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Maturation of glucocorticosteroid activity in the foetal guinea-pig during the end of gestation

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Summary. Cortisol levels were very high in maternal and foetal plasma during the last days of pregnancy. The percentage of free cortisol was maximal in plasmas of the mother and the foetus on the last day before parturition ; the increase in free cortisol levels resulted from the saturation of transcortin binding capacity and from a higher adrenal secretion rate at that time due to stress at the onset of labor. On the last day of pregnancy the foetal adrenals could cover the hormonal needs of the foetus, the foetal-maternal transter of cortisol being similar to the maternal-foetal transfer. However, the low TCM ot cortisol indicated that cortisol catabolism was still immature. The origin of foetal transcortin is still unknown.

Introduction.

In numerous mammalian species, parturition is associated with an increase in foetal, and sometimes maternal, corticosteroid plasma levels (Bassett and Thornburn, 1969 ; Kamoun, 1970 ; Mulay *et al.*, 1973 ; Illingworth *et al.*, 1974 ; Jones, 1974 ; Dalle *et al.*, 1978). Such an increase results from changes in foetal and maternal adrenal activity and in hormonal transfers across the placenta. High plasma cortisol levels have been observed in newborn guinea-pigs (Dalle and Delost, 1974). These high levels result from a regular rise in foetal plasma cortisol levels during the last days of gestation, which parallels the increase in maternal plasma and adrenal cortisol concentrations, foetal adrenal cortisol content increasing only during the last 24 hrs of gestation (Dalle and Delost, 1976). Recent studies (Dalle and Delost, 1979) have shown that 90 p. 100 of the cortisol found in the foetal plasma during the end of gestation is of maternal origin. These studies have also demonstrated that placental transfers of cortisol both from the mother to the foetus and from the foetus to the mother increase near parturition, as does the cortisol secretion rate of the foetal adrenals at that time. Thus, the foetal secretion rate of cortisol becomes similar to its production rate in the newborn. In the work reported here, we have measured levels of free cortisol and the different parameters of cortisol binding in the plasmas of dams, foetuses and newborns.

Material and methods.

Female guinea-pigs (Dunkin-Hartley strain ; length of gestation : 68 days) were caged with a male during 24 hrs at oestrus time when the vagina was entirely open. Thus, mating time was known at ± 12 hrs. The pregnant mothers and their foetuses, obtained by Caesarean section, were killed by decapitation at 60, 62, 64, 66 and 68 days of gestation between 9 and 11 a.m. The newborns and their mothers were killed exactly at 0 hr and 50 hrs *post-partum*. Each group included 6-10 animals.

Plasma cortisol levels were estimated by competitive protein binding according to Murphy (1967), modified as described previously (Dalle and Delost, 1976). Cortisol binding parameters were determined using the method of Pearlman and Crepy (1967), adapted for corticosteroids (Pradier, 1979). Non specific binding of plasma cortisol (other than albumin-bound) was undetectable, and transcortin-binding capacity was similar to that measured with the other usual methods used in our laboratory. The ratio of plasma cortisol : progesterone was high (around 30) (Dalle and Delost, 1976), and progesterone did not interact in the binding of cortisol. The parameters were calculated according to Labrie *et al.* (1966). The binding capacity of transcortin for cortisol was expressed as μg of cortisol/100 ml plasma. The results were expressed as mean ± SEM ; the statistical significance was evaluated using Student's *t* test.

Results.

Pattern of transcortin binding capacity for cortisol (fig. 1). — During the last week of pregnancy the binding capacity of transcortin for cortisol in pregnant guinea-pigs was 5 times higher than in non-pregnant females. It decreased sharply just before (— 38 p. 100 between day 68 and parturition ; $P < 0.05$) and after parturition until day 20 *post-*

