals and humans (22, 35, 36). Hypersecretion of PTH in uremia may thus play a role in the pathogenesis of hypozincemia. Therefore, we used an acute zinc load in combination PTH intravenous infusion to examine the effects of PTH on extrarenal zinc disposition in rats with CRF.

Materials and Methods

Reagents and Animals

Rat PTH (fragment 1-34) and $ZnSO_4 \cdot 7\ H_2O$ were obtained from Sigma (St. Louis, MO, USA). Rat interleukin-6 immunoassay kits were obtained from BioSource International, Inc., Camarillo, CA, USA. Thyroxine (T_4) and triiodothyronine (T_3) enzyme immunoassay test kits were obtained from Maxim Biotech, Inc. (South San Francisco, CA, USA)

The experiments were performed in 40 adult male Sprague-Dawley rats weighing 300-350 g. These animals were obtained from the Animal Center of National Yang-Ming University and kept at 25°C in light (12 h)-dark (12 h) cycles. They were randomly divided into five groups of eight rats as follows: normal control, CRF control, CRF + PTH, CRF + PTX and CRF + PTX + PTH. All animals were fed with standard rat chow, which contained 23% protein, 1.05% calcium, 0.80% phosphate and with total zinc averaging 50-ppm. All animals had free access to food and water. Five-sixths of nephrectomy (5/6 Nx) was used to induce chronic uremia. Some uremic rats underwent thyroparathyroidectoctomy (TPTX) surgery 2 weeks after 5/6 Nx. The procedure for 5/6 Nx and TPTX is described in detail in our previous report (12). The glandular tissue was removed by means of blunt dissection with the animals under ether anesthesia. The plasma calcium level was measured after a 16-hour fast on the 6th day after TPTX. Only animals with a plasma calcium level below 6 mg/dl, which indicated successful parathyroidectomy (PTX), were used for the study. One week after TPTX, all TPTX rats were given subcutaneous injection of 0.8 μg/kg/day L-thyroxine (T_4) and 0.15 μg/kg/day triiodothyronine (T_3) in a combined treatment for 1 week. This treatment has been suggested to completely restore euthyroidism in TPTX rats (19).

Zinc sulfate infusion study was performed at two weeks after PTX or four weeks after 5/6 Nx. Prior to the zinc sulfate infusion study, 24 h urine samples were collected from all animals in a Nalgene plastic metabolic cage, and the animals were fasted for 16 h. The 24-hour urine samples were used to assay the contents of creatinine and urine protein.

On the day of infusion study, the animals were weighed and were anesthetized with sodium pentobarbital 40 mg/kg *via* intraperitoneal injection. A PE 50 catheter was placed in a femoral artery to obtain blood samples. A PE10 catheter was placed in a femoral vein for the zinc sulfate infusion. A midline abdominal incision was then made in order to collect the secretion of bile juice and the excretion of urine. A cannula was inserted into the common duct proximal to the hilum of the liver (between the points where the

ducts bifurcate and the area where the pancreatic tissue first appears) to collect pure bile. Another cannula was inserted into the bladder to collect urine. Before urine collection, the bladders were emptied. After surgery, the rats were placed on a heating plate to keep their body temperature constant. The baseline blood samples were used to determine plasma zinc, creatinine, urea nitrogen, inorganic phosphorus and calcium concentrations as well as T_4 and T_3 levels.

Zinc Sulfate and PTH Infusion

Zinc sulfate alone or zinc sulfate in combination with PTH infusion were performed at two weeks after TPTX or at four weeks after 5/6 Nx. Prior to zinc sulfate infusion, the basal zinc output was determined in bile and urine for 30 min in all rats. The zinc sulfate and Rat-PTH (fragment 1-34) were dissolved in a 0.9% NaCl isotonic solution and using a constant infusion pump (Harvard Apparatus, Millis, MA, USA) delivering either the zinc sulfate saline solution alone at 0.05 mg/kg/min, or the combined solution with 10 μg/kg/min PTH at a rate of 2 ml/h for 90 min. Heparinized blood samples were obtained 10, 30, 60 and 90 min, respectively after the venous infusion of zinc sulfate. These samples were immediately centrifuged at 4°C to obtain plasma and RBC. Plasma samples were used to determine the concentration of zinc. RBC was used to determine alterations in RBC zinc levels during the zinc sulfate infusion.

