

microhematocrit tube and the plasma then diluted 1:400 in 0.1 N HCl. Sodium concentrations in this diluted plasma were then measured on a Zeiss PMQ II flame photometer.

Results. The number of days that *X. hellerii* survived in fresh water after CB 154 injections ranged from 1–15 days. The average survival time was 9.3 ± 2.2 days ($\bar{X} = \text{S.E.M.}$). One fish injected with CB 154 died prior to transfer to fresh water. No vehicle-injected fish died throughout the experiment.

When the CB 154-injected *P. latipinna* were transferred to fresh water they survived from 1–9 days with an average of 4.9 ± 1.0 days. One fish injected with CB 154 and 2 vehicle-injected fish died before transfer to fresh water. No vehicle-injected fish died in fresh water.

There was no significant difference apparent in the *eta* cells of the pituitary glands of the two groups of *P. latipinna* with respect to cell size, nucleus size, or the ratio cell size/nucleus size. However, the cytoplasmic granulation of the *eta* cells in the CB 154-injected fish appeared to exhibit a stronger stainability than in the vehicle-injected fish (Figure). Also, the nucleus more frequently had a folded appearance and the nucleoli were less pronounced in the CB 154-injected fish.

There were no differences between the plasma sodium concentrations in the uninjected control fish and fish that received injections of 2.5 mg CB 154 per kg body weight. These 2 groups, however, had higher plasma sodium concentrations than those groups that had injections of CB 154 in concentrations of 5 or 10 mg/kg body weight ($p < 0.05$) (see Table).

Discussion. The failure of hypophysectomized *P. latipinna* in fresh water has been shown to be due specifically to the lack of prolactin (BALL and OLIVEREAU¹⁴). Thus, it appears that CB 154 has the same effect as hypophysectomy with respect to the available circulating supply of this hormone. The average survival time of 9.3 days for *X. hellerii* is in close agreement with 8.6 days reported by SCHREIBMAN and KALLMAN¹⁵. However, the average survival time of 4.9 days for *P. latipinna* is high compared to the results of 1–2 days reported by BALL and OLIVEREAU¹⁴. Whether this is an effect of CB 154 or a different race of fish is not known.

As previously mentioned, there are a number of investigations which indicate that CB 154 inhibits the secretion of prolactin in several mammals. Further, it has been shown by PASTEELS et al.⁹ that the secretion of prolactin from

rat and human hypophyses in organ culture can be inhibited by CB 154. They thus suggest that CB 154 inhibits the exocytosis of secretory granules by direct action on the prolactin cells. This may also be the case in *P. latipinna* since these fish injected with this same compound appeared to have *eta* cells of the pituitary gland which were more granulated and appeared less active than the *eta* cells of fish injected with the vehicle alone. The *eta* cells are most likely the pituitary source of teleost prolactin (see BALL and BAKER¹⁶).

BALL and ENSOR² have proposed that the reason that hypophysectomized *P. latipinna* fail in fresh water is due to a drop in plasma sodium. This drop in plasma sodium can be specifically prevented by ovine prolactin. This more specific physiological parameter which exists in the absence of prolactin was also evident after CB 154 injections in concentrations of 5 and 10 mg/kg body weight. Injections of 2.5 mg CB 154 per kg body weight were ineffective in lowering the plasma sodium levels. Thus, in appropriate concentrations, CB 154 prevents the maintenance of a normal plasma sodium concentration which is specifically controlled by prolactin.

The action of CB 154 appears to be similar in *X. hellerii* and *P. latipinna* as in several mammals. Whether CB 154 specifically inhibits the release of prolactin but has no effect on the release of any other hormone in these two fish is unknown. If it is a specific action on prolactin release only, then CB 154 could be a most useful compound in investigations concerning prolactin physiology in other species of teleosts.

Zusammenfassung. Die Injektion von 2-Br- α -ergokryptin (CB 154) hemmte das Überleben von *Xiphophorus helleri* und *Poecilia latipinna* im Süßwasser. CB 154 senkte die Plasma-Na-Konzentration von *P. latipinna*, was als Zeichen einer Hemmung der Prolaktinsekretion interpretiert werden kann. Auch erschienen die wahrscheinlich Prolaktin sezernierenden *aeta*-Zellen der Hypophyse inaktiv nach CB 154. Es ist noch unbekannt, ob CB 154 direkt oder indirekt die Prolaktinsekretion der Hypophyse hemmt.

