Target Organs of ^{131}I-Chorionic Gonadotrophine in Immature Female Rat

By means of biological methods it has been ascertained that target organs of chorionic gonadotrophine (HCG) are ovaries, pituitary and suprarenals (for review see LAURITZEN[1]). Limitations of biological experiments did not allow us to solve the problem whether some other target organs exist to which the HCG has a specific relationship in addition to those already known. Experiments using radioactively labeled hormones appear to be an extraordinarily suitable tool for this purpose. We were principally interested in the question of localization of the specific binding sites of the HCG. We used comparatively small doses, with an action just on the border of biological effectiveness, as we are of the opinion that under these conditions an eventual difference in the binding of HCG on the individual organs may manifest itself better than with application of high doses, which overfload the organism with radioactive material and make a finer differentiation according to individual organs either difficult or impossible (SEKI[2]).

The experiment was performed on a total of 33 infantile Wistar female rats 48.5 g of weight in average. HCG (Organon) with a biological activity of 2500 IU per mg was used. The iodation of the hormone was made according to GREENWOOD et al.[3]. Resulting average specific activity was 150 μCi/μg protein. The biological activity of the hormone in protein fraction was determined by immunological method of HORSKÝ et al.[4]. After the determination of the amount of IU HCG per 1 ml of the solution, the active part of the fractions was diluted to contain 1 IU per 0.5 ml. This amount was injected i.v. once to each experimental animal. The animals were killed by decapitation in ether narcosis 20, 60, 120 min after the hormone application. Ovaries, 1 corner of the uterus, suprarenal, a piece of musculus gastrocnaemius, cerebral cortex, whole hypothalamus, median eminence, pituitary and thyreoidea were dissected and weighed. The radioactivity was measured by means of Tesla NZG 319 apparatus with spectrometric sonde. The sensitivity of the apparatus was set so that as the relative frequency of impulses per min was always of a value of 10,000 cpm. The average value of cpm of the total amount applied was 2222.412. The uptake of the ^{131}I-HCG was expressed as a) the total applied radioactivity taken up by 100 mg of the tissue, b) the ratio between this percentual value in the investigated and reference tissue (skeletal muscle or cerebral cortex) and c) the ratio of cpm per 100 mg of investigated tissue to cpm per 100 mg of reference tissue.

The results of uptake of ^{131}I-HCG by the organs examined are summarized in the Tables I and II. The interval of 120 min between the hormone injection and

[1] CH. LAURITZEN, Gynäk. Rdsch. *3*, 1 (1966).
[2] M. SEKI, J. Jap. obstet. gynaec. Soc. *10*, 8 (1963).
[3] F. C. GREENWOOD, W. M. HUNTER and J. S. GLOVER, Biochem. J. *89*, 114 (1963).
[4] J. HORSKÝ, V. JOUJA and Z. KRABEC, Allergie Asthma *13*, 120 (1967).
[5] D. M. DE KRETSER, K. J. CATT, H. G. BURGER and G. C. SMITH, J. Endocr. *43*, 105 (1969).

Table I. Radioactivity in tissues of immature female rats 120 min after i.v. injection of 1 IU ^{131}I-HCG

	Radioactivity of the total dose applied/100 mg tissue (%) (mean ± S.E.)	Radioactivity of the total dose applied/100 mg tissue (%) Radioactivity of the total dose applied/100 mg striated muscle (%) (mean ± S.E.)	cpm/100 mg measured organ cpm/100 mg striated muscle (mean ± S.E.)
Ovaries	1.23 ± 0.7	5.22 ± 1.50[a]	7.58 ± 2.85[a]
Pituitary	0.42 ± 0.18	2.92 ± 1.15	3.38 ± 1.19
Suprarenal	0.32 ± 0.14	1.95 ± 0.37[a]	3.54 ± 1.37
Uterus	0.33 ± 0.11	3.36 ± 0.98[a]	3.21 ± 0.98[a]
Striated muscle	0.14 ± 0.04	1.00	1.00
Thyreoidea	67.42 ± 19.08	2743.30 ± 493.09[a]	1525.29 ± 405.52[a]

[a] The tissue/muscle ratio is different from 1.00 at p 0.05. The hypothesis was evaluated by the t-test that tissue/muscle ratio has an average value of 1. This hypothesis was rejected if the 95% confidence limits for the ratio's mean value did not involve the value of 1.

