

Reticular Fibres in Yoshida's Sarcoma

The histogenesis and classification of the tumour experimentally established by YOSHIDA, and called 'Yoshida's sarcoma', remains undecided^{1,2}. One of the possible reasons for this is the fact that he was unable to demonstrate the presence of reticuline by the method of Wilder.

We found no reference in the literature which might throw any light on the behaviour of this tumour as regards the production of reticuline. For this reason we decided to study the subject, using several other methods.

Portions of different areas of tumours implanted subcutaneously in the cervico-dorsal region of Wistar female rats were used. The animals were killed after 20 days. Macroscopically observable necrotic material was dis-

carded. The samples were fixed in 10% formalin. Besides the usual staining methods, we used Rio Hortega's and Wilder's methods for reticular fibres, PAS and examination with polarized light.

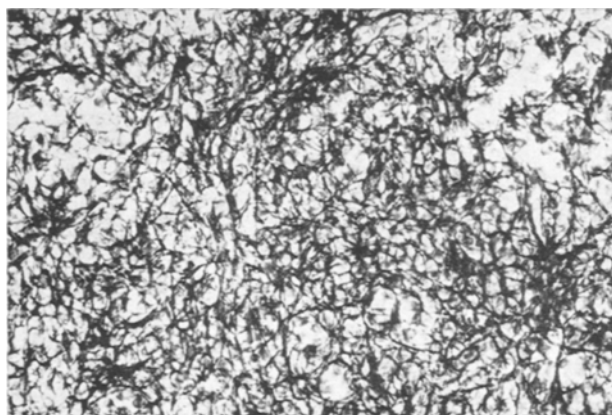
The general morphology was identical with that described by YOSHIDA^{1,2}. The sections stained by the method of Rio Hortega, however, revealed in several areas an abundance of reticular fibres. The PAS was also positive in several areas and examination with polarized light revealed the birefringence which is characteristic of fibrillar material³. These results seem to indicate the presence of reticuline in the material examined.

Two explanations may be offered to explain our results. Either the tumour suffered an alteration of its behaviour in our laboratory, or Wilder's method is not sufficiently sensitive for the detection of reticuline in this material.

Zusammenfassung. Das Yoshida-Sarkom wurde mit Hilfe von PAS, polarisiertem Licht und den Methoden von Wilder und Rio Hortega zur Darstellung retikulärer Fasern untersucht. Letztere Methode führte zum Nachweis grösserer Fasermengen im Neoplasiegewebe.

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Rio Hortega's method showing reticular fibres in the Yoshida sarcoma.

¹ T. YOSHIDA, *Gann.* 40, 1 (1949).

² T. YOSHIDA, *J. natn. Cancer Inst.* 12, 947 (1952).

³ A. G. EVERSON PEARSE, *Histochemistry, Theoretical and Applied*, 2nd Edn. (J. A. Churchill Ltd., London 1961), p. 162.

Effect of Starvation on the Concentration of Ascorbic Acid in the Pedipalp Muscle of the Scorpion *Palamnaeus bengalensis*

There are only a few reports on the nutritional and metabolic status of ascorbic acid for invertebrates. BRIGGS¹ and ROUSELL² reported that *Periplaneta americana* can synthesize ascorbic acid from hexoses. BRIGGS¹ showed that the *Musca* homogenates can synthesize ascorbic acid from a variety of hexoses including glucose. These observations indicated that invertebrate tissues can synthesize ascorbic acid. However, the factors influencing the synthesis in invertebrates have not been studied. In mammalian liver, the synthesis of ascorbic acid declines on starvation (STRIPE, COMPORTI, and DELLACORTE³). In the present investigation, the author has studied the effect of starvation on the concentration of ascorbic acid in the muscle of *Palamnaeus bengalensis*.

The scorpions were collected and fed on live cockroaches for one day. One lot was then used as normals and the other lot was allowed to starve for 12 days. The pedipalp muscles were dissected from the normal and starved scorpions and the concentration of ascorbic acid was determined according to the 2,4-dinitrophenyl-hydrazine method of ROE⁴. The intensity of colour developed was

measured in a Lumetron colorimeter, using a 515 mμ filter. The concentrations of the unknown samples were determined from a standard linear curve. The results were expressed as mg ascorbic acid/100 g wet weight of muscle.

Concentration of ascorbic acid in the pedipalp muscle of normal and starved scorpions

Animal	mg ascorbic acid/100 g wet weight	S.D.	P. 't' test
Normal	10.01 (3)	± 1.41	0.001
Starved	2.50	± 0.86	

¹ M. H. BRIGGS, *Comp. Biochem. Physiol.* 5, 241 (1962).

² G. ROUSELL, *Trans. N.Y. Acad. Sci.* 19, 17 (1957).

³ F. STRIPE, M. COMPORTI, and E. DELLACORTE, *Biochem. J.* 95, 363 (1965).

⁴ J. H. ROE, *Meth. biochem. Analysis* 1, 115 (1954).

⁵ R. C. SINHA, unpublished data.

The concentration of ascorbic acid in the muscle declines significantly on starvation (Table). It is known that on starvation the animals live on the component tissues of their own body for energy purposes. The carbohydrate store of the liver and muscle of the scorpion *Palamnaeus bengalensis* has also been shown to decline on starvation (SINHA⁶). For ascorbic acid synthesis, the animal depends on the dietary source of hexoses. Due to rapid consumption of carbohydrates during starvation, the animal fails to get a sufficient amount of hexoses. Hence, the synthesis of ascorbic acid decreases on starvation as in-

dicated by the low content of ascorbic acid in the starved scorpions.

