Manganese Balance Studies in Infants after Operations on the Heart

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Summary

Manganese balance studies have been performed on 16 infants, aged 3 days to 8 months, in the period following operation for the correction of congenital heart defects. Samples were analyzed by flameless atomic absorption spectroscopy or by solvent extraction

with 8-hydroxyquinoline followed by flame atomic absorption spectroscopy. A higher manganese content was found in either whole blood (710 ± 320 nmole·litre⁻¹) or purified plasma protein (1130 ± 770 nmole·litre⁻¹) compared with fresh frozen plasma (215 ± 35 nmole·litre⁻¹) used in intravenous drips. The manganese content of the milks used in oral feeding was 200-300 nmole·litre⁻¹.

Table 1. *Clinical features of patients*

Patient No.	Age ³ (days)	Length of balance (days)	Weight (kg)	Diagnosis	Blood/plasma intake	Milk intake	
1	23	10	3.3	Pulmonary stenosis	blood day 1; plasma days 2 and 3	(from day 2) SMA	died age 6 months
2	3	9	4.2	TAPVD ⁴	plasma day 1	(from day 3) SMA	died 1 month after admission
3	2	10	3.3	TAPVD ⁴	plasma to day 4	(from day 2) SMA and S26	
4	43	10	3.3	TAPVD ⁴	plasma day 1	(from day 2) S26	
5	36	10	3.8	Congenital aortic stenosis; mitral regurgitation	blood day 1; plasma to day 4	(from day 2) SMA	
6	116	11	4.6	TGA ⁵	blood and plasma day 1	(from day 2) S26	
7	73	11	4.4	TAPVD ⁴	blood day 1; plasma day 1 and 2	(from day 2) S26	
8	218	10	6.7	TGA ⁵	plasma day 1	S26	died 1 year later
9	153	8	5.6	TGA ⁵	plasma to day 3	(from day 2) ¹ SMA % S26	
10	16	11	3.5	TAPVD ⁴	plasma day 1	(from day 2) S26	
11	248	10	5.9	TGA ⁵	plasma day 1	(from day 1) ¹ S26 and SMA	
12	1	10	3.0	TAPVD ⁴	throughout period studied	(from day 2) S26	(died 6/52)
13	238	10	6.9	TGA ⁵	plasma days 1 and 2 ²	(from day 2) ¹ SMA and S26	
14	97	9	4.4	VSD ⁶ , heart failure	plasma day 1	(from day 2) S26	
15	13	9	4.5	TAPVD ⁴	blood day 1; plasma to day 3	(from day 2) S26 and SMA	(died 21 months after surgery)
16	84	10	3.5	TGA ⁵	blood day 1	(from day 2) S26 and SMA	

¹ Received solid food in addition.

² Records of blood and plasma intake incomplete.

³ At start of balance.

⁴ TAPVD, total anomalous pulmonary venous drainage.

⁵ TGA, transposition of the great arteries.

⁶ VSD, ventricular septal defect.

Total manganese intake was under $200 \text{ nmole} \cdot \text{kg}^{-1} \cdot 24 \text{ h}^{-1}$ in infants on milk feeding, but infants receiving solid foods received up to $1500 \text{ nmole} \cdot \text{kg}^{-1} \cdot 24 \text{ h}^{-1}$. There was a significant correlation between net intake and net balance ($P < 0.01$) and between intake and faecal loss ($P < 0.001$). Urinary manganese loss is not significant.

Manganese is an essential trace element for both animals and man (15) as a constituent of a number of vital biochemical pathways. Despite numerous observations of deficiency in animals, manganese deficiency in man has been reported only once in association with vitamin K deficiency (6). The symptoms included impaired growth of hair and nails, weight loss, and a reduced response of blood clotting proteins to vitamin K.

The manganese content of milk is about 0.02 ppm (9, 16) (350 nmole/liter) and the daily intake of manganese by infants is about 180 nmole . When cereal products are added to milk the manganese intake may be much higher (15).

