	d_{obs}(Å)	d_{calc}(Å)	h	k	l	I/I_o
10	4.152	4.150	1	3	1	2
		4.178	$\bar{2}$	2	3	
11	4.040	4.033	$\bar{2}$	1	4	8
12	3.917	3.902	$\bar{3}$	1	1	10
13	3.754	3.765	2	1	2	
		3.774	$\bar{1}$	2	4	10
		3.772	$\bar{2}$	3	2	
		3.760	$\bar{1}$	3	3	
14	3.693	3.696	2	3	0	12
		3.672	0	4	0	
15	3.641	3.634	1	2	3	
		3.632	3	1	0	34
		3.645	$\bar{2}$	2	4	
16	3.527	3.528	$\bar{2}$	3	3	
		3.544	$\bar{3}$	2	1	11
		3.514	0	2	4	
17	3.427	3.439	2	2	2	2
18	3.345	3.339	3	2	0	
		3.352	1	0	4	5
		3.338	0	4	2	
19	3.271	3.272	$\bar{1}$	3	4	6
20	3.198	3.187	$\bar{2}$	3	4	8
		3.182	1	3	3	
21	3.048	3.048	2	3	2	
		3.048	3	2	1	2
		3.049	1	2	4	
		3.057	1	4	2	
22	2.979	2.976	3	3	0	
		2.971	2	2	3	3
		2.976	$\bar{2}$	4	3	
23	2.912	2.919	$\bar{3}$	2	5	
		2.900	$\bar{4}$	1	4	3
		2.899	2	4	1	
		2.894	3	1	2	
24	2.862	2.856	1	5	1	3
		2.847	$\bar{4}$	2	2	
25	2.722	2.722	$\bar{3}$	4	1	
		2.711	2	1	4	2
		2.718	4	1	5	
26	2.571	2.574	0	5	3	
		2.567	$\bar{3}$	4	4	3
		2.582	2	2	4	
27	2.249	2.247	$\bar{1}$	5	5	
		2.252	$\bar{5}$	2	5	5
		2.245	$\bar{2}$	5	5	
		2.247	4	2	2	

ACKNOWLEDGEMENT

The authors express their thanks to Dr. K. N. Rao, Head, Chemical Group for his encouragement in this work.

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CHANGES IN THE TISSUE CONCENTRATION OF GLUTATHIONE AND PROTEIN IN LEAD TOXICITY

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ABSTRACT

The effect of different levels of lead on the reduced glutathione (GSH) and protein concentration of the liver, kidneys and brain was investigated in this study. Lead toxicity increased the GSH concentration of the liver and kidneys but the brain GSH concentration was relatively unaffected. The protein concentration of the organs decreased in lead toxicity. A possible role for GSH in detoxication of lead is indicated.

INTRODUCTION

LEAD poisoning is recogized as a public health hazard, as contamination occurs easily from several sources[1] and only small increases over the level found in average dietaries are needed to produce toxic symptoms[2]. One of the well-documented effects of lead poisoning is a reduced synthesis of haem due to decreased activity of δ-amino levulinic acid dehydrase (ALAD)[3].

The ALAD catalyzes the condensation of two molecules of δ-amino levulinic acid to form porphobilinogen and requires GSH for its activity[4]. However, it is not known whether lead has a direct effect on ALAD, or whether it is mediated through GSH, the cofactor needed for its activity. The possibility that lead poisoning may alter the glutathione concentration in different tissues has been investigated in this study. Further, as generous quantities of good quality protein have been shown to offer some protection in lead toxicity[5,6] the study included investigations at two levels of protein, high and low. The protein content of the tissues was also analyzed.

EXPERIMENTAL

Weanling rats of Charles Foster strain of both sexes weighing on an average 43 g were randomly divided into eight different groups of six animals per group. Four groups were fed on a diet high in protein (HP, 20% Casein) and four others were fed a diet low in protein (LP, 6% Casein). The diets were adequate in all other respects[7,2]. At each level of protein, one group designated as the control did not receive any lead. The other three (Experimental I, II, and III) were given respectively 0.25, 0.50 and 1.00 per cent lead-carbonate (Robert Johnson Co., Bombay) in the diet. All animals were allowed an *ad libitum* access to food and distilled water. The animals were fed the respective diets for a period of 28 days. On the 29th day the animals were killed by decapitation; the liver, kidneys and brain removed, weighed and analyzed immediately for GSH[8] and protein[9].

RESULTS AND DISCUSSION

The GSH concentration was increased in the liver and kidneys of the rats receiving lead, whereas there was no significant change in the brain GSH concentration (table 1). While at the 0.25% level of lead carbonate, the increase in liver GSH concentration over the controls for the HP and LP animals was only 3 and 19 per cent, at the 0.5 and 1% lead-carbonate levels, the increase amounted to 30-33% for the HP animals and 60-75% for the LP animals. In other words, the increase in GSH concentration was more marked at higher levels of lead and was larger (as compared to the respective controls) for the low protein lead fed animals than the high protein lead fed ones. The GSH concentration in the kidneys of all the lead fed groups was significantly higher than that of the controls not receiving any lead.