Zusammenfassung. Bei 12 Tage hungernden Skorpionen (*Palamnaeus bengalensis*) wurde die Ascorbinsäure-Konzentration im Pedipalpen-Muskel mit 2,4-Dinitrophenyl nach ROE⁴ bestimmt und dabei eine signifikante Abnahme der Ascorbinsäure festgestellt.

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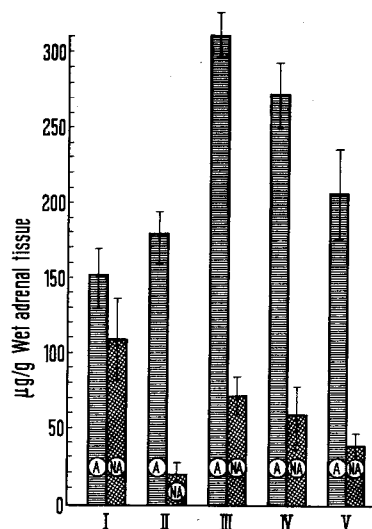
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Effect of Aldosterone, Glucagon and Growth Hormone on the Catechol Amine Content and the Evolution of Chromaffin Hyperplasia in Guinea-Pigs

The neuro-humoral control and growth of chromaffin tissue are but little known¹. There are some experiments concerning the release of catechol amines (adrenalin and noradrenalin) under the influence of hormones (insulin², cortisone and hydrocortisone³, and glucagon⁴) or some drugs (reserpine and chlorpromazine⁵) from the adrenal medulla of cats, rats and dogs. In our previous experiments^{6,7}, we noted the occurrence of chromaffin hyperplasia or pheochromocytoma after the administration of large doses of oestrogens and growth hormone in rats or guinea-pigs. The present paper studies the action of aldosterone, glucagon and growth hormone on the distribution of catechol amines and the evolution of medullary chromaffin hyperplasia induced by oestradiol administration in adult guinea-pigs.

Material and Method. The investigations were carried out in 50 adult guinea-pigs (males and females) which were divided into five experimental groups as follows: 1) 10 guinea-pigs served as controls. 2) 10 received concomitantly during about 4 months the following treatment: twice a week 300 µg α-oestradiol i.p. and 75 µg (75 γ) glucagon (Lilly) i.m., the total amount being 9500 µg oestradiol + 2400 µg glucagon for each guinea-pig. 3) 10 received simultaneously for about 4 months: twice-weekly doses 300 µg oestradiol i.p. and 16 µg d-Aldosterone (Aldocorten, CIBA) i.m., the total dose being 9500 µg oestradiol + 500 µg d-Aldosterone for each guinea-pig. 4) 10 received each week 300 µg oestradiol i.p. and 12 Evans units (EU) of growth hormone intramuscularly, the total dose being 9500 µg oestradiol + 400 EU growth hormone. 5) 10 received only twice a week 300 µg oestradiol by the intraperitoneal route, the end dose being 9500 µg oestradiol, to induce a medullo-chromaffin hyperplasia. After 4 months, all guinea-pigs were killed by light ether anaesthesia and the adrenal glands were collected: one adrenal gland for estimation of the adrenalin and noradrenalin content by an improved technique of VON EULER and LISHAJKO⁸, and the other adrenal gland in order to evaluate the intensity degree of chromaffin hyperplasia after fixation in Bouin fluid and staining with PAS-hematoxyline eosin or the chromaffin reaction after fixation in chrome salt fixatives. Recent histochemical techniques⁹ allow a good identification of adrenalin and noradrenalin-storing cells in adrenal medulla.

Results. The biochemical methods show a significant change in the adrenalin and noradrenalin content of the adrenal medulla of guinea-pigs after administration of these hormones. Thus a striking increase of adrenalin was observed after aldosterone and growth hormone administration, a moderate increase after oestradiol, and



Variation of adrenalin (A) and noradrenalin (NA) content in the adrenal medulla of control guinea-pigs (I), treated with oestradiol and glucagon (II), with oestradiol and aldosterone (III), oestradiol and growth hormone (IV), and oestradiol only (V). The vertical lines at the top of the columns indicate the standard deviation [S.E.

$$= \pm \sqrt{\sum d^2/n(n-1)}].$$

¹ J. MALMEJAC, *Physiol. Rev.* 44, 186 (1964).

² G. VITRY, CH. CHAMBOST, and S. DURAND, *C. r. Soc. Biol.* 157, 1029 (1963).

³ J. ROFFI, *C. r. Acad. Sci.* 249, 574 (1959).

⁴ L. STRAND, A. GOLDFIEN, and W. GANONG, *Endocrinology* 74, 656 (1964).

⁵ H. WEIL-MALHERBE and H. POSNER, *J. Pharmac. exp. Ther.* 140, 93 (1963).

⁶ A. LUPULESCU, *Ann. Endocr.* 22, 459 (1961).

⁷ A. LUPULESCU, *Endokrinologie* 48, 164 (1965).

⁸ U. S. VON EULER and F. LISHAJKO, *Acta physiol. scand.* 51, 348 (1961).

⁹ J. TRAMEZZANI, J. CIOCCIO, and G. WASSERMANN, *J. Histochem. Cytochem.* 12, 890 (1964).