

binding" of high affinity with a $K_d(4^\circ\text{C})$ of $3.3 \times 10^{-10}\text{M}$.

It is concluded that in the experimental conditions used (intact animals), the formation of nuclear $[^3\text{H}]$ -estradiol complexes is temperature dependent and can occur either through an intermediate cytosol complex or by the "two-step" mechanism being carried out inside the nucleus which the $[^3\text{H}]$ -estradiol can reach by direct diffusion.

3. The binding of 18-hydroxydeoxycorticosterone (18-OH-DOC) of 18-hydroxyprogesterone (18-OH-progesterone) of a new urinary 18-hydroxy-steroid and a set of fluorinated steroids to mineralocorticoid and glucocorticoid receptors in the rat kidney, M. PALEM-VLIERS, P. GENARD and H. VAN CAUWENBERGE, Department de Clinique et de Pathologie Médicales (Professeur H. VAN CAUWENBERGE), Hôpital de Bavière, Université de Liège, Liege, Belgium

Cytosol prepared from the rat kidney slices of adrenalectomized male rats was incubated 30 min at 25°C with $2 \times 10^{-9}\text{M}$ $[^3\text{H}]$ -aldosterone, $2 \times 10^{-8}\text{M}$ dexamethasone, and increasing concentrations of unlabeled aldosterone, 18-OH-DOC, 18-OH-progesterone and a new urinary 18-hydroxy-steroid (compound x). $2 \times 10^{-8}\text{M}$ dexamethasone included in all flasks were required to prevent $[^3\text{H}]$ -aldosterone binding to glucocorticoid sites. Bound and free ^3H steroid were separated with G_{50} Sephadex column. 1.5×10^{-14} mol of $[^3\text{H}]$ -aldosterone were bound per mg of protein, this value was taken as control (100%). The apparent K_{Diss} for the mineralocorticoid receptors were $1.4 \times 10^{-7}\text{M}$ for 18-OH-DOC, $9 \times 10^{-7}\text{M}$ for 18-OH-progesterone. $2 \times 10^{-5}\text{M}$ cp x were able to compete with aldosterone for the cytosolic receptor. The apparent K_{Diss} of cp x for the mineralocorticoid receptor was $4 \times 10^{-6}\text{M}$. Our preliminary results have shown that cp x had a weak affinity for mineralocorticoid receptors sites; this affinity was less than that obtained with 18-OH-progesterone. cp x has a affinity for kidney glucocorticoid binding sites labelled by $[^3\text{H}]$ -dexamethasone. The affinity of some fluorinated steroids for the mineralo- and gluco-corticoid was also estimated.

4. Binding proteins for androgens and estadiol in rat perineal and skeletal muscles, ROLAND R. TREMBLAY, JEAN Y. DUBE and R. LESAGE. Department of Endocrinology, Laval University Hospital Center, Quebec, Canada

The presence of specific binding proteins for androgens and estrogens has been previously demonstrated in target tissues; however, until recently, there has been perplexity as to whether muscles would contain such highly specific binding proteins or receptors. The aim of our work was therefore to study some of the characteristics of androgens (testosterone (T) and dihydrotestosterone (DHT)) and estradiol- 17β (E_2) binding proteins in rat levator ani/bulbocavernosus muscle complex (LA/BC) and in thigh muscle (TM). Specific *in vitro* binding of T, DHT and E_2 was demonstrated in the cytosol (30,000 g supernatant) of LA/BC and TM by gel filtration through Sephadex G-25 columns. Animals were castrated 24 h prior to the experiment. In LA/BC cytosol, T, DHT and E_2 were bound with high affinity ($K_a = 1.9, 0.5$ and $0.3 \times 10^9\text{M}^{-1}$ respectively). In TM cytosol, T and E_2 had similar affinities ($K_a = 1.1$ and $2.3 \times 10^9\text{M}^{-1}$ respectively), whereas DHT had a lower affinity for its receptor ($K_a = 5.0 \times 10^7\text{M}^{-1}$). The number of binding sites for T, DHT and E_2 in LA/BC cytosol was respectively 7.5, 14.5 and 12.0 fmol/mg prot., while it was significantly lower, 1.8, 5.3 and 4.2 fmol/mg prot. in thigh

muscles. Moreover, competition experiments strongly suggested the conclusion that the binding of the 3 steroids in these sites was due to different proteins. A fundamental difference is therefore demonstrated between the muscles where T, as opposed to DHT in the prostate, is the steroid bound with high affinity to the cytosol receptor.

5. Response of the immature rat to androgen and estrogen following treatment on day one of life with estrogen, testosterone or an estrogen antagonist, LEONARD J. LERNER and ADRIANA VITALE, Lepetit Research Labs., Milan, Italy

Administration of androgen or estrogen to the newborn rat has been shown to profoundly influence the endocrine system and sexual behaviour of the matured animal. It was of interest to determine if alteration of the hormonal environment in the newborn animal could alter its responsiveness to hormones at later stages in its development. Newborn male and female rats were administered single subcutaneous injections of estradiol benzoate (EB) (10 μg), testosterone propionate (TP) (0.5 mg) or the antiestrogen MER-25 (ethamoxytriphetol) (2 mg) within 24 h after birth. At 21 days of age the females were subcutaneously administered EB at daily doses of 0.1 or 1 μg , or TP at daily doses of 1 or 5 mg for 3 days and on the following day, body, uterine and ovarian weights were determined. At 21 days of age the males were subcutaneously administered EB at daily doses of 1 or 10 μg , or TP at daily doses of 1 or 5 mg for 7 days and on the following day, body, testis, epididymis, seminal vesicle and ventral prostate weights were determined. Body weights of the rats were unaltered by any of the treatments. EB or TP on day 1 of life reduced ovarian weight on day 24 by 50% regardless of subsequent treatment. Early treatment with EB reduced the later response of the uterus to EB or TP. Early treatment with TP reduced later uterine response to TP only. Early treatment with MER-25 did not alter uterine response to EB or TP. Testis weight was significantly decreased by administration of EB or TP regardless of type of treatment at birth, however, early treatment with TP magnified the reduction of the size of this organ. The weights of the epididymes, seminal vesicles and ventral prostates and their responses to the steroids were not altered by treatment of the newborn rat with any of the compounds. This study indicates that sex steroid treatment of the newborn rat can alter the response of the prepuberal animal to EB or TP at some hormone target tissues.

6. Transformation of glucocorticoid receptor complex from rat thymocytes and its subsequent uptake on chromatin in a cell-free system, PETER A. ANDREASEN, Institute of Experimental Hormone Research, Norre Allé 71, 2100 Copenhagen O, Denmark

The uptake of glucocorticoid receptor complex from rat thymocytes on isolated chromatin from the same tissue has been studied. A thymocyte 100,000 g supernatant was prepared and made 40% with respect to glycerol. Tritiated glucocorticoid receptor complex was formed by incubation of $[^3\text{H}]$ -triamcinolone acetonide or $[^3\text{H}]$ -dexamethasone with the supernatant at -5°C . When the supernatant was incubated with chromatin at 4°C , an uptake of complex on the chromatin was found. A rapid uptake was seen after incubation of the diluted supernatant at 4°C prior to the addition of the chromatin, whereas a slow uptake was seen without preincubation. This indicated a transformation from one form of the complex to another during the preincubation. However, the total uptake was not changed by preincubation. This