

cases result in perinatal deaths or require cortisone treatment within some weeks after birth.

The present communication describes a case of otherwise uncomplicated pregnancy associated with an extremely low maternal oestriol excretion (1.9–3.3 mg/24 h during the 36–37th week), but with an excretion of oestrone + oestradiol-17 β at the lower normal limit. A male infant was born which, with exception of a very mild ichthyosis, showed normal clinical findings at birth as well as 10 months later. There were no signs of cerebral dysfunction. The placental steroid sulphatase activity was normal. 5 days after birth the plasma levels of cortisol were within the lower limit of the normal values but raised later. Urinary pregnanetriol excretion and response to ACTH stimulation were normal.

Gas chromatographic – mass spectrometric analysis of the infants urine revealed a steroid pattern resembling that found in anencephaly with a total absence of 3 β -hydroxy-5-ene-steroids. This particular child might therefore have lacked the foetal zone of the adrenal cortex, which is the case in anencephaly. Due to the normal clinical findings it might be speculated that the excessive production of 3 β -hydroxy-5-ene-steroids in the foetal and early infancy stages is not of vital importance.

76. Urinary excretion of aldosterone, tetrahydrocortisone (THE), and tetrahydrocortisol (THF) in premature infants, W. RAUH, L. WILLE, P. VECSEI, W. VIELHAUER and H. WILL, Department of Pediatrics and Pharmacology, University of Heidelberg, Germany

Urinary excretion rates of aldosterone, THE, and THF were determined radioimmunologically in 7 premature infants during the first 10 days of life. Specific radioimmunoassays for THE and THF were developed. White New Zealand rabbits were immunized against complexes of THE- and THF-20-oximes and bovine serum albumine. After an immunization period of 6 months specific antibodies against THE and THF were obtained. Radioimmunochemical analysis showed that THE- and THF-antibodies also bind with the glucuronates of THE and THF. Urinary excretion rates of THE (0.002–0.1 mg/m²/24 h) and THF (0.005–0.05 mg/m²/24 h) were extremely low when compared with normal values of older children and adults (THE: 0.3–3.0 mg/m²/24 h, THF: 0.3–2.5 mg/m²/24 h). Aldosterone excretion ranged from 3.0–60.0 μ g/m²/24 h, the normal values for adults being 2.0–12.0 μ g/m²/24 h. There was a definite increase in THE excretion during the first 10 days of life, whereas THF and aldosterone excretion did not change during this period of time. In this study a special pattern of adrenal steroid excretion in premature infants is demonstrated. The differences between premature infants and older children can be explained by changes in adrenal function and hormone metabolism.

77. Role of aldosterone in sodium homeostasis in premature neonates, J. W. HONOUR, C. H. L. SHACKLETON and H. B. VALMAN, Northwick Park Hospital and Clinical Research Centre, Watford Road, Harrow, Middlesex HA1 3UJ, England

The urinary steroid excretion of pre-term infants (22–30 weeks gestation age) has been studied following the repeated observation of a persistent period of urinary salt loss and hyponatraemia developing between the first and third weeks of life. This investigation was therefore undertaken to study the production of steroids by pre-term

infants during this period. Aliquots of 24 h urine collections from pre-term infants were hydrolysed enzymically, and the steroids extracted on Amberlite XAD-2 columns and fractionated by Sephadex LH-20 chromatography (Shackleton C. H. L., Gustafsson J.-A. and Mitchell F. L.: *Acta endocr., Copenh.* **74** (1973) 157). Steroids present in fractions were analysed as methyloxime-trimethylsilyl ethers using capillary column gas chromatography and gas chromatography-mass spectrometry (GC-MS). The steroid excretion was similar to that of full-term neonates, apart from the finding of relatively large amounts of tetrahydroaldosterone, the principal urinary metabolite of aldosterone. Selected ion-monitoring GC-MS was used for the specific quantitative determination of urinary tetrahydroaldosterone, 3 β -allo-tetrahydroaldosterone being used as internal standard (Shackleton C. H. L. and Honour J. W.: *Z. Klin. chem. klin. biochem.* **12** (1974) 259). A weak positive sodium balance was observed during the first weeks of life and this was associated with low plasma sodium and elevated urinary tetrahydroaldosterone excretion (100–300 μ g/24 h). In full-term infants the excretion of tetrahydroaldosterone is between 1 and 25 μ g/24 h. These results show that pre-term infants are capable of synthesizing aldosterone in response to low plasma sodium concentrations. An increasingly positive sodium balance was observed from the third week of life but tetrahydroaldosterone excretion remained high and was still elevated when normal plasma sodium concentration was established. The results suggest for the first time that the renal tubular response to aldosterone is low in pre-term infants although the juxta-glomerular apparatus is considered to be functional.

