

Lead levels in human placentae from normal and malformed births.

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SUMMARY Placenta lead levels were studied in a series of Birmingham births classified by stillbirth, neonatal death, or survival beyond one week. There was an appreciable range of lead levels even in normal births ($0.15 - 3.56\mu\text{g/g}$) but nevertheless average results showed a pronounced excess of lead in those who failed to survive both birth and the neonatal period. There was no association of placental lead with impaired birthweight among survivors but, in common with other authors, we noted a seasonal variation. The placentae from Indian women had a similar lead levels to those from European women and lower values were found in the normal sibs of stillbirths and neonatal deaths. The possibility is discussed that under conditions of impaired fetal health in pregnancy the placenta may concentrate lead.

The possibility exists that current environment levels of heavy metals such as lead, cadmium, and mercury may present a hazard to the fetus. It is undoubtedly true that exposure of the mother to abnormally high levels of any of these metals can result in fetal death or abnormality. The best documented cases are those associated with mercury, and in particular the Minnemata Bay (Matsumota *et al.*, 1965) and Iraq incidents (Bakir *et al.*, 1973), but in addition several workers (Cantarow and Trumper, 1944) have shown the increased liability to abortion of women employed in industries with a high exposure to lead. The presence of lead in human fetal tissues has been reported and high levels of lead in bone have been reported in two studies of still births (Lanzola *et al.*, 1973).

All three metals have been shown to be capable of crossing the placental barrier in animal studies and to cause fetal malformations and deaths (Ferm and Carpenter, 1967), and both synergistic and antagonistic reactions of lead (Ferm, 1969) and cadmium are known (Holmberg and Ferm, 1969). In view of such evidence it seemed to us that if current environmental levels of these heavy metals is in any way responsible, or partly responsible, for fetal abnormality then one would expect a correlation between the heavy metal levels in placental tissue and some malformations. To examine this assumption we have embarked upon detailed analysis of heavy metals in the placentae of still births, normal births, and abnormal births. There are relatively few published reports (Table 1), and no previous workers have attempted this type of correlation. The present report describes our initial findings with respect to lead.

Table 1 Reported metal levels in normal human placentae

Author	Country	Year	Metal level ($\mu\text{g/g}$ fresh wt)		
			Pb	Cd	Hg
Baglan <i>et al</i>	USA	1974	0.30	0.017	-
Colluci <i>et al</i>	USA	1973	0.96	0.068	-
Einbrodt <i>et al</i>	Germany	1973	0.56	-	-
Horiuchi <i>et al</i>	Japan	1959	0.57	-	-
Suzuki <i>et al</i>	Japan	1971	0.071	-	0.071
Thieme <i>et al</i>	Germany	1974	0.40	-	-
Thurauf <i>et al</i>	Germany	1975	-	0.07	-

Materials and methods

Placental samples from births at the Birmingham Maternity Hospital have been stored at -20°C for the past six years. In the present work 1971 samples were studied from the 1st week of each month and it was shown that the moisture levels (mean level 79.6%) compared favourably with those of fresh samples (80%).

Placental samples were obtained from:

- (1) All neonatal deaths and stillborn births.
- (2) The next two normal births whose mothers did not have Indian names.
- (3) The next birth to a mother with an Indian name.
- (4) Placentae from sibs of all those births were then obtained using a hospital numbers of mothers on an ordered computer listing which printed out only women who delivered at the hospital more than once since 1970.
- (5) Finally, a series of 24 premature normal births (less than 2.5kg) was defined by a consecutive series with non Indian names in 1971.

The samples were thawed, excessive blood removed, the tissue dissolved in Soluene, and the lead estimated by flameless atomic absorption spectroscopy. We have described our solubilisation procedure (Barlow and Khera, 1975) and the

problems concerning trace metal analysis in analytical papers (Khera and Wibberley, 1976). Results have been checked by double blind experiments in our laboratories and by assays in other laboratories by alternative procedures. Extremely high attention to environmental contamination was required to ensure reproducible results.