Casey and Robinson (4) found that the manganese content of human foetal tissue might be up to 9 ppm ($160 \text{ } \mu\text{mole/kg}$) dry weight in the liver, but was much lower in other tissues. This is contrary to the earlier findings of Widdowson *et al.* (18) and Seeling *et al.* (12) that whereas copper and zinc are accumulated in foetal liver, manganese is not. The nutritional requirement for manganese in the neonate and infant is uncertain, but it has been suggested that manganese is lost in the bile regardless of the intake (3). Alexander *et al.* (1) have recommended an intake of about $1.0 \text{ } \mu\text{mole/kg/day}$ in children although, as indicated above, the intake required by neonates may be rather less. In rats the ability to maintain homeostasis develops only 17–18 days after birth (10), and manganese injected intraperitoneally is retained mainly in the liver.

Table 2. Analytical recovery of manganese (values expressed as mean $\pm$ S.D.)

Amount of Mn added	Amount of Mn found	Recovery
by electrothermal atomisation		
0 ¹	12.3 ± 0.8^1	
25	13.6 ± 2.3	97%
50	59.3 ± 3.9	94%
by solvent extraction		
0 ²	1.4 ± 0.2^2	
4	5.0 ± 0.4	93%
8	9.2 ± 0.6	98%

¹ $\text{nmole} \cdot \text{liter}^{-1}$.

² $\mu\text{mole} \cdot \text{liter}^{-1}$.

Table 3. Precision of manganese assay

	Concentration	No. of determinations	Coefficient of variation $\pm$ S.D.
Within batch precision ¹			
Electrothermal atomisation	30 ²	29	$9.4 \pm 3.9\%$
	500 ²	29	$9.7 \pm 4.6\%$
Solvent extraction	1 ³	25	$9.1 \pm 5.3\%$
	10 ³	25	$5.5 \pm 3.2\%$
	Concentration ⁵	No. of determinations	Coefficient of variation
Between batch precision ⁴			
Electrothermal atomisation	33.3 ± 3.2	17	9.6%
	536 ± 62	17	11.6%

¹ Mean coefficient of variation over several determinations.

² $\text{nmole} \cdot \text{liter}^{-1}$.

³ $\mu\text{mole} \cdot \text{liter}^{-1}$.

⁴ Repeated assay of samples over several determinations.

⁵ Mean $\pm$ S.D.; $\text{nmole} \cdot \text{liter}^{-1}$.

Table 4. Manganese content of milk and infused fluids

	$\text{nmole} \cdot \text{liter}^{-14}$	Range
Milks		
S26 ¹	270 ± 140 ($n = 8$)	105–1150
SMA ²	215 ± 140 ($n = 5$)	70–420
EBM ³	110 ($n = 1$)	
Dextrose 5%	26 ± 28 ($n = 17$)	<5–200
Saline	40 ± 51 ($n = 6$)	5–145
Whole blood	710 ± 320 ($n = 8$)	265–1110
Fresh frozen plasma	215 ± 35 ($n = 3$)	195–270
Purified plasma protein	1130 ± 770 ($n = 18$)	295–2920

¹ Manufacturer, John Wyeth Ltd.

² Manufacturer, Cow and Gate Ltd.

³ Expressed breast milk.

⁴ Mean $\pm$ S.D.

Table 5. Total intake and output of manganese over the balance period of 16 patients after cardiac operation

Patient No.	Intake nmole/kg	Output			Balance nmole/kg
		Urine nmole/kg	Stool nmole/kg	Other ¹ nmole/kg	
1	179	44.0	473	0	–338
2	147	49.0	644	0	–497
3	214	53.2	428	0	–267
4	989	38.4	1165	0	–214
5	222	36.3	178	0	8
6	233	69.1	1045	0	–880
7	227	28.5	465	0	–267
8	221	60.5	986	38.5	–874
9	1000	31.1	551	0	418
10	208	41.0	1015	0	–848
11	4413	83.8	3712	163	454
12	318	145	1266	57	–1150
13	3095	118	1253	5	1719
14	340	24.5	568	0	–253
15	264	14.7	360	0	–111
16	300	35.9	1094	0	–830

¹ Vomit, aspirate etc.

