mated before and after Sephadex-LH 20 chromatography gave identical results[7].

The biological effects of the active immunization of male rabbits against androstenedione differ in some respects from the results of Nieschlag et al.[2], as we found a significant increase of the concentration of androstenedione and testosterone in peripheral blood, as well as Leydig's cell hyperplasia, but no significantly increased testicular weight. An explanation for this difference may be the different specificity of the antibody as the antigens were conjugated in a different position of the steroid molecule to the protein.

1 E. Nieschlag, K.-H. Usadel, U. Schwedes, H.K. Kley, K. Schöffling and H.L. Krüskemper, Endocrinology *92*, 1142 (1973).
2 E. Nieschlag, K.-H. Usadel, H.K. Kley, U. Schwedes, K. Schöffling and H.L. Krüskemper, Acta endocr. (Kbh.) *76*, 556 (1974).
3 E. Nieschlag, K.-H. Usadel, E.J. Wickings, H.K. Kley and W. Wuttke, in: Immunization with Hormones in Reproduction Research, p. 155. Ed. E. Nieschlag. North-Holland Publishing Co., Amsterdam 1975.
4 S. Nechansky, Thesis, Vet. med. University, Vienna 1979.
5 J. Vaitukaitis, J.B. Robbins, E. Nieschlag and G.T. Ross, J. clin. Endocr. *33*, 988 (1971).
6 E. Nieschlag and D.L. Loriaux, Z. klin. Chem. klin. Biochem. *10*, 164 (1972).
7 E. Möstl, Thesis, Vet. med. University, Vienna 1978.

Hemicastration-induced changes in the electrophoretic pattern of some enzymes in the brain of the skink, *Mabuya carinata*

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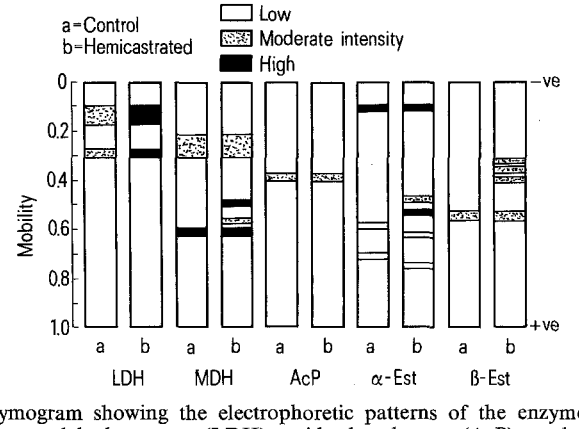
Summary. Hemicastration in the skink induces change in the electrophoretic pattern of some enzymes like LDH, MDH, acid phosphatase and esterases.

Hemicastration-induced compensatory hypertrophy of the contralateral gonad is said to be due to the removal of gonadal hormone feedback check on the hypophysis and consequent increased release and/or utilization of hypophysial gonadotrophins[2,3]. This hemicastration-induced compensatory hypertrophy is blocked by gonadal hormones like estrogen, progesterone and testosterone, indicating that the former explanation may be true[4–6]. We report here some interesting information about the electrophoretic patterns of some enzymes in the brain of the hemicastrated male skink.

Materials and methods. Sexually mature male skinks, weighing 18–25 g, collected in and around Mysore city during the month of September, were hemicastrated by surgical removal of the right testis. Sham-operated controls were also used. Each group contained 5 animals. On the 21st day of hemicastration, the animals were autopsied; the brain was dissected out and immediately homogenized in 0.1 M phosphate buffer pH 7.0 (1:2.5 w/v) using a tissue homogeniser at 5 °C. The homogenate was centrifuged at 3000 rpm at 5 °C for 1 h in an MSE refrigerated centrifuge. Protein concentration in the homogenate was estimated by the method of Lowry et al.[7]. About 150 µg of protein from this sample was layered on the gel in 40% sucrose to carry out polyacrylamide disc gel electrophoresis as described by Davis[8]. Tris-glycine buffer at pH 8.5 was used in the run at 5 °C, applying a current of 4 mA per gel. The gels were stained using suitable procedures[9,10] for acid phosphatase (AcP), α- and β-esterases (Est), lactate dehydrogenase (LDH) and malate dehydrogenase (MDH).

Results and discussion. Electrophoretic patterns for various enzymes and their mobilities are given in the figure. There is no difference in the electrophoretic patterns of LDH bands between the hemicastrated animals and sham-operated controls. But an increase in the total activity of both the bands was observed in the hemicastrated skinks. Acid phosphatase does not show any difference in the number of bands or their activity between the control and experimental groups. The electrophoretic pattern with reference to MDH and both esterases was altered, however. All 3 enzymes show new bands in addition to the bands present in the enzymes from the brains of the control animals. 2 new additional MDH-bands appeared, and 3 new additional β-esterase bands were found. The α-esterase even has 4 new cationic bands in hemicastrated animals, and 2 cationic bands found in the controls disappear.

The results indicate an overall increase in the total activity as also in the number of bands representing isozymic patterns in 3 enzymes. The oxidative enzymes have been reported to be involved in lipid metabolism and carbohydrate metabolism. Ascribing any definite function to the hydrolytic enzymes, especially esterases, in any tissue is not possible as their occurrence is ubiquitous. But it can be said



Zymogram showing the electrophoretic patterns of the enzymes lactate dehydrogenase (LDH), acid phosphatase (AcP), malate dehydrogenase (MDH), α- and β-esterases (Est) in the brain of normal and hemicastrated skink, *Mabuya carinata*.

that these may be involved in the release or utilization of the components necessary to maintain the probable physiological imbalance created by hemicastration. Hence it can be said that there is a de novo synthesis of hydrolytic and oxidative enzymes.