The percentage of zinc load translocated from plasma (X) was calculated by using the following equation.

$$X = \frac{\text{Zn injected} - (Zn_{90} - Zn_0)\ 0.2BW - UZnV}{\text{Zn injected}} \times 100,$$

where Zn_0 and Zn_{90} are plasma zinc concentration at baseline and at 90 min, respectively. BW is body weight and UZnV is the amount of zinc excretion, including urine and bile zinc excretion during the 90-min infusion. It is assumed in this calculation that the ECF space in the rat is 20% of its body weight (18, 49).

Both bile and urine were collected throughout the 90-min zinc sulfate infusion. At the end of experiment, the volume of bile secretion was determined by weight and the result was used to determine the total output of bile zinc within the 90-min zinc sulfate infusion. The bladder was perfused with 1.5 ml 0.3% phenol red zinc-free saline solution to remove the urine. The total volume of urine and phenol red was corrected for the 1.5 ml perfusate shifts as reflected by changes of the concentration of phenol red, which was used as an unabsorbable marker. The perfusate was then used to determine the urinary excretion of zinc. Blood was drawn from the abdominal aorta and the liver was washed with 20 ml of 0.9% heparinized NaCl solution *via* the abdominal aorta. After that, the liver was partially harvested and the samples were used to assays the zinc and MT contents.

Zinc, MT, IL-6 and Biochemical Analysis

The concentrations of plasma urea nitrogen, creatinine and calcium, and the level of urine phenol red were determined in an automatic biochemistry analyzer (Express Plus, Ciba-Corning Diagnostics Corp, MA, USA). The levels of plasma IL-6 were monitored at the beginning and the end of the PTH infusion by using a rat IL-6 immunoassay kit. The methods of atomic absorption spectrophotometer (AAS) were used to determine the concentrations of zinc and MT in all samples, including plasma and tissue samples, in a Perkin Elmer 3110 AAS. Plasma creatinine and urea nitrogen levels were used to monitor the development of uremia.

For the assays of RBC zinc, the RBCs were washed two times with isotonic saline solution and centrifuged at 4°C 5000 g for 10 min to yield packed RBC. The packed RBCs were resuspended in an equal volume of isotonic saline solution, and then the cyanomethemoglobin method was used to determine the hemoglobin (Hb) content. A total of 0.2 ml of the RBC suspension was digested in a 95°C dry heat bath with 0.2 ml 14 N HNO_3 for 20 min. It was then diluted to 2 ml with deionized distilled water to determine the RBC zinc content by means of AAS. RBC zinc concentrations were expressed in micrograms zinc per gram of hemoglobin.

For the determinations of liver Zn, MT and total protein levels: 0.3-0.4 g of fresh or frozen liver tissues was homogenized in 1 ml of a 0.25 M saccharose solution. Then, they were diluted with 1 ml of cold 0.25 M saccharose solution. The homogenate was centrifuged at 650 g for 10 min at 4°C to remove particles and then the resultant supernatant was used to measure the concentrations of tissue zinc, MT and total protein. The Lowry method was used to determine the tissue total protein on a Ciba-Corning autoanalyzer. A 0.2 ml sample of resultant supernatants was digested in a 95°C dry heat bath with 0.2 ml 14 N nitric acid for 20 min, and then dissolved in 2 ml deionized distilled water to determine the contents of zinc by AAS. Zinc concentrations in the liver were expressed as μg/g protein.

Liver MT levels were measured using the modified silver saturation hemolysate method (50). Briefly, 1 ml of the resultant supernatant was heated at 95°C in a dry heat bath for 5 min for deproteinization. Then 1 ml of a 0.25 M saccharose solution was added to dissolve the MT. Subsequently, the mixture was centrifuged at 12,400 g for 8 min to remove the

Table 1. Plasma creatinine, calcium, and zinc levels, well as the area under the plasma zinc response cure (AUC) during zinc sulfate (0.05 mg/kg/min) infusion period in CRF rats with various treatment

