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Study of Possible Correlations Between Prenatal Brain Development and Placental Weight [1]

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One of several factors, studied in our laboratory, that influence DNA synthesis and cell number in prenatal brain [4, 6, 12], is placental development [review in 13]. WINICK [10] and STEPHAN and CHOW [5] have demonstrated that maternal dietary restriction results in placentas of lower cell number, and lower weight, respectively. The newborns of such mothers have lower neonatal brain cell number [10, 12] but the relationship between the placental and the cerebral development has not been established.

Several reports indicate a positive correlation between placental weight and birth weight [review in 3]. The development of the placenta, especially from the point of view of placental cell number (DNA) has been the subject of a recent study by WINICK and NOBLE [8] and by WINICK *et al.* [9].

The object of this work is the study of correlations between various neonatal parameters, especially placental weights and brain development in *normal* animals (rabbits). In particular, the aim of the study was to establish whether, on a statistical basis, an individual with a heavier term placenta is also likely to have a higher neonatal cerebral weight or cerebral cell number.

Materials and Methods

11 6-month-old does of the New Zealand White strain, weighing 3–4 kg, were mated and 80 fetuses and their (fetal) placentas were removed by cesarean section on day 30 of gestation; an additional 10 does were used for cesarean sections between days 20 and 28

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providing 7–20 fetuses for each of the days. The fetuses were weighed, then cerebral hemispheres (without cerebellum and olfactory lobes) were dissected out, and their wet weight as well as the wet weight of the corresponding placentas, were determined. Total cerebral DNA was determined as described previously [4, 11]. From this DNA content one can calculate the total number of cerebral cells [13]: neonatal cerebral DNA 1,804 μg (mean value, figure 3); DNA content of diploid rabbit cell [7] $5.3 \times 10^{-6}\mu$g; total neonatal rabbit cerebral cell number 3.4×10^{8}.

In the rat, neurons (neuroblasts) cease to proliferate around birth and the neonatal cell number (as measured by neonatal DNA) is indicative of the final neuron number [for a discussion see 4, 6, 12, 13]. To our knowledge, the time of cessation of neuron proliferation in the rabbit has not been reported; thus, for the time being, the neonatal cerebral DNA determined in this work, indicates only the total cerebral cell number. As in the case of term in the rat and hatching in the chick [13] we now find that this number (DNA content) also reaches a plateau around term in rabbit cerebrum: 45% at 20 days, 67% at 22, 79% at 24, 88.5% at 25.5, 97.5% at 27.5, assuming 100% at 30 days. This time, then, is convenient for DNA determination, because a small error in the estimated fetal age does not result in

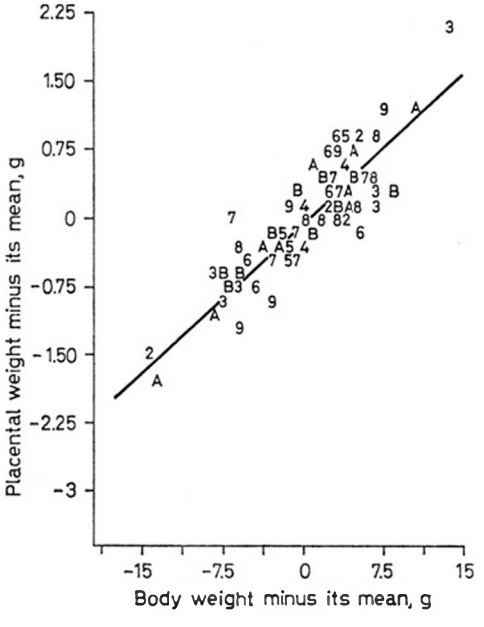

Fig. 1–3. Correlations between parameters at 30 days of pregnancy (rabbit). Numbers 1 to 9 indicate offspring of the same litter, with A and B representing litter number 10 and 11, respectively. Since the graphs illustrate the spread around the slope, identical values for littermates were eliminated due to technical difficulties in plotting.

Fig. 1. Placental weight minus its mean (g) vs body weight minus its mean (g). Equality of slopes, $F = 1.89$ on 9 and 54 degrees of freedom (DF) with probability, $p = 0.07$. Common slope regression coefficient, $b = 0.109$, $t = 14.66$ on 63 D F, probability (zero slope error), $p < 0.0001$.

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an appreciable difference in cerebral DNA content. Day 30 was chosen as close enough to term (31.5), without the risk of natural birth and the loss of the placenta; the corresponding values are referred to as 'neonatal'.