The protein concentration in the liver, kidneys and brain, unlike the GSH concentration, was reduced in lead toxicity (table 2). This effect was clearly seen in the groups receiving lead at 20% protein whereas in the groups receiving the low protein diet, and particularly at higher levels of lead, the trend was not consistent.

The data reported here indicate that GSH concentration of liver and kidneys is increased significantly in lead toxicity. Thus the reduced activity of ALAD may not be due to the reduced amount of cofactor available (GSH). However, it appears that the increase seen in the liver and kidney GSH concertration may relate to

TABLE 1

Effect of different levels of lead on the GSH concentration of liver, kidneys and brain of rats fed a low (LP) and high protein (HP) diet

Lead carbonate %	GSH mg/ 100 g fresh weight.					
	Liver		Kidneys		Brain	
	HP	LP	HP	LP	HP	LP
0.00 (control)	202.9 ± 16.6	92.0 ± 7.6[b]	110.9 ± 14.3	100.3 ± 10.2	89.1 ± 7.4	76.3 ± 5.0
0.025 (Exptl I)	209.0 ± 21.0	109.5 ± 16.6[b]	204.2 ± 18.6[a]	190.9 ± 26.2[a]	80.1 ± 3.3	77.6 ± 7.7
0.50 (Exptl II)	264.1 ± 29.8	155.6 ± 10.4[ab]	204.8 ± 22.5[a]	177.8 ± 27.2[a]	76.3 ± 5.9	83.0 ± 5.2
1.00 (Exptl III)	270.0 ± 26.6	161.0*	194.9 ± 22.0[a]	149.0*	89.5 ± 3.4	76.0*

Values are mean ± SE based on six animals in each group except in LP Exptl III

[a] Significantly different from controls receiving no lead carbonate, $p < 0.05$.

[b] Significantly different from the groups receiving high protein, $p < 0.05$.

* Only two animals survived.

TABLE 2

Effect of different levels of lead on the protein concentration of liver, kidneys and brain of rats fed a low (LP) and a high protein (HP) diet

Lead carbonate %	Protein g/ 100 g fresh weight					
	Liver		Kidneys		Brain	
	HP	LP	HP	LP	HP	LP
0.00 (control)	13.5 ± 0.7	11.2 ± 0.8	11.6 ± 0.9	10.2 ± 0.9	10.1 ± 0.8	8.4 ± 0.5
0.25 (Exptl I)	12.5 ± 0.5	10.5 ± 1.0	10.3 ± 0.7	10.5 ± 0.2	9.7 ± 0.5	8.5 ± 0.5
0.50 (Exptl II)	10.4 ± 0.5*	10.8 ± 0.7	10.3 ± 0.4	11.5 ± 1.4	9.5 ± 0.4	9.7 ± 0.5
1.00 (Exptl III)	10.5 ± 0.5*	—	9.8 ± 0.6	—	9.1 ± 0.4	9.1*

Values are mean SE, based on six animals in each group except LP Exptl III.

* Significantly different from control, $p < 0.05$.

* Only two animals survived.

the role of GSH in conjugating lead. While a GSH conjugate of bromosulphathalein has been reported[10], no studies seem to have been carried out on the GSH conjugates of lead. It has also been reported that liver slices of rats convert naphthalene into a glutathione derivative-S (1:2-dihydro-2-hydroxy-1-naphthyl) glutathione[11]. Thus it appears that the increase seen in GSH concentration in lead toxicity may be a protective mechanism. The fact that despite the low level of protein, the LP lead fed animals showed a substantial increase in GSH over the control, argues in favour of a role for GSH in detoxication of lead and a preferential use of amino acids, to maintain the reduced glutathione concentration high.

An alternative explanation is also possible. It has been reported elsewhere that liver and brain ascorbic acid is reduced significantly in lead toxicity[6,7]. GSH is necessary for the reduction of dehydroascorbic acid to ascorbic acid. A reduction in ascorbic acid concentration and perhaps other such metabolities, which require the mediation of GSH, may affect the steady state level of GSH, resulting in higher concentration in the tissues. Further, the reduced oxygen tension due to anaemia, which is reported to occur in lead toxicity[1], may reduce the oxidation of GSH and result in higher levels of GSH in the tissues. However, it is difficult to reconcile these postulates with the increased incorporation of cysteine ^{35}S into liver and kidney GSH, reported by Hsu[12] in rats with lead toxicity. This observation of Hsu implies that lead has a stimulatory effect on the biosynthesis of glutathione. It was reported in the same study that an increase occurs in the liver, kidney and brain GSH concentration in lead toxicity, the increase in liver and kidneys being much larger than the increase in brain, results which are essentially similar to ours. Investigations to clarify the possible role of GSH in conjugating lead are needed. The decreases seen in the protein concentration of the organs may imply a reduced synthesis of protein in these organs. Some support for this is derived from the observations of Hsu[12] who has reported a decreased ^{35}S cysteine incorporation into liver and kidney protein of rats fed lead. Lead toxicity may thus alter the balance between protein and GSH synthesis in some tissues.

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