Table II. Radioactivity in brain and pituitary of immature female rats 120 min after i.v. injection of 1 IU ^{131}I-HCG

	Radioactivity of the total dose applied/100 mg tissue (%) (mean ± S.E.)	Radioactivity of the total dose applied/100 mg tissue (%) Radioactivity of the total dose applied/100 mg cerebral cortex (%) (mean ± S.E.)	cpm/100 mg measured organ cpm/100 mg cerebral cortex (mean ± S.E.)
Pituitary	0.42 ± 0.18	7.67 ± 1.48[a]	3.38 ± 1.19[a]
Hypothalamus	0.02 ± 0.0097	1.31 ± 0.35	0.67 ± 0.13
Median eminence	0.06 ± 0.03	1.65 ± 0.37	1.50 ± 0.37
Cerebral cortex	0.04 ± 0.01		1.00

[a] The tissue/cerebral cortex ratio is different from 1.00 at p 0.05. The hypothesis was evaluated by the t-test that tissue/cerebral cortex ratio has an average value of 1. This hypothesis was rejected if the 95% confidence limits for the ratio's mean value did not involve value of 1.

killing of the experimental animals showed most significant results. The control group of 6 infantile female rats of the same weight was injected i.v. with ^{131}I-bovine serum albumin (2 µg in 0.5 ml of phosphate buffer). Iodation was done in the same way as in the case of HCG. The value of cpm of the total amount applied was 6,000,000. The experimental design was the same as in the experimental group. No preferential incorporation was found in comparison with reference tissue, similarly to what has been shown by DE KRETSER et al.[5]. In our experiment with ^{131}I-HCG it was possible to establish a statistically significant uptake of radioactivity by the ovary, uterus and pituitary, but not by the suprarenal. It may be of interest in this connection that while LAURITZEN et al.[6] found a high concentration of HCG in the hypophysis of the human embryo by biological methods, they were unable to show its presence in the ovaries, just as TSUMUJI[7] some years ago did not find any preferential accumulation of ^{131}I-HCG in the ovaries of the rabbit.

We believe, however, that a most interesting result is demonstrated, namely a high accumulation of ^{131}I-HCG in the uterus. A direct effect of HCG on the uterus was only supposed by some authors up to now (for review see LAURITZEN[1]). This effect should be furthermore intensively studied.

Zusammenfassung. Die Einlagerung von ^{131}I-HCG und ^{131}I-Serumalbumin in einige Organe unreifer Rattenweibchen wurde gammaradiometrisch gemessen. Eine signifikant erhöhte Akkumulation der Radioaktivität nach ^{131}I-HCG-Verabreichung wurde im Ovarium, dem Uterus, der Hypophyse und der Schilddrüse festgestellt.

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[6] CH. LAURITZEN, G. DE RIDDER and M. MENTZ, Acta endocr., Copenh. *61*, Suppl. 138, 218 (1969).
[7] Y. TSUMUJI, Ber. III. Weltkongress Int. Fed. Gynäk. Geburtsh., (1961), vol. 1, p. 119.