Results

The lead levels in 126 analyses of placental samples are recorded in tables 2-9. There is an appreciable variation between the lead levels in samples from normal births to non-Indian women whom we will refer to as European (Table 3, mean levels 0.93) and those in samples from malformations (Table 4, mean level 1.49), neonatal deaths (Table 5, mean value 1.73), or still births (Table 6, mean value 1.45). There was no clear relation with cause of death: in fact the three highest values included the rare condition of Trisomy 18 (about 1 in 3000 births), a ring D chromosome, and a congenital cerebral tumour: both the latter conditions are of exceptional rarity (less than 1 in 50 000 births).

Another group of samples were those from Asian mothers from the Indian, sub continent (i.e. India, Pakistan, and Bangladesh) and these showed average levels of 1.13 μg . Our 1971 results also appear to show a seasonal variation in lead levels in normal births with the lowest levels occurring in May for the European births and in September for the Asian births. Dawson et al. (1968) and Baglan et al. (1974) have both reported seasonal variations with minimum levels in summer months. If placental lead is solely an indicator of maternal exposure then one would have expected sibs to have closely similar levels. This has not so far proved to be the case (Table 7).

In order to evaluate the possible contribution of low birth weight to both high lead levels and still births or neonatal deaths, a series of placentae from premature European babies was then studied (Table 8). No correlation was found between low birth weight for these normal births and high mean lead values. The mean values for the various categories are collated together in Table 9.

Discussion

Our first conclusion at this state is that the results appear to be significant and to justify further work. In particular we wish to examine other groups such as Asian samples where stillbirth or neonatal death has resulted. Further work concerning the background of the mother is also required. Just as blood-lead levels increase with urban environment

Table 9 Summary of placental results

Nature of samples	Pb ($\mu\text{g/g}$ fresh wt basis)
Normal Indian Samples	1.13 \pm 0.68
European samples from	
(a) Normal births	0.93 \pm 0.64
(b) Normal premature births	0.96 \pm 0.28
(c) Malformed stillbirths and neonatal Deaths	0.48 \pm 0.69
(d) Neonatal deaths	1.73 \pm 0.57
(e) Stillbirths	1.45 \pm 0.50
Indian and European sibs samples	0.75 \pm 0.19

(Waldron and Stofën, 1974) it is obviously possible that placental lead levels could show some variation within the Birmingham environment, and Thieme et al. (1974) have shown that the placental lead levels were higher for mothers in dense urban areas. A previous blood level survey in Birmingham by Betts et al. (1973) has implicated "Surma" as a possible reason for higher Asian blood-lead levels and in another family study the blood-lead level of an Asian mother who did use "Surma" was found to be higher than that of a father who did not (Warley et al., 1968). However, the Indian placental lead levels were not particularly high: in a fairly random survey of Indian households about a third of women used "Surma", which is mostly lead sulphide (75% PbS). The higher placental lead levels in the stillbirths and neonatal deaths themselves of course do not prove that current environmental lead levels are having any adverse effects on the fetus. It could be argued at this stage that placental lead is an environmental indicator rather than an environmental hazard and as such its correlation with the other well-known environmental indicators such as social class and home environment should be investigated further.

In preliminary work which we have carried out on placental specimens from Stoke on Trent were some of the mothers were working in the pottery industry no high values were obtained and mean values were only half those found in similar Birmingham samples.

If substantiated by further work, our findings concerning placental lead levels in stillbirths and neonatal births are remarkable. In only 7% of the normal births were placental lead levels greater than 1.5 $\mu\text{g/g}$ whereas 61% of the stillbirths or neonatal deaths had levels greater than this. This does not of course mean that lead must be a causal factor in such deaths but it certainly suggests the necessity for further lead analysis on stillbirth tissues. The alternative explanation, that lead accumulated in times of fetal stress is one which the present evidence appears to support. For an assessment of this proposal we have planned further work which will include in addition the analysis of maternal tissues or fluids.