	Plasma Cr (mg/dl)	Plasma Ca (mg/dl)	Plasma Zn (μg/ml)		AUC (μg/ml-90 min)
			initial	90 min	
Normal control	0.51±0.03	9.85±0.09	1.33±0.03	8.73±0.17	437.1±10.5
CRF control	1.21±0.05*	9.78±0.12	1.19±0.03*	7.10±0.23*	335.6±12.1*
CRF + PTH	1.08±0.06*	10.6±0.10	1.23±0.02*	5.45±0.21$^{*\#}$	233.1±11.6$^{*\#}$
CRF + PTX	1.16±0.08*	5.24±0.17$^{\#}$	1.17±0.03*	8.17±0.30$^{\#}$	379.8±13.8$^{\#}$
CRF + PTX + PTH	1.20±0.07*	5.15±0.14$^{\#}$	1.10±0.04*	4.55±0.38$^{*\#\$}$	189.5±11.5$^{*\#\$}$

Each value indicates the mean ± SE of 8 rats.
$^{*}P < 0.01$ *vs.* normal control, $^{\#}P < 0.01$ *vs.* CRF control, $^{\$}P < 0.01$ *vs.* CRF + PTX

protein. Next, 0.5 ml of the resultant supernatant was incubated with 0.5 ml of a 0.314 mg/ml silver nitrate solution for 10 min to bind MT. Excessive silver was removed by adding 0.1 ml rat RBC hemolysate, obtained according to the procedure of Onosaka and Cherian (43), followed by heat treatment in a dry heat bath for 5 min and centrifuging at 12,400 g at 4°C for 10 min to remove the hemoglobin and excessive silver. The process was able to remove 99.5 % of excessive silver in these samples. The final supernatant was used to determine the silver concentration by means of AAS. The amount of silver in the final supernatant was proportional to the amount of MT present. The amount of MT in the sample was calculated with the following equation

$$\mu g\ MT/g\ protein = \frac{(C_{ag}-C_{bkg})X3.55XSDF}{P}$$

The terms are defined as follows: C_{ag} is the concentration of silver in the final supernatant; C_{bkg} is the background reading in the supernatant of the blank (without the tissue sample); SDF is the sample dilution factor and P is the concentration of protein in the final supernatant. For calculation, 1 μg of silver represented 3.55 μg of MT (50).

Statistical Analysis

All values were expressed as the mean ± SE. Student's unpaired *t* test and one-way analysis of variance (ANOVA) were used to evaluate the variance between the different treatments. The integration of the plasma zinc response from 0 up to 90 min (areas under the curve, AUC) was calculated by the trapezoidal rule. These AUC in different treatment rats were analyzed by one-way analysis of variance, and the difference of specific mean was analyzed for significance using the Duncan's multiple range test. A *P* value less than 0.05 was considered significantly different.

Results

Increased urinary zinc excretion and decreased plasma zinc levels were found in those uremic rats 4 weeks after 5/6 Nx surgery as compared with normal control rats (Table 1 and 2). Their levels of plasma creatinine and urea nitrogen were about two times those of the normal control rats (Table 1). Renal creatinine clearance was significantly lower than that in normal rats (data not shown). Thus, those 5/6 Nx rats were in markedly CRF status. because PTH plays an important role in the regulation of plasma calcium homeostasis, TPTX resulted in hypocalcemia in those CRF rats. Their plasma calcium level was below 6.0 mg/dl after TPTX (Table 1), but, their plasma T_4 and T_3 levels were still similar to those of non-TPTX rats (96.5 ± 3.0 *vs* 95.1 ± 4.1 ng/ml in T_4 and 1.92 ± 0.18 *vs* 2.23 ± 0.20 ng/ml in T_3, respectively), because they had received T_4 and T_3 intraperitoneal injections for 1 week after TPTX. For zinc sulfate (0.05 mg/kg/min) infused alone, significantly decreased area under the curve of plasma zinc response was found in CRF rats, as compared with normal control rats (Table 1, $P < 0.01$) but the CRF rats with PTX were found to have increased response of plasma zinc (Figure 1). The concomitant infusion of PTH along with zinc sulfate to CRF rats resulted in a mean low in plasma zinc (ΔPZn) which was lower than zinc sulfate infusion alone (Figure 1, $P < 0.05$), especially in CRF rats with PTX.