We have performed manganese balance studies on a group of infants recovering from surgery to correct congenital defects of the heart to assess the potential manganese requirement in the very young.

MATERIALS AND METHODS

All sixteen patients who were studied had been admitted to the Hospital for Sick Children for surgical correction of congenital defects of the heart or great vessels (Table 1). They were all boys aged from 2–248 days. Patients were studied with the informed consent of their parents or next of kin.

Balance collections were begun within 6 h after operation by the method described by Stevens *et al.* (14), and continued for up to 11 days. The manganese content of the containers used to store the samples before analysis was negligible (less than $0.2 \text{ } \mu\text{g}$). All samples were deep frozen until analysis.

Urine samples were analyzed directly by atomic absorption spectroscopy using an electrothermal graphite tube atomiser. Manganese content of other samples was determined by extracting ashed samples with 8-hydroxyquinoline in 4-methylpentan-2-one followed by flame absorption spectroscopy (5).

RESULTS

The recovery of manganese added to a normal urine sample as determined by electrothermal atomisation, and added to a pooled plasma sample as determined by solvent extraction and flame atomisation is shown in Table 2. The within batch precision of

the assays, as the mean within batch precision over several analytical runs is shown in Table 3. Between batch precision data for the electrothermal atomisation technique are also shown in Table 3.

The manganese contents of the milks and other fluids administered to the patients are shown in Table 4. The drugs used were also analysed and found to contribute a negligible proportion of the daily intakes (less than 0.5%). The manganese content of the milks was low whereas the manganese content of both the whole

blood and purified plasma protein (used in transfusions) was high. Fresh frozen plasma had a relatively low manganese content, similar to that of the milks. The high manganese content of the blood is due to the manganese content of the erythrocytes, whereas that of the purified plasma protein may have been acquired during processing.

The balances on the sixteen patients are summarised in Table 5. The high intakes of patients 9, 11, and 13 were due to being fed solid foods with a high manganese content during the latter stages

Fig. 1

Fig. 2

of the balance period (up to 18 μ mole/day). Patient 12 received blood and plasma infusions throughout the balance period. The blood and plasma infusions during the first few days of the balance gave a much higher manganese intake in the earlier period of the study when compared with the intake of children being fed orally. The final milk intake reached 300–600 ml per day, giving a manganese intake of 30–300 nmole per day.

There was a significant correlation between net intake and net balance ($P < 0.01$) and between the actual daily intake and daily balance ($P < 0.001$) (Fig. 1). There was no correlation between intake and urinary loss of manganese but there was a high correlation between intake and faecal loss ($P < 0.001$). There was also a correlation between the age of the patient and the intake of manganese, presumably due to the high manganese content of the food eaten by the older children. This gives an age-related correlation for the manganese balance (Fig. 2).

DISCUSSION

The urinary manganese loss was small compared to that in the faeces (<10%). A significant correlation was found to exist between urinary chromium excretion and chromium intake (Sampson, Barlow and Wilkinson, unpublished work) in the same group of patients as studied here. The absence of such a correlation for manganese may be an indirect confirmation of the low importance of the kidneys as an excretory route for this trace element. The negative balances observed in these patients may be indicative of a degree of manganese deficiency and support the observations of Hughes and Cotzias (7) of possible manganese deficiency in neonatal mice. Widdowson records negative manganese balances in neonates of approximately 160 nmole/kg/day (19).

Most manganese is excreted in the bile (8, 11), but biliary obstruction does not cause increased urinary loss of manganese. There is, however, increased faecal loss through intestinal secretion (2). There is an apparently important enterohepatic circulation and a mixed degree of hepatic damage (raised bilirubin and/or elevated transaminases) was observed in some patients and may account for some manganese retention. This could not be quantified. Raised plasma manganese levels are seen following myocardial infarction (13) and during acute hepatitis or cirrhosis (17).

The large negative manganese balances observed in some of the older patients (Nos. 6, 8, and 16) may be indicative of a need for supplementation with manganese. The infant may be incapable of resorbing manganese excreted in the bile and it is unlikely that positive balance can be achieved without supplementation until reaching an age of 4–5 months.

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