together to steroid-free serum in appropriate concentrations for the added specific antigen levels. Therefore, T and D can be simultaneously determined. The values calculated as described have shown acceptable correlations with the respective values determined after celite-chromatography.

46. Centriole steroids: proposal for a new cell control mechanism by steroid hormones, ITALO NENCI, ELISABETTA MARCHETTI, GUIDO A. FABRIS and ANDREA MARZOLA, Istituto di Anatomia e Istologia Patologica, Università di Ferrara, Via Fossato di Mortara 64b, 44100 Ferrara, Italy

Centrioles are known to play important roles in fundamental cell activities, such as mitosis, ciliogenesis and ciliary motility, establishment of the cell polarity, etc., which are deeply involved in the regulation of cell behaviour. Specific steroid antibodies, by immunocytochemical methods at U.V., light and electron microscopy, are able to trace immunoreactive steroids at the centriole level in cells from several steroid target tissues. Rat endometrium displays definite changes of centriole steroids after major endocrine upsets. Moreover, as steroid hormones appear to be vitally taken up by centrioles grown *in vitro*, the nature of the centriolar binder and its relationships with the receptor system have been fruitfully investigated. It seems justified to suggest that centriole steroids can play some important role in mediating steroid hormone action on the cell duplication, growth and differentiation.

47. Mechanism by which foetal cortisol induces parturition in sheep and goats, A. P. F. FLINT, E. JANE KINGSTON, J. S. ROBINSON and G. D. THORBURN, Agricultural Research Institute of Animal Physiology, Babraham, Cambridge CB2 4AT, and Nuffield Institute for Medical Research, and Nuffield Department of Obstetrics & Gynaecology, University of Oxford, Oxford OX3 9DU, England

Parturition in sheep and goats appears to be initiated by increasing foetal levels of cortisol. In sheep, foetal cortisol increases oestrogen synthesis, and decreases progesterone synthesis, by the placenta; uterine prostaglandin synthesis resulting from these endocrine changes causes myometrial contractions and labour. Our previous work has suggested that the effect of foetal cortisol in sheep is to activate placental steroid 17α-hydroxylase and C-17,20 lyase, enzymes catalysing the conversion of progesterone into oestrogens. Such a mechanism satisfactorily explains why glucocorticoid infused into foetal lambs causes increased oestrogen synthesis characteristic of term labour while suppressing foetal adrenal androgen secretion. Recent work indicates that a similar mechanism may operate in goats. Dexamethasone infused to 4 foetal kids (7–11 days after implanting foetal vascular catheters at 115–123 days gestation) induced parturition in 58 ± 6 h (mean ± S.E.M.); maternal endocrine changes in these animals were characteristic of term labour. Examination of steroid metabolism by placental extracts *in vitro* indicated the activation or induction of 17α-hydroxylase after exposure to either endogenous ($n = 7$) or exogenous ($n = 4$) glucocorticoid (but not after prostaglandin-induced labour). It is suggested therefore that in the goat foetal cortisol stimulates placental oestrogen synthesis by activating placental enzymes, and that increased oestrogen secretion causes progesterone withdrawal (luteolysis) and increased myometrial contractility through stimulating uterine prostaglandin $F_{2\alpha}$ production.

48. Identification of unknown steroids by NMR spectroscopy and mass spectrometry: discovery of a natural spirolactone derivative in man and animal, P. GENARD*, M. PALEM-VLIERS* and W. EECHAUTE,† *University of Liege (Belgium), Hôpital de Bavière, Departement de Clinique et de Pathologie Médicales (Professeur H. Van Cauwenberge) and †University of Ghent (Belgium), Laboratorium van normale en pathologische fysiologie (Professeur J. Leuzen)

Nuclear Magnetic Resonance and Mass Spectrometry are without doubt the most sophisticated methods for the study of molecular structures in biochemistry. In the present paper we relate the discovery by means of this method of an unknown steroid in the urine of normal man, dog and rat. The experimental subjects did not receive any drug or treatment. Our identification studies indicate that the molecular structure of that natural steroid should be very similar to that of the synthetic antialdosterone canrenone. The difference seems to be the presence of two oxygenated groups at C_6 and C_7, in place of a double bond. In our opinion, such a natural compound was never detected in mammalias.

49. Estimation of a natural spirolactone derivative in normal and hypertensive man by gas–liquid chromatography, M. PALEM-VLIERS and P. GENARD, University of Liege (Belgium), Hôpital de Bavière, Departement de Clinique et de Pathologie Médicales (Professeur H. Van Cauwenberge), Belgium

G.L.C. is certainly the most appropriate method to estimate very small quantities of compounds non labelled with radioactive atoms. Recently, the heptafluorobutyration of steroids resulted in an appreciable increase of sensibility. Very small amounts (≃1 pg) can be detected. Using this method we have estimated a new natural spirolactonic steroid in the urine of normal and hypertensive man. The steroid is extracted from the urine with dichloromethane and washed with NaOH 0.1 N. The residue is partitionated between benzene–water. The water phase is extracted. After periodic oxydation and heptafluorobutyration the residue becomes detectable by G.L.C. A prudent and preliminary estimation of that spirolactone shows that its level is higher in 20% of hypertensive patients than in normal subjects.

50. Effects of spironolactone on the plasma binding and unbound levels of testosterone and oestradiol in healthy men, C. E. HORTH, PATRICIA J. LOBO, J. R. SHELTON, M. J. ASBURY, JOAN M. CLARKE and G. R. VENNING, Division of Scientific Affairs, G. D. Searle & Co. Ltd., High Wycombe, Bucks, U.K.

Earlier investigations (Pentikäinen *et al.*, 1974; Stripp *et al.*, 1975; Loriaux *et al.*, 1976) into the endocrine effects of treating healthy men for 5–14 days with spironolactone (S) showed no significant changes in plasma oestradiol (E_2) and unremarkable changes in plasma testosterone (T). In order to learn more about the kinetics of T and E_2, a study which extended the duration of treatment with S and which included assessments of the plasma binding, the total levels and the unbound concentrations of T and E_2 by means of equilibrium dialysis (ED) and radioimmunoassay (RIA) was designed. Four healthy, male volunteers aged 26–35 years received S, 200 mg day^{-1}, for 21 days. Venous blood samples were collected on various days prior to, during and following treatment. Although the total plasma concentrations of T and E_2 showed no significant changes during treatment, plasma unbound T was slightly but significantly increased ($P < 0.05$). This was not due to a reduction in T binding by SBG which was not significantly altered during treatment. The within subject correlation ($r = 0.42$) between unbound T and unbound E_2 was significant ($P < 0.05$) in the absence of an association between total T and E_2 levels. During treatment, a slight