Zinc sulfate infusion was found to increase excretions of basal bile and urine zinc in normal and CRF rats (Table 2). PTH combination with zinc sulfate infusion increased liver zinc uptake (Table 3), but PTH combined infusion did not increase the excretion of bile zinc (Table 2). During PTH infusion, the concentrations of plasma IL-6 were found to elevate, including in the CRF rats and CRF rats with PTX (Table 3), but did not alter the contents of RBC zinc (Table 2).

Increased liver MT and zinc levels were found

Table 2. The excretion of zinc in bile juice and urine, and the alteration of RBC zinc during basal and zinc sulfate (0.05 mg/kg/min) infusion period in CRF rats with various treatment

	Bile Zn (μg/kg/h)		Urine Zn (μg/kg/h)		RBC Zn (μg/g Hb)	
	Basal	$ZnSO_4$	Basal	$ZnSO_4$	Basal	$ZnSO_4$
Normal control	4.55±0.35	5.85±0.41*	0.26±0.04	0.48±0.05*	24.8±1.5	25.2±2.2
CRF control	4.61±0.38	5.98±0.48*	0.42±0.03†	0.85±0.10*†	23.8±1.8	24.5±2.3
CRF + PTH	4.58±0.26	6.12±0.38*	0.48±0.04†	0.88±0.08*†	24.2±1.6	26.1±1.8
CRF + PTX	4.55±0.38	5.87±0.47*	0.44±0.03†	0.81±0.13*†	23.2±1.1	25.4±.2
CRF + PTX + PTH	4.88±0.37	6.21±0.44*	0.46±0.04†	0.78±0.10*†	24.5±1.2	23.8±1.8

Each value indicates the mean ± SE of 8 rats.
*Excretion significantly different from values obtained from basal excretion ($P < 0.05$).
†Significantly different from normal control ($P < 0.05$).

in CRF rats, especially in CRF rats with PTH infusion, but they were not found in CRF rats with PTX (Table 3). CRF rats showed significantly increased percentage of zinc load translocated from plasma at 90 min ($p < 0.01$), as compared with normal control rats (73.1 ± 1.5% *vs* 66.7 ± 0.8 %), but it was not found in CRF rats with PTX (67.2 ± 1.8% *vs*. 66.7 ± 0.8%). The PTH combination with zinc sulfate infusion increased more zinc translocation from plasma in both CRF and CRF + PTX rats than zinc sulfate infusion alone (Figure 2).

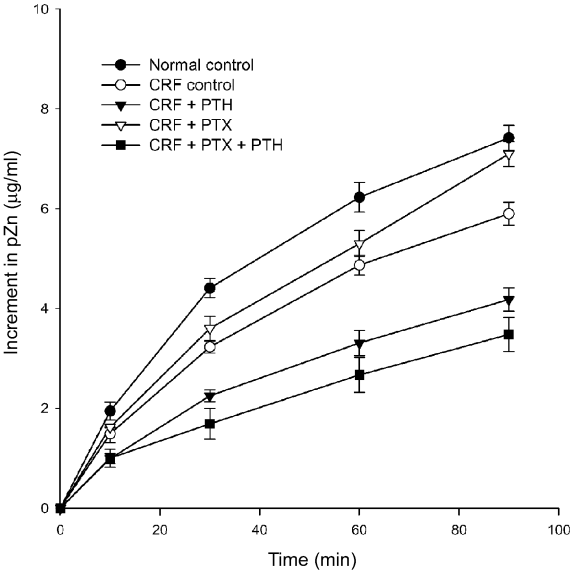

Fig. 1. Time course of the change in plasma zinc concentration (ΔPZn) resulting from the infusion of zinc sulfate (0.05 mg/kg/min for 90 min) in normal rats (●-●), in CRF rats (○-○), in CRF rats receiving combination PTH infusion (10 μg/kg/min, ▼-▼), in CRF rats with PTX (▽-▽) and in CRF with PTX receiving combination PTH infusion (10 μg/kg/min, ■-■). All values represent the mean in eight rats; the vertical lines denote 1 SE. ΔPZn in CRF control and PTH-infused groups were significantly lower than those of the normal control ($P < 0.05$).