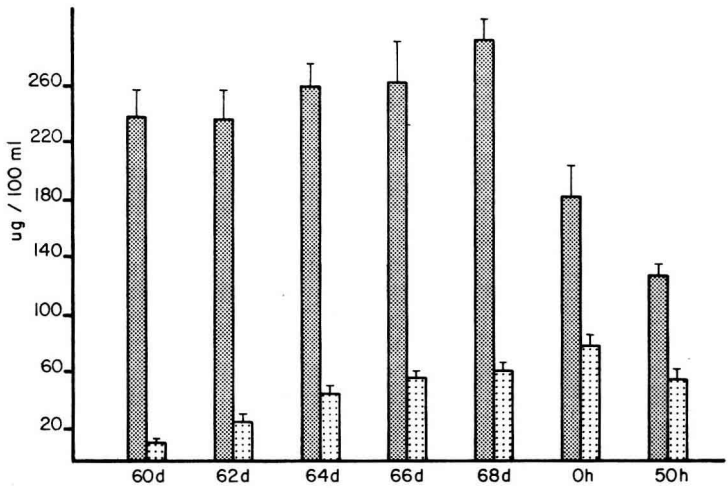

FIG. 1. — *Changes in transcortin binding capacity for cortisol in maternal* (▒ ; n = 6-10) *and foetal and neonatal* (▭ ; n = 6) *guinea-pigs during the perinatal period.*

partum when the values were similar to those measured in adult females (about 50 μg/100 ml). In foetuses the binding capacity of transcortin for cortisol was low on day 60 of gestation (15 μg/100 ml). Thus, it increased during the last week, being maximal at birth (80 μg/100 ml). However, it always remained under the maternal values.

Pattern of $S_A K_A$ (*albumin binding factor for cortisol*). The part of cortisol bound to albumin equalled free cortisol $\times$ $S_A K_A$ (S_A : albumin binding capacity for cortisol ; K_A : albumin-cortisol binding constant).

In dams $S_A K_A$ values were similar to those measured in non-pregnant females (1.5-3.0). These values did not vary significantly during the end of gestation. Foetal $S_A K_A$ values ranged between 0.4 and 0.7, increasing at birth (+ 245 p. 100, $P < 0.05$). $S_A K_A$ values were then not significantly different in newborns, adult females and pregnant animals.

K_T *values* (*transcortin binding constant for cortisol*). During the whole experimental period, K_T values were similar in mothers ($1.5 \pm 0.2\ 10^8\ M^{-1}$), foetuses ($1.3 \pm 0.2\ 10^8\ M^{-1}$) and adult females ($1.0 \pm 0.3\ 10^8\ M^{-1}$).

Free and bound cortisol plasma levels in mothers (fig. 2 and fig. 3). — During the last week of pregnancy plasma cortisol levels were very high ; they increased until day 68 and then regularly decreased (— 45 p. 100 between day 68 and parturition ; $P < 0.001$; — 53 p. 100 between parturition and 50 hrs *post-partum* ; $P < 0.001$) until day 20 *post-partum* when they were similar to those measured in the non-pregnant females (50 μg/100 ml). A high percentage (96 p. 100) of cortisol bound to transcortin was found at day 60 of gestation. This percentage decreased to 63 p. 100 on day 68 then rose to 93 p. 100 during the 50 hrs *post-partum*. This last value was similar to the one measured in non-pregnant females. Plasma cortisol bound to albumin increased from 2 p. 100 on day 60 to 28 p. 100 on day 66. It remained high on day 68 (23 p. 100)

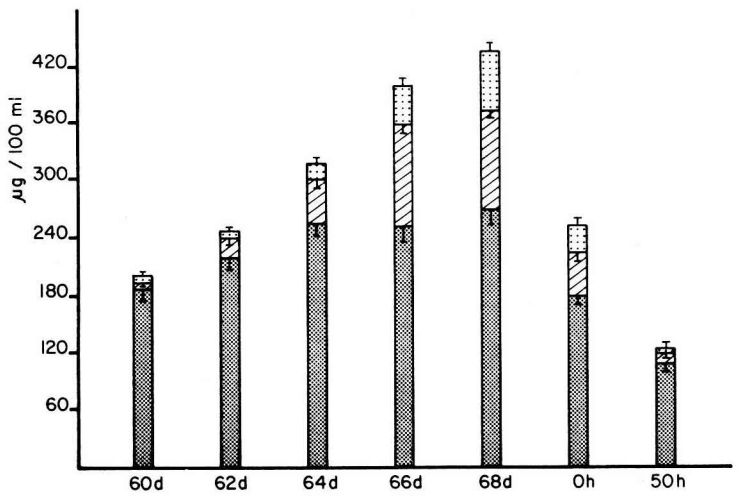

FIG. 2. — *Development of free* ( ), *albumin-bound* ( ) *and transcortin-bound* ( ) *cortisol in the plasma of the mother guinea-pig during the perinatal period.*

