

Table I. A similar response has been demonstrated by other workers¹⁻³. The higher temperatures proved lethal to the control fish acclimatized at 12°C: results of tests on 20°C acclimatized fish are shown in Table II. The controls selected the lower temperatures or, perhaps more correctly, avoided the higher temperatures. This was not shown by the thiourea treated fish.

At lower temperatures the thiourea treated fish were more mobile than the controls. An increase in tempe-

rature increased the mobility of the controls so that they became the more mobile group but did not affect the experimental group. A change in behaviour of salmon fry as a result of thiourea treatment has been shown by HOAR et al.⁴.

The behaviour of the thiourea treated fish could be explained by a loss of temperature perception. However, experiments following BULL's⁵ method have shown that these fish can still establish a conditioned reflex to a temperature increase. An alternative explanation depends on the anti-thyroid action of the drug. Thyroid changes affect behaviour in fish⁴, and as thiourea augments tolerance to high temperatures⁶⁻⁸ this may explain the movement of the thiourea treated fish into the higher temperatures. It is suggested that a change in thyroid activity is correlated with changes in swimming behaviour which lead to a variation in the selected temperature.

Résumé. Le traitement par le thiourée a augmenté la mobilité chez *Phoxinus phoxinus* aux basses températures, mais non aux hautes températures. Les cas témoins ont évité les hautes températures; ce qui n'était pas le cas chez les poissons soumis à l'expérience. Ces faits suggèrent une corrélation entre le nouveau comportement et la glande thyroïde.

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¹ J. R. BRETT, J. Fish. Res. Board Can. 9, 265 (1952).
² P. DOUDOROFF, Biol. Bull. 75, 494 (1938).
³ E. T. GARSIDE and J. S. TAIT, Can. J. Zool. 36, 563 (1958).
⁴ W. S. HOAR, D. MACKINNON, and A. REDLICH, Can. J. Zool. 30, 273, (1952).
⁵ H. O. BULL, J. mar. biol. Assoc. 15 (N. S.), 485 (1928).
⁶ N. V. EVROPEITZEVA, Dokl. Akad. Nauk., U.S.S.R. 68, 977 (1949); Quoted in G. E. PICKFORD and J. W. ATZ, *The Physiology of the Pituitary Gland of Fishes* (New York Zoological Society, New York 1957).
⁷ R. SUHRMANN, Biol. Zbl. 74, 432 (1955).
⁸ P. Y. FORTUNE, J. exp. Biol. 32, 504 (1955).

Table I						
T °C		19	20	21	22	23
Group		% no. times stationary				
C						
Acclimatised at 12°C		1.2	1.1	4.7	22.9	70.0
+ T						
Acclimatised at 12°C		4.3	17.4	18.2	29.8	30.2

Table II							
T °C		25	26	27	28	29	30
Group		% no. times stationary					
C							
Acclimatised at 20°C		50.5	33.7	12.4	0.25	0.5	2.6
+ T							
Acclimatised at 20°C		38.4	6.7	16.5	20.6	3.4	14.4

Average of all records. Each figure represents the average of number of times a temperature was selected in ten tests.

2,2'-Dihydroxy-6,6'-dinaphthyl Disulphide (DDD) Diazo Blue B Reactive Granules in the Parathyroid Gland of the Rat and Toad

It is commonly accepted that the parathyroid gland is an endocrine organ which elaborates and releases a polypeptide hormone. Although several intracellular materials have been interpreted as secretion, or its antecedents in the parenchymal cells of the gland¹⁻⁷, attempts to demonstrate morphologically parathyroid hormone or a related substance within the cells have not been very successful. During work on chemocytological features of the rat parathyroid gland⁸, I have noted the presence of 2,2'-dihydroxy-6,6'-dinaphthyl disulphide (DDD) diazo blue B reactive granules which consist of proteins with sulphhydryl and disulphide groups. The granules were seen primarily in the cytoplasm of the parenchymal cells, and additionally on the outer surface of the plasma membrane and within the interstitial connective tissue space including the vascular endothelial cells (Figure 1). Such distribution pattern has led me to the consideration that the

granules can pass through the plasma membrane of the parenchymal cells and may thus be associated with the secretory activity of the cells. Therefore, the object of my present work was to examine the morphology of the granules under varied conditions, in an attempt to elucidate their cytophysiological significance related to the elaboration and elimination of hormone in the parathyroid gland of rats of the Wistar strain and toads (*Bufo vulgaris japonicus*).

