same *in vitro* as *in vivo*, suggesting that no movement of binding protein(s) from cytoplasm to nucleus had occurred after the aldosterone injection.

The possibility that inactivation of kidney protein(s) might be prevented by aldosterone was investigated by the inclusion of [^3H]aldosterone in the homogenization medium. It was found under these conditions that the amount of aldosterone bound was increased three- to five-fold for the cytosol fractions and up to twofold for the nuclear fraction. However, this binding was inhibited by oestradiol-17 β , unlike the binding occurring after normal isolation. This suggests that, in addition to aldosterone-specific binding protein, kidney also contains an unstable and relatively non-specific aldosterone-binding protein. The amount of the aldosterone-specific component was not significantly increased by homogenization in the presence of aldosterone.

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Steroid Assays in Human Pregnancy Blood

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Little information is available about steroid concentrations in the human foeto-placental unit throughout gestation. This communication describes a method developed to provide such information.

Blood plasma was obtained at various times of gestation from maternal peripheral and uterine veins and umbilical artery and vein. An ethanolic solution containing radioactive tracers of cholesterol, progesterone, pregnenolone, androstenedione, 17 α -hydroxyprogesterone and testosterone was added to the plasma sample (10 ml). The plasma was extracted four times with 15 ml of A.R. ethanol and the combined dried extracts were then applied to a washed paper strip (Whatman 3MM) for chromatography. This chromatogram was developed with *n*-heptane-70% methanol (Yannone, 1968), a system that separates cholesterol, progesterone, pregnenolone and androstenedione into discrete zones but does not resolve 17 α -hydroxyprogesterone and testosterone. After a scanning for radioactivity

(Nuclear-Chicago Actigraph III) all radioactive zones were eluted from the chromatograms. The cholesterol zone was rechromatographed in a hexane (b.p. 67–70°C) adsorption system and then all zones were acetylated with acetic anhydride-pyridine (1:1, v/v). Cholesterol acetate was chromatographed again in the adsorption system; progesterone and androstenedione were rechromatographed in the *n*-heptane-70% methanol system and pregnenolone acetate was developed with hexane (b.p. 67–70°C)-90% methanol.

After elution all of these compounds were ready for quantitative determination. Progesterone 17 α -acetate and testosterone acetate were separated by paper chromatography in hexane (b.p. 60–80°C)-60% methanol. Testosterone acetate was then ready for quantitative determination. Progesterone 17 α -acetate was hydrolysed with sodium hydrogen carbonate (3.8 mM in 50% methanol) and the free steroid was chromatographed in Bush system LT11/A85 (Bush, 1961) before quantitative determination. Recoveries of compounds up to the quantitative step were all in the range 44–65%.

Cholesterol and progesterone were determined quantitatively by g.l.c. (Pye series 104 model 4 gas chromatograph with flame ionization detection) by using the peak-area measurement technique, after chromatography at 230°C in a 7 ft column of 3% XE 60. The extracts were sufficiently clean to allow quantitative determination of these compounds without interference from early eluted material. Proof of identity is based on the results of paper chromatography and g.l.c. on columns containing 3% XE 60, 3% QF 1, 3% SE 30 and 3.5% OV 17. Quantitative determination of the other four steroids is the subject of continuing research.

By using this method it was possible to obtain purified extracts of all six compounds from individual plasma samples. There is good correlation between the results obtained for progesterone by using this method and similar results published by workers who used different methods (Fuchs, Fuchs & Short, 1963; Yannone, McCurdy & Goldfien, 1968).

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