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Effect of CB 154 on plasma sodium concentrations in *P. latipinna* in freshwater

Treatment	Sodium concentration (mEq/l) ^a
1. No injection	159.3 \pm 1.6
2. 2.5 mg CB 154/kg body weight	166.6 \pm 11.0
3. 5.0 mg CB 154/kg body weight	127.2 \pm 9.4
4. 10 mg CB 154/kg body weight	130.7 \pm 8.5

^a Mean \pm S.E. Sodium in freshwater 3.98 mEq/l.

¹⁴ J. N. BALL and M. OLIVEREAU, C. r. Séance. Soc. Biol., Paris 259, 1443 (1964).

¹⁵ M. P. SCHREIBMAN and K. D. KALLMAN, Gen. comp. Endocr. 6, 144 (1966).

¹⁶ J. N. BALL and B. I. BAKER, in *Fish Physiology* (Eds. W. S. HOAR and D. J. RANDALL; Academic Press, New York 1969), vol. 2, p. 1.

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Neurosecretion in Ehrlich Ascites Carcinoma-Bearing Mice

The dependence of tumours of endocrine and target organs on the internal hormonal environment, as well as the production of such tumours by a hormonal imbalance, is widely recognized as important in human and animal

carcinogenesis¹. Some hormones may act as carcinogens or promote the growth of human and experimental tumours^{1–3}. The role of neurohormones also appears to be significant in this respect. It has been shown, for example,

that the level of the antidiuretic hormone is elevated in the blood plasma of tumour patients⁴⁻⁶. On the other hand, it has been shown that the neurohypophysial hormone lysine-vasopressin stimulates RNA and DNA synthesis in *in vitro* cultivated cerebral tumours^{7,8}.

In the present paper the results are reported of a karyometric investigation of the hypothalamic, vasopressin-producing centre, viz. the supraoptic nucleus, in tumour-bearing animals. Adult male mice of the C57 Bl strain were used. The animals were inoculated i. p. each with ca. 10⁷ living Ehrlich ascites carcinoma cells. Each animal inoculated with the tumour developed a marked ascites during the second week of the experiment. The animals were killed by decapitation on the 14th day post inoculation. Simultaneously a control group of 4-month-old healthy animals of the same strain, sex and age was sacrificed. Immediately after decapitation the brains were exposed, trimmed, and the hypothalami fixed by immersion in BOVIN's fluid. 7 μ m serial sections were prepared from each brain and stained with GOMORI's chrome haematoxylin by BARGMANN's modification. For the karyometric study, the volumes of 100 nuclei of the neurosecretory cells of the supraoptic nucleus in each animal of the experimental and the control group were estimated. This was done by measuring the long and the short axis of the nucleus, the volume being calculated according to the formula: $V = \pi/6 LB^2$, where L is the long and B the short axis of the nucleus⁹. The arithmetical means obtained in the experimental and the control group were compared statistically by means of the Student's t -test.

The results of the present study are summarized in the Table. As can be seen, significantly larger nuclear volumes are observed in the neurosecretory cells of the supraoptic nucleus of the tumour-bearing animals in comparison with those found in the controls. This clearly indicates an enhanced neurosecretory activity of the supraoptic nucleus on day 14 following tumour inoculation.

The present experiment confirms the clinical studies of BERNARD-WEIL et al.^{4,5} who reported an elevated plasma

level of the antidiuretic hormone in cancer patients. Moreover, they showed that the excess hormone is of hypothalamic origin and not of tumour origin as considered possible by BERNARD-WEIL and ADAM⁶.