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2 R.A. Edgren, A.F. Parlow, D.L. Peterson and R.C. Jones, Endocrinology *76*, 97 (1965).
3 M.X. Zarrow, S.K. Sundaram and M. Stob, Proc. Soc. exp. Biol. Med. *119*, 551 (1965).
4 S.V. Goswami and B.I. Sunderaraj, Gen. comp. Endocr. *11*, 393 (1968).
5 R.E. Short, J.B. Peters, N.L. First and L.E. Casida, J. Anim. Sci. *27*, 705 (1968).
6 M. Suvarnalatha and H.B.D. Sarkar, Horm. Metab. Res. *6*, 417 (1974).
7 O.H. Lowry, N.J. Rosebrough, A.L. Farr and R.J. Randall, J. biol. Chem. *193*, 265 (1961).
8 B.J. Davis, Ann. N.Y. Acad. Sci. *121*, 404 (1964).
9 C.R. Shaw and A.L. Koen, Chromatographic and electrophoretic techniques, vol. 2, 2nd edn. John Wiley, New York 1968.
10 G.J. Brewer, An introduction to isozymic techniques. Academic Press, New York 1970.

Bilateral ovarian hypertrophy in pituitary-grafted rats[1]

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Summary. Large masses that looked like tumours were found in place of the ovaries in rats with isotransplants of pituitaries, 11 months after this procedure. They were interpreted as being due to massive bilateral cystic hyperplasia and hypertrophy of both ovaries.

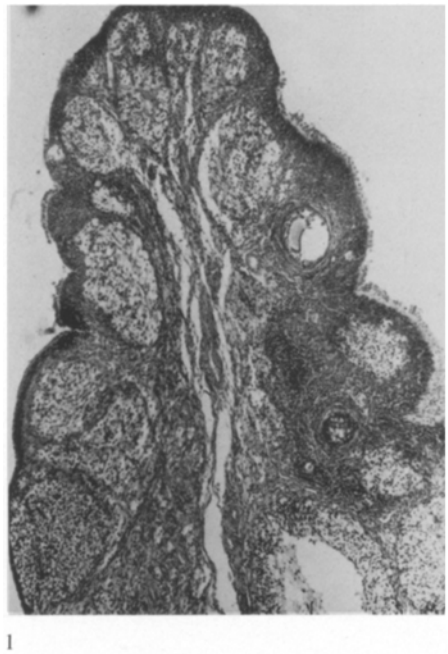
There are many experimental and clinical reports showing that prolactin excess or deficit influences ovarian function in women and in experimental animals[2-9]. As it is known that pituitary isografts, placed under the renal capsule, induce increased prolactin production in the rat[10,11], it was considered interesting to use this method to observe the effects of chronic prolactin excess on ovarian function in the rat.

Pituitary isografts were placed under the kidney capsule in 15 normal female Wistar rats, 3 months old. 1 pituitary was grafted in each kidney. Another 10 female rats from the same group were kept as controls.

Vaginal smears were performed daily during the first 2 months and afterwards during 10 consecutive days each month, during the 11-month experimental period. After the operation the normal vaginal cycle and the estrus and proestrus disappeared in the experimental animals, the vaginal smears presenting a metaestrus-like picture with residues of cornified cells, some navicular cells and many leucocytes; the latter sometimes predominated and the smears looked like diestrus.

Blood was extracted from the heart 2.5 months after the operation. Prolactin was assayed in the serum by radioimmunoassay using a rat NIAMDD prolactin kit, which was generously sent by A.F. Parlow. The serum prolactin level of the 10 control rats was 5.80 ± 0.92 ng/ml while that of the 17 pituitary-grafted ones it was 22.47 ± 5.39, the difference being significant ($p < 0.01$). But looking at the individual data it could be seen that only 7 out of 15 pituitary-grafted rats were clearly hyperprolactinemic.

The most outstanding feature in the autopsy was the bilateral presence, in place of each ovary, of a large,



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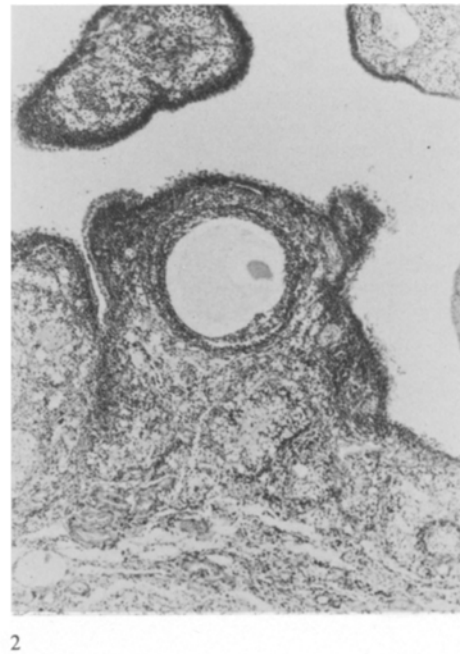

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Fig. 1. Histologic section of tumoral-looking mass showing ovarian tissue with follicles in different degrees of development. Clumps of clear cells of luteinic aspect are also seen.
Fig. 2. A section with greater magnification showing a well developed ovarian follicle and clear cells of luteinic aspect.