Table 2 Lead in Indian placental samples from normal births

<i>Delivery Data</i>	<i>Mother's age (y)</i>	<i>Gestation period (w)</i>	<i>Mother's weight (kg)</i>	<i>Child's weight (kg)</i>	<i>Sex of child</i>	<i>Social class of father</i>	<i>Pb ($\mu\text{g/g}$ fresh wt basis)</i>
January	20	40	56.5	2.18	M	3	1.33
January	28	36	58.1	3.69	M	4	0.87
January	27	39	63.5	3.14	M	3	0.73
January	36	38	50.5	3.64	F	5	1.20
January	21	40	78.0	3.46	M	3	1.13
January	37	37	77.5	3.50	M	4	1.27
January	39	42	52.2	3.2	F	4	1.00
May	26	-	-	-	-	4	2.16
May	25	40	98.0	3.54	F	3	0.86
May	26	-	-	-	-	3	1.37
May	28	33	61.7	2.58	M	4	3.56
May	20	40	48.5	3.44	F	-	1.25
May	20	36	45.5	2.78	M	3	0.47
May	26	40	85.0	3.76	F	3	1.46
September	18	42	47.0	3.45	M	3	0.52
September	34	40	69.0	3.24	F	4	0.99
September	32	38	48.5	3.86	M	4	0.53
September	24	41	76.0	4.11	F	5	0.63
September	28	40	61.0	3.66	M	-	0.75
September	19	39	53.0	2.58	F	4	0.97
September	28	28	57.5	3.65	F	4	0.76
Arith. Mean	26.8	38.9	62.5	3.34	-	3.7	1.13 \pm 0.68

Range 0.47-3.56 Pb $\mu\text{g/g}$ fresh wt basis
Arith. mean for Jan. 1.08 $\mu\text{g/g}$ fresh wt basis
May 1.59 $\mu\text{g/g}$ fresh wt basis
Sept. 0.72 $\mu\text{g/g}$ fresh wt basis

Table 3 Lead in European placental samples from normal births

<i>Delivery Data</i>	<i>Mother's age (y)</i>	<i>Gestation period (w)</i>	<i>Mother's weight (kg)</i>	<i>Child's weight (kg)</i>	<i>Sex of child</i>	<i>Social class of father</i>	<i>Pb ($\mu\text{g/g}$ fresh wt basis)</i>
January	16	41	46.0	2.90	F	3	1.40
January	33	40	62.5	3.66	F	3	1.20
January	24	40	65.0	3.36	M	3	1.10
January	21	40	57.1	3.26	M	-	0.90
January	32	42	81.5	2.90	F	3	1.20
January	32	42	81.5	2.90	F	3	2.65
May	38	41	73.0	2.62	M	3	0.32
May	32	40	-	4.08	F	-	0.93
May	40	38	62.5	3.32	M	-	0.61
May	32	42	58.5	3.20	F	3	0.55
May	33	37	47.0	3.12	M	3	0.74
May	33	42	83.0	3.42	M	3	0.15
May	39	38	59.5	3.40	M	1	0.38
May	35	39	55.0	2.74	F	4	0.37
May	30	38	76.2	3.30	-	3	0.34
May	23	-	-	-	M	-	0.79
May	20	39	54.5	3.46	F	3	0.77
May	22	41	57.5	3.60	F	2	2.64
September	28	41	58.0	2.94	F	3	0.65
September	33	41	-	3.38	M	-	0.68
September	24	41	57.0	3.61	M	3	1.62
September	25	41	66.5	3.34	M	3	1.19
September	22	43	49.5	4.28	F	1	0.76
September	27	38	65.5	2.32	F	3	0.43
Arith. Mean	38.9	40.2	62.7	3.26	-	2.8	0.93 \pm 0.64

Range 0.15-.2.65 Pb $\mu\text{g/g}$ fresh wt basis
Arith. mean for Jan. 1.41 $\mu\text{g/g}$ fresh wt basis
May 0.72 $\mu\text{g/g}$ fresh wt basis

Sept. 0.89 µg/g fresh wt basis

- = no information.