The following statistical method was utilized [14] to determine if significant linear correlations exist between any of the following measured parameters: placental weight, body weight, cerebral weight, and cerebral DNA. A slope was estimated for each individual litter of pups and an F test for equality of slopes applied to demonstrate the constancy of the linear relationships amongst all of the litters. If the slopes were indeed similar, yielding $p > 0.05$, a regression coefficient, b, was estimated for the entire group. The degree of correlation was determined by testing the equality of the regression coefficient to zero, with $p < 0.01$ constituting significance.

Results and Discussion

The problem of quantification in the placenta is a difficult one because of the number of factors involved. The permeability of the placenta, the exchange surface, and the maternal blood flow to the placenta are all involved, to a not well defined degree. It has been reported that maternal blood flow is greater to the placentas of larger fetuses [2]

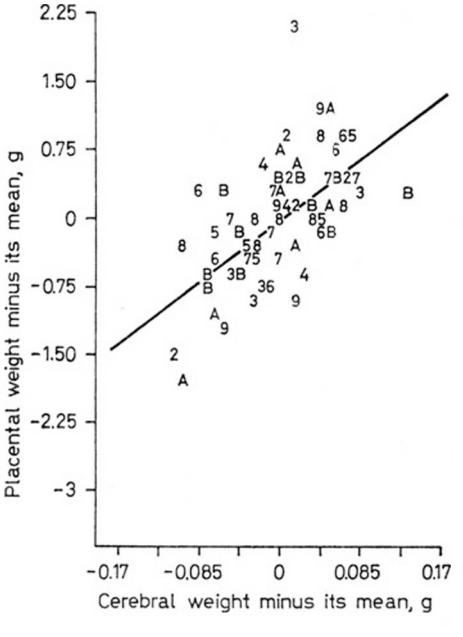

Fig. 2. Placental weight minus its mean (g) vs cerebral weight minus its mean (g). $F = 1.56$ 9 and 54 DF, $p = 0.15$; $b = 7.785$, $t = 5.73$, 63 D F, $p < 0.0001$.

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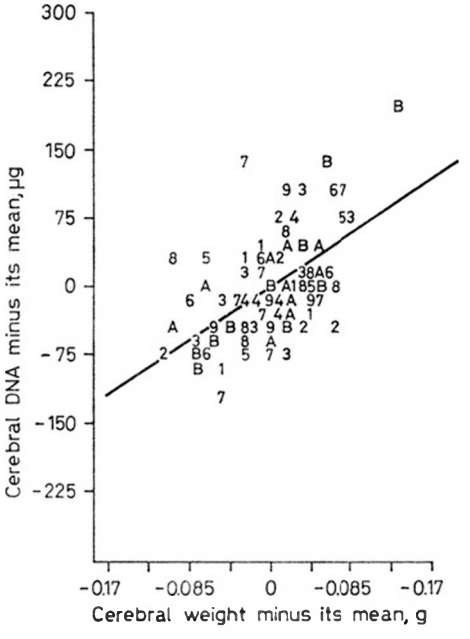

Fig. 3. Cerebral DNA minus its mean (μg) vs cerebral weight minus its mean (g). $F = 1.06$, 10 and 58 DF, $p = 0.406$; $b = 682.41$, $t = 5.56$, 68 DF, $p < 0.0001$.

and that blood flow to the uterus increases in proportion to the placental weight [1].

In our work a sample of 20 placentas were dried to constant weight; their water content so determined was practically identical (83.35% $\pm$ 0.63% SD). Thus, for all practical purposes, the weights of placentas are also indicative of the dry weights of their tissues and of the blood therein. This parameter then, the placental weight, was chosen in the present work, bearing in mind the limitations involved.

Our results (fig. 1–3) indicate that highly significant correlations can be demonstrated between 'neonatal' body weight and placental weight, cerebral weight and placental weight, and cerebral DNA (cell number) and cerebral weight. The correlation between cerebral DNA and body weight may be considered as being on the borderline of significance ($p = 0.0186$); the correlation between cerebral DNA and placental weight for this size sample was not significant ($p = 0.0589$).

Younger fetuses show the same trend of correlations down to at least the 20th day.

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In conclusion, on a statistical basis, an individual animal with a heavier term placenta is also likely to have a higher neonatal cerebral weight; the latter is also likely to have a higher number of neonatal cerebral cells (DNA), although for the size of the sample studied, a direct significance of correlation between placental weight and cerebral DNA could not be demonstrated.

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Summary

Correlations between various neonatal parameters, especially placental weights and brain development in normal animals (rabbits) have been studied. It was found that, on a statistical basis, an individual animal with a heavier term placenta is also likely to have a higher neonatal cerebral weight; the latter is also likely to have a higher number of neonatal cerebral cells (DNA).

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