14. *In vitro* characterization and assay of the progestin receptor in the rat pituitary and hypothalamus, M. MOGUILEWSKY and J. P. RAYNAUD, Centre de Recherches, Roussel-Uclaf, 93230 Romainville, France

The following properties of the progestin receptor were studied in the hypothalamus, pituitary and uterus of the estrogen-primed immature 25-day-old female rat using the highly potent progestin R 5020 (17,21-dimethyl-19-nor-4,9-pregnadiene-3,20-dione) which does not bind to CBG (corticosteroid binding globulin). (a) *Sedimentation pattern*: in sucrose density gradients, the R 5020-progestin receptor complex sedimented with a coefficient of about 6*S*. (b) *Binding parameters*: R 5020 bound to the progestin receptor with an intrinsic dissociation constant (K_D) of about $5 \times 10\,M^{-1}$ as measured by a Dextran-coated charcoal adsorption technique. (c) *Kinetic constants*: the dissociation rate (k_{-1}) of the R 5020–receptor complex was $1 \times 10^{-2}\,min^{-1}$ at 0°C and 6 times slower than that of progesterone. (d) *Specificity*: R 5020 binding to the progestin receptor was hormone-specific; progesterone, norprogesterone and norgestrel competed markedly with R 5020, whereas neither estradiol, testosterone, nor dexamethasone had any effect. Following characterization of the receptor, progestin binding sites were assayed *in vitro* by an exchange technique. The concentration of binding sites/g tissue in the hypothalamus, pituitary and uterus of the estrogen-primed rat was in the ratio 1:4:30. Progestin receptor level in tissue that contain estrogen receptor increased with estrogen (primer) dose in both intact and castrated immature rats, was maximum 40 h after priming and depended upon the estrogenic activity of the primer.

15. Progesterone receptors of the anterior pituitary gland[1], W. W. LEAVITT, R. W. EVANS, L. J. SHOLITON and J. O. JOHNSTON, Worcester Foundation for Experimental Biology, Shrewsbury, MA 01545, VA Hospital, Cincinnati, OH, Merrell National Laboratories, Cincinnati, OH, U.S.A.

Recently, a specific progesterone receptor (Rp) was identified in the cytosol fraction of hamster pituitary and hypothalamus[2]. The present study was done to determine if an Rp is present in rat pituitary. Cytosol was labelled with [^{3}H]-progesterone (^{3}HP) or [^{3}H]-R5020 (^{3}HR)[3] and subjected to sucrose–glycerol density-gradient centrifugation[2]. Serum progesterone was measured for correlation with Rp levels. Two ^{3}HP-binding peaks (4*S* + 6*S*) were evident in uterine and pituitary cytosols. The 4*S* peak was eliminated by competition with unlabeled cortisol leaving a single 6*S* peak (Rp). Estradiol (E) priming of the male or female rat increased Rp levels in pituitary cytosol as demonstrated using ^{3}HP and ^{3}HR, and pituitary Rp bound ^{3}HR with higher affinity than ^{3}HP. Following adrenalectomy of gonadectomized rats, Rp levels were increased in pituitary cytosol of both the E-primed and unprimed groups. An inverse relationship was established between serum progesterone and Rp levels in the uterus and pituitary showing that stress-induced adrenal progesterone secretion significantly influences Rp levels in the rat. These results demonstrate an estrogen-inducible Rp in rat pituitary with properties similar to those of the uterine Rp.

References

1. Supported by NSF grant PCM 75-16135, VA grant MRIS 7877 and Merrell National Labs.
2. Leavitt W. W. *et al.*: *Annls N.Y. Acad. Sci.*, **286** (1977) 210.
3. Supplied by Dr. J.-P. Raynaud, UCLAF.

16. Feedback action of progesterone and oestrogen on LH responses after administration of GnRH in cycling gilts, D. F. M. VAN DE WIEL*, E. A. VAN DE BOEZEM†, L. P. B. M. VERGROESEN‡ and F. P. W. DE VRIES‡, Research Institute for Animal Husbandry "Schoonoord", Zeist*; Agricultural University, Dept. of Animal Production, Wageningen†; Agricultural School, Dordrecht‡, The Netherlands

It was found by Foxcroft *et al.* (1975) that the LH response after GnRH administration in the immature female pig is inhibited by pretreatment of the animal with oestradiol-17β. These authors discussed that this finding seemed somewhat contradictory to the known stimulating effect of oestrogen on plasma LH levels in mature females of other species. Therefore we have investigated the LH responses after GnRH injection in mature female pigs during all stages of the oestrous cycle. Five cycling gilts, 6–8 months old, were anaesthesized and permanent indwelling catheters were introduced through the ear vein. Each animal was injected intravenously with 100 μg GnRH every two days during one cycle. Blood samples were taken at −15, 0, 10, 20, 30, 40, 60, 90, 120 and 240 min after injection, and plasma levels of LH, progesterone and oestrogen were determined by radioimmunoassay. It was found that the LH responses show gradual increases and decreases during the oestrous cycles, closely paralleling the variations in plasma progesterone levels. From these results it is difficult to conclude if this is due to a stimulatory effect of progesterone or an inhibitory effect of oestrogen. To elucidate this question, further work with ovariectomized steroid-implanted animals is now in progress.

Reference

1. G. R. Foxcroft, D. K. Pomerantz and A. V. Nalbandov: *Endocrinology* **96** (1975) 551–557.

17. Effect of a novel anti-fertility compound on plasma prolactin and gonadotrophin levels in the rat, LEONARD J. LERNER and IRIT GIL-AD, Department of Endocrinology, Lepetit Research Laboratories, 20158 Milan, Italy, First Chair Department of Pharmacology, University of Milan, 20129 Milan, Italy

Reciprocal control of physiological functions by the central nervous system and the gonads involves not only steroid and protein hormones but also catecholamines, cyclic nucleotides, prostaglandins and other endogenous substances. Modification of the physiological state by drugs or pathological conditions probably would be reflected in changes in hormonal levels. The compound DL 204 IT or L-11204, (2-(3-ethoxyphenyl)-5,6-dihydro-s-triazole [5,1-a] isoquinoline), has been shown to block HCG-induced hypertrophy and increased 5-ene-3β-hydroxysteroid dehydrogenase activity in the adult rat ovary. DL 204 IT also blocks PMS-induced superovulation in the rat; accelerates oviductal ova transportation in the rat, hamster and rabbit; terminates pregnancy in all species studies; inhibits prostaglandin catabolism in a number of tissues; increases prostaglandin synthesis in the rat ovary; and increases histamine and histaminase activity in selected rat tissues. In the present study single injections of 100 mg/Kg in a sesame oil vehicle were administered subcutaneously to groups of pregnant rats on days 2, 6, 9, 12, 14, 16 or 19 of gestation and blood plasmas were taken 48 h post-injection for RIA of prolactin, LH and FSH. Vehicle-treated animals had elevated PRL levels on days 1 and 2 of gestation and on the day of parturition. DL 204 IT administered on pregnancy days 2 or 6 increased PRL by 270 and 200% respectively but treatment after the first third of pregnancy failed to alter PRL levels. In contrast LH levels were unchanged by DL 204 IT at any time of pregnancy and FSH was significantly decreased (35%) only when injections were made on day 9 of gestation. This