

Table II. Effect of adrenalectomy on the activity of kidney and liver arginase

Experimental conditions	No. of animals	Arginase activity-units (M \pm SD)	Difference (%)	Significance
Kidney				
Controls (I)	7	9.86 \pm 1.56	—	—
Adrenalectomy (control II)	7	11.85 \pm 1.28	+ 22.46	> 0.05
Adrenalectomy + hydrocortisone 10 mg/100 g body wt.	7	26.33 \pm 2.52	+ 163.86	< 0.001
Liver				
Controls (I)	7	308.7 \pm 41.4	—	—
Adrenalectomy (control II)	7	206.3 \pm 85.0	— 33.0	< 0.01
Adrenalectomy + hydrocortisone 10 mg/100 g body wt.	7	375.1 \pm 48.0	+ 21.59 (Con. I) + 81.70 (Con. II)	> 0.05 < 0.01

adrenalectomy (in contrast to hepatic arginase) can be used as the material basis for the idea that arginase activity in the kidney and in the liver is regulated in a different way. The finding that arginase in the kidneys remained unchanged after adrenalectomy introduces the question whether corticosteroids have any direct influence at all on the regulation of the activity of this enzyme in the kidney. The increase in arginase activity under the influence of exogenous hydrocortisone may be secondary in nature, namely it may be in close association with the increased excretion of the products of protein catabolism.

Résumé. On a constaté que l'arginase des reins est plus sensible que l'arginase du foie à l'hydrocortisone injecté. Au contraire, l'adrénalectomie provoque la réduction de l'activité arginasique du foie mais ne modifie pas le niveau de l'arginase rénale.

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Inhibition of Ovarian Compensatory Hypertrophy by Water Deprivation¹

The establishment of an anoestrous condition in the rat by prolonged water depletion² suggests that this procedure induces inhibition of estrogenic secretion by the ovary in spite of the observed increase both in production and release of gonadotrophin³.

It is known that hemicastration in rats produces ovarian hypertrophy as a result of increased gonadotrophic activity⁴ and that water deprivation causes no changes in ovarian weight⁵. We produced water deprivation in rats to study its inhibitory action on ovarian compensatory hypertrophy.

The experiments were carried out in 20 female adult albino rats of our stock weighing between 105 and 233 g and having had at least 3 regular 4–5-day estral cycles. The animals were kept on a 14-h light and 10-h dark schedule. The rats were randomly divided into 2 groups: 1. control water deprived (CWD) and 2. hemicastrated and then water deprived (HWD).

Hemicastration was accomplished by the lumbar route under pentobarbital anesthesia (33 mg/kg body wt.). The excized ovaries were used to control the weight of the remaining ovary in each animal. In a previous experiment bilateral ovariectomy was performed in 26 rats, and both ovaries' weights were compared by the Student's test for paired data. The mean difference was 0.77 mg \pm 0.98 SE ($t = 0.78$ NS). 48 h after hemicastration both groups were deprived of water; a standard dry diet for rats (13% water content) was given ad libitum. Vaginal smears and body weight were both controlled daily. The rats were killed under pentobarbital anesthesia when their weight loss had reached 40% of the initial body weight; i.e. after

an average of 12.3 \pm 0.3 and 11.5 \pm 0.4 days in the (CWD) and (HWD) groups respectively.

The ovaries were carefully dissected and weighed on a Monopan balance (0.05 mg precision). They were fixed in 10% formalin for routine histologic techniques and examination.

The vaginal smear showed a continuous di-oestrous during the whole water deprivation period in both groups.

Histologic studies showed that in the ovaries of the water deprived rats there is a decrease in the number of large follicles with an increase in atresia. No changes were observed in the number of corpora lutea. Thus, in the ovaries of the water deprived animals there is an increase in the relation corpora lutea-normal follicles. A decrease in the ovarian stroma was also observed. 2 types of abnormalities were observed in the interstitial cells of the ovaries. Some cells were sphere-shaped and of a larger than normal size, their cytoplasm filled with vacuoles (xanthomatous appearance). These sphere-shaped cells

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³ K. KOVÁCS, A. JACOBVITS, M. DAVID, H. HORVATH, D. BACH-RACH and B. KÖRPASSY, *Endokrinologie* 32, 281 (1955).

⁴ C. G. HELLER, C. J. HELLER and E. L. SEVRINGHAUS, *Endocrinology* 30, 309 (1942).