The significance of the enhanced secretion of vasopressin in cancer patients and in tumour-bearing animals can only be surmized. One possibility is a reduced feedback inhibitory control of adrenal corticoid hormones on the hypothalamic neurosecretory centres. The neurohypophysial hormones stimulate the adrenal cortex both directly¹⁰ and by the intermediary of the hypothalamic CRF-producing centre¹¹. On the other hand, adrenal steroid hormones appear to inhibit the neurosecretory centres since increased secretory activity in the hypothalamus is observed in cases where there is a decreased level of adrenal cortical hormones in the blood¹²⁻¹⁴. The functional state of the adrenal cortex in Ehrlich ascites carcinoma-bearing mice is now being investigated in our laboratory.

Zusammenfassung. Karyometrische Untersuchungen am Nucleus supraopticus von Mäusen des Stammes C57Bl, die Träger des Ehrlich-Ascites-Tumors sind, zeigen eine statistisch gesicherte Zunahme der Zellkernvolumina im Vergleich zu gesunden Kontrolltieren.

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Nuclear volumes (μm^3) of neurosecretory cells of the supraoptic nucleus of C57Bl mice bearing the Ehrlich ascites carcinoma and in healthy controls

Group	Experimental	Control
No. of nuclei measured	700	700
Mean (μm^3)	272.15	221.09
Standard error of the mean (S.E.)	± 4.07	± 3.75
Significance of the difference between the means	$t = 9.20 \quad P < 0.001$	

¹ E. STERN, M. R. MICKEY and R. A. GORSKI, *Ann. N.Y. Acad. Sci.* 164, 494 (1969).

² A. LACASSAGNE, *C. r. hebdomadaire. Seance. Acad. Sci., Paris* 195, 630 (1932).

³ W. A. MEISSNER, S. C. SOMMERS and G. SHERMAN, *Cancer* 10, 500 (1957).

⁴ E. BERNARD-WEIL, GILBERT-DREYFUS, M. DAVID and J. SEBAOUN, *Lancet* 11, 324 (1967).

⁵ E. BERNARD-WEIL et Y. ADAM, *Annls. Endocr.* 30, 393 (1969).

⁶ E. BERNARD-WEIL, *Annls. Endocr.* 30, 549 (1969).

⁷ E. BERNARD-WEIL, C. PIETTE et L. OLIVIER, *Path.-Biol.* 16, 445 (1968).

⁸ E. BERNARD-WEIL, C. PIETTE, L. OLIVIER et M. DAVID, *Annls. Endocr.* 30, 399 (1969).

⁹ M. PALKOVITS, *Endokrinologie* 45, 227 (1963).

¹⁰ J. G. HILTON, L. F. SCIAN, C. D. WESTERMANN and O. R. KRUSEI, *Science* 129, 971 (1959).

¹¹ G. A. HEDGE and P. G. SMELIK, *Neuroendocrinology* 4, 242 (1969).

¹² K. SREBRO and Z. SREBRO, *Folia biol. Kraków* 18, 353 (1970).

¹³ A. CORNIL, A. DE COSTER, G. COPINSCHI and J. R. H. FRANCKSON, *Acta endocr. Copenh.* 48, 163 (1965).

¹⁴ A. VIRÙ and H. ÅKKE, *Acta endocr. Copenh.* 62, 385 (1969).

Granulationes leptomeningicae bei *Gallus domesticus*

Die beim Menschen und bei Primaten regelmässig vorkommenden Granulationes und Villi arachnoideales sind auch für die Haussäugetiere¹, nicht jedoch allgemein für Säugetiere² als konstante Bildungen anzusehen. Für niedere Wirbeltiere liegen Angaben nicht vor; es sei denn, man schliesst das Fehlen arachnoidealer Granulationen aus der verbreiteten Ansicht, dass nur bei Säugern die Leptomeninx in eine Arachnoidea und eine Pia mater differen-

ziert ist. Bei niederen Wirbeltieren werden allerdings den Granulationen ähnliche Bildungen vermutet³.

Nach eigenen Untersuchungen ist die Struktur der Leptomeninx des Huhnes zwar regional unterschiedlich und weist nur abschnittsweise ein Cavum leptomeningicum auf; es lassen sich aber unabhängig davon stets 2 Blätter der Leptomeninx differenzieren. Danach ist dem Huhn konstant eine Arachnoidea zuzuschreiben. Die aus einem