Identification of Acrocentric Chromosomes Involved in the Formation of 'Fusion'-Metacentrics in Mice. Proposal for Nomenclature of *M. poschiavinus* Metacentrics

Cytogenetic methods have restricted value in the field of mouse genetics unless means are available to make the identification of individual chromosomes possible. Therefore, structural variations due to centric fusion of acrocentric chromosomes are of considerable interest because 'fusion'-chromosomes such as the T163H-[1], the T1Ald-[2,3], and the T1Wh-[4] metacentrics are recognizable cytogenetic markers of the respective mouse strains. Compared to these strains with a single pair of metacentric chromosomes each, the karyotype of the tobacco mouse, *M. poschiavinus*, is characterized by the presence of 7 pairs of Robertsonian 'fusion'-metacentrics[5,6].

Any further step contributing to the identification and individualization of the acrocentrics involved in translocation-fusions should be helpful in genetic studies in mice. Therefore, attempts were undertaken to identify the acrocentric constituents (chromosome arms) of the 'fusion'-metacentric of the T1Ald-translocation by crossing female AKR-mice homozygous for the T1Ald-chromosome ($2n = 38$; N.F. = 40) with the wild type tobacco mouse (*M. poschiavinus*) carrying 7 pairs of metacentrics ($2n = 26$; N.F. = 40). It can be assumed that the chromosomes of the AKR-strain as well as the acrocentrics of the laboratory mouse strains in general are homologous to the acrocentrics or to the arms of the metacentrics of the tobacco mouse. Whether or not the T1Ald-metacentric is among the 7 metacentrics of the tobacco mouse should be revealed by meiotic studies in F_1 offsprings of such crosses. However, other combinations could have been established by the fusions of the acrocentrics in both strains. F_1-animals can be expected to be heterozygous for 7 metacentrics of the tobacco mouse and for one T1Ald-metacentric. Yet, in the first case mentioned, primary spermatocytes of F_1♂ examined in diakinesis and first metaphase stages would show 6 trivalents and 6 bivalents including the bivalent built up by a homologous pair of metacentrics, and the *XY*-bivalent. In the other case more complicated figures would result.

In fact, a diploid chromosomal set (spermatogonia, bone marrow) of $2n = 32$ (N.F. = 40) with 8 metacentrics was found in the 3 F_1 males (Figure 1) and the 2 F_1 females from different litters so far studied. Rough measurements indicate that the T1Ald-chromosome

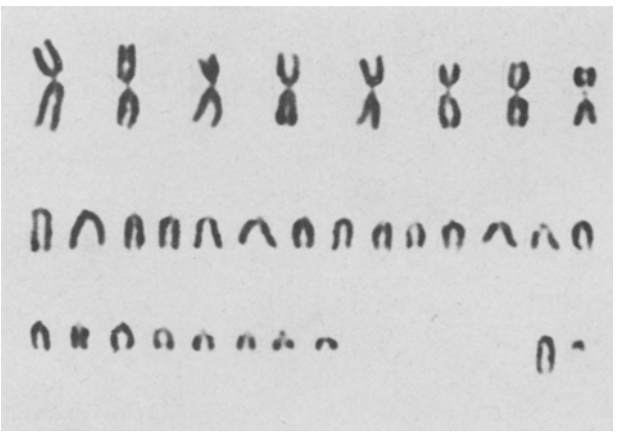

Fig. 1. Karyotype (bone marrow metaphase) of a (*M. poschiavinus* ♂ × AKR/T1Ald-T1Ald ♀) F_1♂. $2n = 32$; N.F. = 40.

[1] E. P. EVANS, M. F. LYON and M. DAGLISH, Cytogenetics *6*, 105 (1967).
[2] A. LÉONARD and GH. DEKNUDT, Experientia *22*, 715 (1966).
[3] A. LÉONARD and GH. DEKNUDT, Acta zool. pathol. antverp. *48*, 43 (1969).
[4] J. B. WHITE and J. H. TJIO, Hereditas *58*, 284 (1967).
[5] A. GROPP, U. TETTENBORN and E. VON LEHMANN, Experientia *25*, 875 (1969).
[6] A. GROPP, U. TETTENBORN and E. VON LEHMANN, Cytogenetics *9*, 9 (1970).