and at parturition (18 p. 100). However at 50 hrs *post-partum* the percentage of plasma cortisol bound to albumin (6 p. 100) was similar to the level measured in non-pregnant females. Plasma free cortisol paralleled albumin-bound cortisol. It increased from 2 p. 100 on day 60 to 14 p. 100 on day 68 (highest value ; 61 μg/100 ml), and plasma free cortisol levels were similar to those of the adult at 50 hrs *post-partum*.

Free and bound cortisol levels in the foetus (fig. 3 and fig. 4). — The percentage of transcortin-bound cortisol (80 p. 100 of plasma cortisol between days 60 and 64)

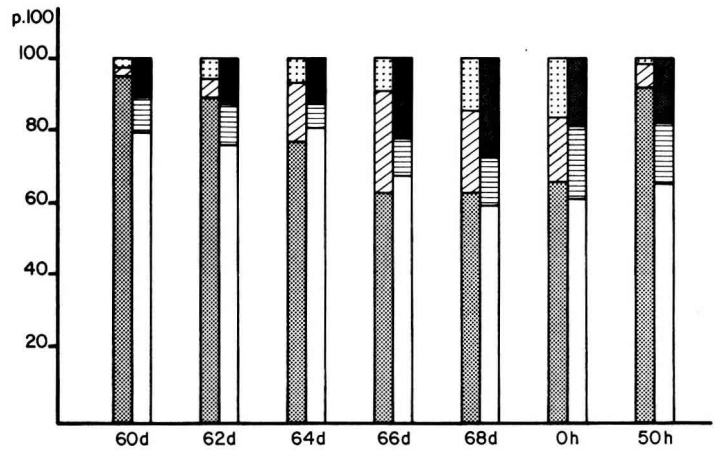

FIG. 3. — *Changes in percentage of free, albumin-bound, and transcortin-bound cortisol in mother, foetal and newborn guinea-pigs during the perinatal period.* Mother : *free* ( ) ; albumin-bound ( ) ; transcortin-bound ( ). Foetal and newborn : free ( ) ; albumin-bound ( ) ; transcortin-bound (open bars).

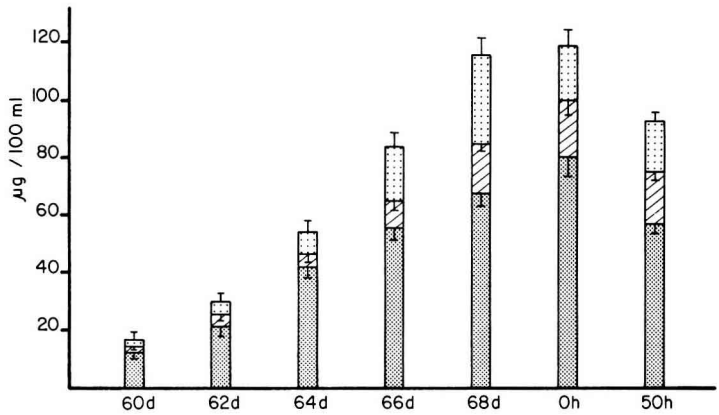

FIG. 4. — *Development of free cortisol* ( ), *albumin-bound cortisol* ( ) *and transcortin-bound cortisol* ( ) *in foetal and newborn guinea-pigs around parturition.*

decreased until birth (57 p. 100). During the whole experimental period the percentage of albumin-bound cortisol remained around 10 p. 100, although free cortisol rose sharply from 13 p. 100 on day 60 to 27 p. 100 on day 68. At those stages free cortisol levels were 7 μg/100 ml on day 64, 19 μg/100 ml on day 66 (+ 171 p. 100, $P < 0.01$) and 33 μg/100 ml on day 68 (+ 74 p. 100 between days 66 and 68, $p < 0.05$).

Discussion.