Table 4 Lead in European placental samples from malformed stillbirths and neonatal deaths

<i>Delivery Data</i>	<i>Mother's age (y)</i>	<i>Gestation period (w)</i>	<i>Mother's weight (kg)</i>	<i>Child's weight (kg)</i>	<i>Sex of child</i>	<i>Social class of father</i>	<i>Nature of malformation</i>	<i>Pb (µg/g fresh wt basis)</i>
11.2.71	17	-	-	-	-	3	Anencephaly	1.37
19.2.71	-	-	-	-	-	-	Multiple Malformations	1.51
11.3.71	22	42	71.0	3.43	F	3	Meningocele	1.60
22.3.71	30	32	49.0	1.10	F	3	Anencephaly	1.74
17.5.71	23	27	52.0	1.08	M	3	Microcephaly	1.07
19.5.71	25	35	72.0	3.30	F	3	Persistent ductus arteriosus	0.53
1.6.71	26	36	62.0	2.67	M	3	Diaphragmatic hernia	0.64
15.6.71	24	41	58.0	3.82	F	3	Haemangioma liver	1.65
29.7.71	30	32	55.5	1.40	M	3	Anencephaly	0.98
24.9.71	25	3*	59.5	3.58	F	4	Extra fingers	0.82
18.10.71	23	47	63.7	2.42	M	2	Chromosomal Abnormality ring D	2.41
5.11.71	31	40	58.0	2.06	F	3	Chromosomal abnormality (trisomy 18)	2.41
25.11.71	24	35	-	2.50	M	3	Congenital Tumour on CNS	2.67
Arith. Mean	25.0	36.9	60.1	2.49	-	3.0	-	1.49 ± 0.69

Range 0.53.-2.67 µg/g fresh wt basis - = no information

Table 5 Lead in European placental samples from neonatal deaths

<i>Mother's age (y)</i>	<i>Gestation period (w)</i>	<i>Mother's weight (kg)</i>	<i>Child's weight (kg)</i>	<i>Sex of child</i>	<i>Social class of father</i>	<i>Pb (µg/g fresh wt basis)</i>	
-	28	-	-	-	-	0.83	
28	-	-	-	-	3	1.42	
19	36	53.0	2.40	M	3	1.89	
31	36	-	2.67	M	-	1.58	
29	32	65.5	1.84	M	3	2.77	
24	35	55.0	3.14	M	3	2.06	
21	38	52.0	2.00	M	3	2.14	
18	29	48.5	1.06	F	3	2.18	
20	38	63.2	2.16	F	3	2.20	
17	39	66.5	3.61	M	-	1.26	
33	39	74.5	3.34	M	4	2.06	
19	31	63.0	1.50	M	3	0.72	
37	27	72.7	0.98	M	2	1.32	
23	31	57.5	1.48	F	3	1.77	
Arith. Mean	24.5	33.8	61.0	2.18	-	3	1.73 ± 0.57

Range 0.72.-2.77 µg/g fresh wt basis - = no information

Table 6 Lead in European placental samples from stillbirths without malformations

<i>Mother's age (y)</i>	<i>Gestation period (w)</i>	<i>Mother's weight (kg)</i>	<i>Child's weight (kg)</i>	<i>Sex of child</i>	<i>Social class of father</i>	<i>Pb (µg/g fresh wt basis)</i>	
34	35	52.2	1.68	F	3	1.18*	
26	41	61.5	3.60	M	3	0.61	
25	40	76.0	3.08	F	3	0.86*	
27	30	82.2	1.15	F	3	1.55*	
27	33	-	1.36	M	4	1.87*	
-	-	-	-	-	-	2.19	
30	41	50.0	2.75	F	3	1.81	
20	41	62.0	2.80	F	-	1.56	
30	34	68.5	1.40	M	3	1.42*	
Arith. Mean	27.4	36.9	64.6	2.23	-	3.1	1.45 ± 0.50