Discussion

Abnormalities of zinc metabolism have been reported in humans and animals with CRF (37, 38, 40). The present study found that CRF rats with intact parathyroid gland presented a lower response in the plasma zinc level and a higher percentage of zinc translocated from the plasma than normal rats during the zinc sulfate intravenous infusion, especially during zinc sulfate combination PTH infusion. These observations were consistent with the notion that PTH enhances the disposal of extrarenal zinc, and that secondary hyperparathyroidism (HPT) may occur in these CRF rats, because CRF rats with PTX markedly reduce the percentage of zinc translocated from the plasma, as compared with CRF control rats. It has been reported that the secondary HPT may occur in rats within hours after the induction of renal failure (26). Thus, in our CRF rats, the hypozincemia might have partially resulted from the secondary HPT.

Further studies were then conducted to assess whether the PTH enhanced the uptake of liver zinc and then reduced the response of plasma zinc levels during the zinc sulfate intravenous infusion, because the liver appeared to be a major site of zinc uptake (11, 39). In the present study, we suggested that PTH might enhance uptake of liver zinc because PTH infusion groups had a higher contents of liver zinc, as compared with the non-PTH infusion groups.

The uptake of liver zinc has been shown to be a biphasic process (45, 47, 54). The fast uptake phase of zinc reflects net zinc accumulation into a labile pool. This process may be completed in ~2 min (41). The labile zinc is free or only loosely bound to protein or bound to low-molecular weight protein, because it can easily exchange between different compartments of the cell, bind to numerous proteins (25), and act as

Table 3. The contents of zinc and metallothionein (MT) in liver after 90-minute zinc sulfate (0.05 mg/kg/min) infusion, and the alternation of plasma interlukin-6 (IL-6) during CRF rats with various treatment

	Liver MT (μg/g protein)	Liver Zn (μg/g protein)	Plasma IL-6 (pg/ml)	
			Initial	90 min
Normal control	1.87±0.08	189.4±10.9	8.5±0.8	8.9±0.6
CRF control	2.24±0.10*	228.2±12.1*	10.9±1.2	15.2±4.1
CRF + PTH	2.68±0.18$^{\#}$	265.1±11.5$^{\#}$	11.2±0.9	56.2±6.8$^{\#}$
CRF + PTX	2.13±0.15	212.2±12.9	14.2±1.8	10.2±2.5
CRF + PTX + PTH	2.78±0.20$^{\$}$	269.5±15.1$^{\$}$	13.3±1.5	48.6±7.2$^{\$}$

Each value indicates the mean ± SE of 8 rats. MT and zinc contents in liver are expressed as μg/g protein.
$^{*}P < 0.05$ *vs.* normal control, $^{\#}P < 0.05$ *vs.* CRF control, $^{\$}P < 0.05$ *vs.* CRF + PTX

a source of zinc for the slow-exchange phase (45). Based on results, we suggested that PTH might enhance the uptake of liver zinc in the fast uptake phase, because $ZnSO_4$ combination PTH infusion increased the contents of liver zinc within a short time during the 90 min zinc sulfate infusion. The mechanism of PTH stimulated uptake of liver zinc is unclear, but PTH has a calcium ionophoric property (4, 5) and it enhances entry of calcium ion into hepatic cell (13). It has been shown that zinc is able to enter postsynaptic neurons through a Ca-A/K channel (51). Thus, PTH may also enhance uptake of liver zinc through calcium channel, because zinc ion is the transported species in the uptake of liver zinc (54). However, further *in vitro* studies are required to elucidate the actual mechanism of PTH stimulated liver zinc uptake. Hence, intracellular MT is a major zinc-binding protein (3, 6), since the absorbed zinc may be bound by MT. MT-bound zinc reduces the degradation of MT (7, 14) and thus increases intracellular MT concentration, so liver MT levels are found to rise in rats with PTH infusion.

Increased liver MT level may enhance the transfer of zinc from plasma to the liver (14, 15). Many hormones, such as glucocorticoids, epinephrine, and glucagons, as well as IL-1, have been shown to increase liver zinc uptake through a MT-linked mechanism (20, 28, 45) to enlarge the labile pool size and increase the uptake of liver zinc. In the present study, we found that the concentrations of liver MT increased in PTH infusion rats. Thus, we suggested that PTH may simultaneously be through MT-linked mechanism to increase liver zinc uptake. Our present results were consistent with other authors' report (23) which suggests that PTH infusion increases the concentration of plasma IL-6, while IL-6 is a major cytokine mediator of MT gene expression in hepatocytes (52). Thus, PTH may partially be associated with the effect of IL-6 to increase the uptake of liver zinc through MT-linked mechanism.