The estimation of plasma and adrenal cortisol concentrations (associated with the measurement of metabolism) and the transfer and the binding of plasma cortisol in maternal and foetal guinea-pigs in the last days of gestation and at birth could provide a better understanding of the maturation of foetal adrenal activity under maternal influence. High plasma cortisol levels and high transcortin binding capacity were found in pregnant guinea-pigs (Gala and Westphal, 1967 ; Diamond *et al.*, 1969). These values were maximal on day 68, and decreased sharply at parturition as did progesterone and PBG plasma levels (Lea *et al.*, 1976). High plasma PBG levels were associated with very low progesterone TCM during gestation (Illingworth *et al.*, 1970). Similarly, high transcortin plasma levels paralleled a low cortisol TCM (3 times lower than in non-pregnant females) (Dalle and Delost, 1979).

During the last week of gestation plasma cortisol levels increased significantly (+ 125 p. 100, $P < 0.001$). During the same period plasma transcortin levels did not vary significantly but free cortisol levels increased from 7 to 61 μg/100 ml (+ 770 p. 100, $P < 0.001$) ; simultaneously, cortisol transfer across the placenta from the mother to the foetus increased (+ 130 p. 100, $P < 0.01$). However these results do not indicate whether the two processes were correlated. Indeed, numerous morphological changes occurred in the guinea-pig placenta during the end of pregnancy which might explain the increase in cortisol transfers (Firth and Farr, 1977 ; Kaufmann and Davidoff, 1977).

In foetal guinea-pigs the increase in plasma cortisol levels paralleled that of transcortin during the end of gestation ; similar results have been demonstrated in foetal sheep (Fairclough and Liggins, 1975) and rat (Martin *et al.*, 1977 ; Van Baelen *et al.*, 1977). As in rat foetuses (Van Baelen *et al.*, 1977), free cortisol levels in foetal guinea-pigs increased sharply during the last days of gestation. The transfer of corticosteroids across the placenta has never been studied in rats. In guinea-pigs the rise in free cortisol levels occurring just before parturition, in foetuses as in dams, seemed to be due to an increased cortisol secretion rate of the foetal and maternal adrenals. This increased rate might result from a response of the adrenals to the stress associated with the onset of labor. At that time, the transfer of cortisol across the placenta from the foetus to the mother equals that from the mother to the foetus (Dalle and Delost, 1979).

In conclusion, during the end of gestation in guinea-pig the simultaneous measurement of plasma and adrenal cortisol levels, binding, and cortisol metabolism and transfer in both dams and foetuses, demonstrates that, although a large part of foetal cortisol is of maternal origin, the foetal adrenals are able to cover the hormonal needs of the foetus just before term. At that time, the enzymatic systems involved in cortisol catabolism are still immature.

In spite of high foetal levels, the origin of transcortin remains unknown. It may be of foetal origin, as in birds (Gasc and Martin, 1978), its production being regulated by

hormones such as thyroxine and cortisol, or it may be of maternal origin, as in the mouse (Savu *et al.*, 1977). The hypothesis of a placental transfer of this protein in the guinea-pig will be investigated in our laboratory. The increase in free cortisol levels observed in both dams and foetuses during the last two days of pregnancy results from adrenal response to the onset of labor. The rise in foetal free cortisol levels argues in favour of foetal adrenal maturity occurring just before birth.

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Résumé. Les cortisolémies fœtale et maternelle augmentent régulièrement et fortement au cours de la dernière semaine de la gestation chez le cobaye. Le pourcentage de cortisol libre est maximum chez la mère et le fœtus la veille de la parturition ; son augmentation provient d'une part de la saturation de la capacité de liaison de la transcortine pourtant très élevée à ce moment et d'autre part d'une sécrétion surrénalienne maternelle et fœtale en cortisol stimulée par le stress de la parturition. Chez le fœtus en effet, les surrénales ont atteint, juste avant la naissance, un degré de maturation suffisant pour assurer les besoins de l'organisme du nouveau-né en hormone et le fœtus retransfère à la mère la totalité du cortisol que celle-ci lui envoie. Toutefois, le catabolisme hépatique du cortisol reste faible au cours de cette période et le problème de l'origine de la transcortine du fœtus reste posé.

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