Range 0.61.-2.19 $\mu\text{g/g}$ fresh wt basis- = no information

* Macerated

Table 7 Placental lead in sibs of 1971 samples

<i>Racial Group</i>	<i>Nature of previous birth</i>	<i>Nature of present (w) birth</i>	<i>Mother's Age(y)</i>	<i>Gestation period (kg)</i>	<i>Mothers weight (kg)</i>	<i>Child's weight (w)</i>	<i>Sex of child</i>	<i>Social class of father</i>	<i>Pb ($\mu\text{g/g}$ fresh wt basis)</i>
Indian	Normal	Normal	28	37	70.0	3.15	M	4	0.93
Indian	Normal	Normal	27	40	72.0	3.26	F	3	0.79
Indian	Normal	Normal	22	41	57.2	3.74	F	4	0.91
Indian	Normal	Normal	28	39	67.5	3.24	F	4	0.67
Indian	Normal	Normal	27	40	46.5	2.90	M	4	0.78
Indian	Normal	Normal	28	40	67.5	3.52	F	3	0.76
Indian	Normal	Malformed	22	34	52.0	2.22	M	4	0.79
Indian	Malformed	Malformed	24	39	46.2	2.34	F	3	0.81
Indian	NND	Died (RDS)	32	33	47.0	2.00	M	4	0.81
Indian	Malformed	Died due to Infection	29	31	67.0	1.86	M	3	0.94
European	Normal	Normal	21	39	53.0	2.62	M	3	0.55
European	Normal	Normal	21	41	71.0	3.72	M	3	0.83
European	NND	Normal	20	40	52.5	4.10	M	3	0.25
European	NND	Normal	21	38	44.5	2.40	F	4	0.82
European	NND	Normal	28	40	58.5	3.48	M	3	0.71
European	NND	Normal	21	41	62.2	4.22	F	3	0.60
European	NND	Normal	26	37	58.5	5.12	F	3	0.40
European	Stillbirth	Normal	26	39	53.0	3.39	M	4	1.00
European	Stillbirth	Normal	31	41	54.0	3.26	F	3	0.69
European	Stillbirth	Normal	26	39	67.5	3.26	M	3	0.79
European	NND	Malformed	33	38	60.5	3.36	M	3	0.99
Arith. Mean			25.8	38.4	58.5	3.19		3.4	0.75 \pm 0.19
Indian		European							
Range 0.67.- 0.94		0.25 – 1.00 $\mu\text{g/g}$ fresh wt basis							
Arith. Mean 0.82		0.59 $\mu\text{g/g}$ fresh wt basis							

Table 8 Placental lead in European premature birth samples

<i>Mother's age (y)</i>	<i>Gestation period (w)</i>	<i>Mother's weight (kg)</i>	<i>Child's weight (kg)</i>	<i>Sex of child</i>	<i>Social class of father</i>	<i>Pb ($\mu\text{g/g}$ fresh wt basis)</i>
23	39	44.5	2.34	M	2	0.75
28	30	-	1.62	F	5	0.70
28	40	77.0	2.12	M	4	1.59
24	34	59.5	2.23	F	1	1.09
31	37	68.0	2.40	M	3	0.96
23	39	53.0	2.22	F	-	0.90
33	40	-	2.30	M	1	0.98
27	30	57.0	1.43	M	3	0.21
30	40	58.2	2.42	F	3	1.25
31	37	59.5	2.48	F	-	1.19
27	40	61.0	2.26	M	4	0.55
25	39	49.0	2.22	F	3	0.92
24	42	49.0	2.42	M	4	1.25
38	33	47.5	1.98	M	3	0.88
33	36	59.0	2.32	M	4	0.91
32	37	62.5	2.32	F	3	0.76
27	35	51.0	2.40	M	-	1.25
27	38	54.5	2.40	F	3	0.84
27	40	69.5	2.32	F	3	1.29
28	37	57.0	2.34	M	2	0.85
32	39	54.5	2.37	M	2	0.91
27	-	-	2.43	-	-	0.87
23	-	-	2.38	-	-	1.11
27	-	-	2.12	-	-	0.97
Arith. Mean 28.1		37.2	57.4	2.24	2.9	0.96 \pm 0.28

Range 0.21.-1.59 $\mu\text{g/g}$ fresh wt basis - = no information

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