Based on the present results, we also suggested

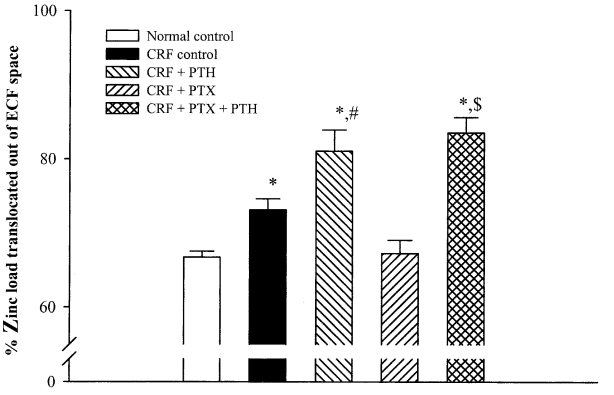

Fig. 2. The percentage of zinc load translocated from plasma during 90-min 0.05 mg/kg/min zinc sulfate infusion. All values represent the mean in eight rats; the vertical lines denote 1 SE. Statistical analysis was conducted using the standard unpaired *t* test after ANOVA. $^{*}P < 0.05$ *vs.* normal control, $^{\#}P < 0.05$ *vs.* CRF control, $^{\$}P < 0.05$ *vs.* CRF+PTX.

that PTH enhanced the uptake of liver zinc, but it did not increase the secretion of bile zinc, which might have been resulted from PTH while enhances liver zinc uptake and simultaneously stimulates MT synthesis to increase accumulation of liver zinc. Thus, chronic subcutaneous PTH administration has been found to increase the concentration of liver zinc in normal and uremic rats (11, 24).

Intracellular zinc concentration is strictly regulated, and intracellular zinc homeostasis is maintained through various mechanisms, including zinc sensing, binding, and sequestrating. Zinc sensing is mediated by regulating the expression of zinc transport proteins, including those processes for zinc influx during zinc deficiency and efflux during zinc excess (42). Consequently, in the present study, we observed that during zinc sulfate intravenous infusion, the secretion of basal bile zinc increased, which might have been resulted from the increase of liver zinc

uptake to enhance zinc efflux from bile.

Since parenteral zinc administration may increase the concentrations of plasma ultrafilterable zinc (56), increased plasma ultrafilterable zinc may increase the amount of glomerular filterable zinc and then increase the source of zinc destined for urinary excretion (46). Our results showed that the excretion of urinary zinc increased during zinc sulfate intravenous infusion in all rats.

Zinc in the blood is mostly transported in plasma proteins and red blood cells. However, the total zinc content of the red blood cells is ~10 times more than that of the plasma. Hence, the uptake of RBC zinc may influence the metabolism of body zinc. A Ca^{2+}/ Zn^{2+} exchanger that has been found in red blood cells may drive RBC zinc efflux (53), and yet our results indicated that the property of PTH calcium iontophoresis to enhance entry of calcium into many cells (13, 21, 51) did not to enhance the efflux of red blood cells zinc because PTH infusion did not reduce the content of red blood cell zinc.

Hypozincemia is commonly found in patients with CRF and has been shown to be related to their renal insufficiency, because the reabsorption of zinc in the renal tubules plays a vital role in zinc balance (46). Increased urinary zinc excretion has been found in patients or animals with renal insufficiency (27, 34). However, many other factors may also contribute to the hypozincemia in patients with CRF, e.g., decreased protein intake (8), decreased absorption of intestinal zinc (2, 16), and/or altered distribution of zinc in the body (10, 48). We hence concluded that in CRF, the secondary HPT mighty play a role in altering body zinc distribution because acute PTH venous infusion increased shift of zinc from the extracellular compartment to the intracellular compartment, which might have been resulted from PTH's enhancement of the uptake of tissues zinc, such as liver, kidney and small intestine tissues.

Acknowledgments

This work was supported by a grant no. 92-117 from the Taipei Veterans General Hospital, Taiwan, Republic of China. We appreciate Ching-Chu Wang's and Fanny Lin's technical assistance in this study.

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