78. Steroid metabolism by mouse placental tissue *in vitro*, G. H. OKKER-REITSMA, Laboratory of Cell Biology and Histology, University of Leiden, Leiden, The Netherlands

It is known that the mouse placenta is capable of steroid metabolism at some time of its existence, but to what extent is still not clear. It is generally accepted that in the rodent placenta steroid formation is one of the functions of a particular cell type – the mononuclear giant cells. The present study was designed to look into the capacity for steroid synthesis of the mouse placenta during development, with special emphasis on the role of the giant cells. Mouse placental tissue of 10 and 15 days gestation was incubated for 4 h in 1 ml medium 199 with either [³H]-progesterone or [³H]-dehydroepiandrosterone. The placenta of 10 days gestation shows many well developed giant cells. The labyrinth and the "basal zone" are still very small; consequently a division of the placenta in parts with and without giant cells is not possible. The explants of placental tissue of 15 days gestation, however, consisted of a part of the placenta which contains many giant cells or a part of the labyrinth which should not contain any giant cell. At 15 days the giant cells are smaller than earlier in development. The steroid metabolites were extracted and subjected to paper- and thin layer chromatography. The identified metabolites implicate the presence of enzymes involved in steroid synthesis. In the placenta of 10 and 15 days 3 β -hydroxysteroid dehydrogenase (3 β -HSD), 5 α -reductase and C₂₁-C₁₉ lyase could be detected. Incubations of placental tissue of 10 and 15 days with giant cells showed 20 α -HSD activity. Placental tissue of 15 days with giant cells showed also 17 β -HSD activity. 21-Hydroxylase was found in the labyrinth of the placenta of 15 days gestation. From these results we can conclude that giant cells as well as the labyrinth of the mouse placenta are

capable of steroid metabolism. They differ in the steroid enzyme pattern. This enzyme pattern changes during development.

79. Steroid hormones production of the embryonic chick ovary, C. T. TENG and C. S. TENG, Department of Cell Biology, Baylor College of Medicine, Houston, Texas, U.S.A.

Ample evidence has shown that steroidogenesis begins early in the embryonic chick ovaries and the hormones secrete from the ovary effect the sexual differentiation of the Mullerian ducts in the embryonic chicks. Therefore, the amount and types of the steroid hormone secreted from the developing embryonic ovary are critical for the sex tract development. By radioimmunoassay, we were able to detect precisely the amount of steroid released into the culture medium from the embryonic left and right ovaries during organ culture. Approximately 20 mg of ovarian tissue from 15-day-old embryos were cultured in 1 ml of Hanks' solution at 41°C for 4 h, the amount of estradiol (E₂) and testosterone (T) released from the left ovary into the culture medium were 165 pg and 11.8 pg

per ovary respectively. With the presence of 100 IU of HCG in the culture medium the amount of E₂ and T released into the medium were 500 pg and 98 pg per ovary respectively. Steroid hormone production from the 15th day embryonic ovary in response to HCG stimulation was dosage dependent. In the presence of 20 IU of HCG in the culture medium could cause maximum effect on both E₂ and T production. With the presence of 1 mM dibutyl cyclic AMP in the culture medium causes the same effect as 20 IU HCG does. When MIX (a potent phosphodiesterase inhibitor) was also present in the culture medium the steroid production was enhanced to 758 pg of E₂ per ovary. The involuting right ovary of the 15th day embryonic chick also releases steroid hormones into the culture medium, yet in a significantly lower amount. Right ovary responses to HCG stimulation with 250% increase in E₂ production and 800% increase in T production. This report suggested that the 15th day embryonic ovaries (developing and involuting ones) produced steroid hormones and responded to gonadotropic hormone stimulation, and the gonadotropic hormone action probably mediated through cyclic AMP. (Supported by NIH Grant HD-08218-03).