Weight of ovaries

Group	No. of animals	Evolution (days)	Mean (mg/100 g body weight)	Standard error
Control	10		19.52	± 2.45
HWD	10	10-14	18.86	± 2.21
CWD*	10	10-14	20.80	± 3.32

* The half weight of both ovaries was taken into account for each animal.

were more scarce in the CWD than in normal animals and distributed either in small and medium nodes or sparsely in the ovarian stroma. The other abnormalities were shrunk spindle-shaped cells showing a greater than normal nuclear cytoplasm relationship. A gross accumulation of granulated inclusions stained with PAS, Sudan Black B and Long Ziehl-Neelsen techniques (like lipofuscin pigment) appeared to be contained, both intra and extracellularly, at least in some areas of the ovaries.

In the hemicastrated animals no significant weight change was found in the remaining ovary as compared with both their own control and those of the control water deprived animals. (Table). That is, water depletion was shown to inhibit completely the ovarian compensatory hypertrophy in spite of the reported increase in gonadotrophin release. We assume this to be related to the histologic changes that dehydration produces in the ovaries.

Resumen. En ratas hemicastradas deshidratadas no se observa la hipertrofia compensadora del ovario remanente. Se sugiere que ello pueda estar relacionado con los cambios histológicos del ovario deshidratado.

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Plasma Prolactin Levels in Fetal Sheep

Radioimmunoassays for ovine prolactin have been developed in recent years¹⁻³ permitting the measurement of the levels of prolactin in the plasma of sheep. In this paper we report the development of a double antibody radioimmunoassay for ovine prolactin and its application in determining the prolactin levels in the plasma of fetal sheep.

Methods. The development of the rabbit antiovine prolactin serum used in the radioimmunoassay has been previously described by us⁴. Ovine prolactin standard (L3458B, 35 IU/mg, the kind gift of Dr. C.H.Li) was iodinated with ¹²⁵I by the method of GREENWOOD et al.⁵. Because the labeling procedure caused some damage to the prolactin it was passed through a Sephadex G-100 superfine column (0.9 × 27 cm), equilibrated and eluted with an 0.08M barbital-acetate buffer, pH 8.6, before being used in the assay. Figure 1 shows a representative elution profile, whose individual tubes were counted on a partially-shielded gamma-sensitive scintillation detector coupled with a scaler. The material in the major peak (fractions 28-30) was diluted with a diluent made by adding bovine serum albumin (Fraction V) to the barbital-acetate buffer to a concentration of 0.5%. 200 µl of the diluted material contained 4000-5000 cpm when counted in a Nuclear Chicago Model 4216 Well Gamma Counter. For the assay, 200 µl of the ¹²⁵I-labeled ovine prolactin was added to glass culture tubes (13 × 100 mm) to which was added 200 µl of the rabbit antiovine prolactin serum diluted 1:18,000 with diluent. Data for the standard curve were obtained by adding 100 µl of ovine prolactin solutions in concentrations ranging from 0 to 50 ng/ml in diluent. Plasma was also assayed in 100 µl aliquots. Each concentration of standard and each sample was run in triplicate. The tubes were incubated at 4°C for 5 days at which time 100 µl of 1:10 dilution of goat anti-rabbit γ-globulin serum and 100 µl of a 1:20 dilution of normal rabbit serum were added to each tube. The tubes were incubated for another 6 to 16 h, centrifuged, the supernatants aspirated off, and the precipitates counted in a Nuclear

Chicago Model 4216 Well Gamma Counter. The results are expressed as a percentage of the number of counts in the precipitate of the zero prolactin tubes (referred to as percentage of zero binding). Figure 2 shows a representative standard curve and the parallel curves obtained by diluting two samples of wether plasma.

Blood samples were obtained from 11 white-faced range sheep fetuses between 60 and 138 days of fetal age. The fetuses were taken while the ewes were under pentobarbital anesthesia, their hearts removed quickly for other experiments, and the blood sample obtained, in a heparinized syringe, from the aorta.

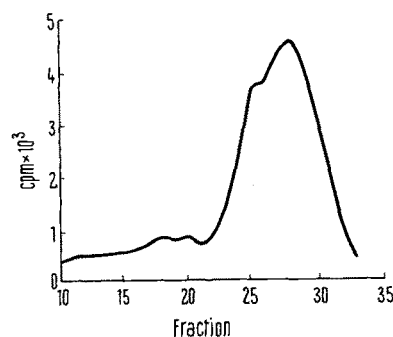


Fig. 1. Elution profile of ¹²⁵I-labeled ovine prolactin from a Sephadex G-100 superfine column (0.9 × 27 cm). Fraction volume approximately 0.27 ml; 2.0 ml of eluate was collected before the fractions.

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