The method employed in the demonstration of the DDD diazo blue B reactive granules in the tissue was the

¹ G. BOBEAU, J. Anat. Physiol. 47, 371 (1911).
² J. A. ROSOF, J. exp. Zool. 68, 121 (1934).
³ A. M. PAPPENHEIMER and S. L. WILENS, Amer. J. Path. 11, 73 (1935).
⁴ E. V. COWDRY and G. H. SCOTT, Arch. Path. 22, 1 (1936).
⁵ A. L. GRAFFLIN, Endocrinol. 26, 857 (1940).
⁶ A. L. GRAFFLIN, Endocrinol. 30, 571 (1942).
⁷ R. J. WEYMOUTH and B. L. BAKER, Anat. Rec. 119, 519 (1954).
⁸ J. HARA and K. YAMADA, Z. Zellforsch. 57, 360 (1962).

original method of BARNETT and SELIGMAN^{9,10}. The parathyroid glands were taken from normal and bilaterally nephrectomized rats, and also from toads captured seasonally throughout the year. The glands were fixed overnight in trichlor acetic acid alcohol (1% trichlor acetic acid in 80% ethanol), embedded in paraffin, sectioned at 8 μ and stained.

In the parathyroid glands from bilaterally nephrectomized rats, a noticeable abundance of intensely DDD diazo blue B reactive granules was observed as compared with those from normal animals. These granules were most numerous in the cytoplasm of the parenchymal cells and particularly within its peripheral layer. An example of this abundance is shown in Figure 2.

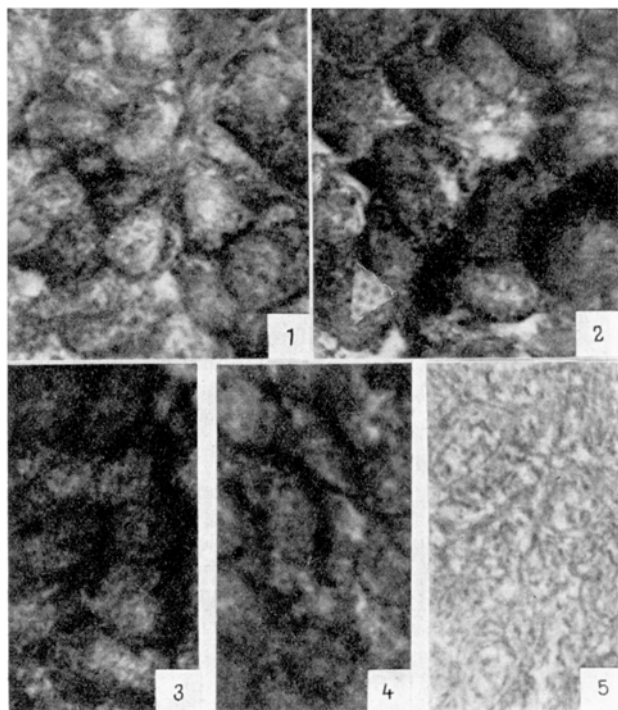


Fig. 1. DDD diazo blue B reactive granules in parathyroid tissue of a normal rat. $\times 1500$.

Fig. 2. An abundance of intensely DDD diazo blue B reactive granules in parathyroid tissue of a rat at 12 h after bilateral nephrectomy. $\times 1500$.

Fig. 3. An abundant amount of intensely DDD diazo blue B reactive granules in parathyroid tissue of a toad captured in July. $\times 1500$.

Fig. 4. A moderate amount of DDD diazo blue B reactive granules in parathyroid tissue of a toad captured in October. $\times 1500$.

Fig. 5. A minimal amount of faintly DDD diazo blue B reactive granules in parathyroid tissue of a toad captured in February. $\times 1500$.

In the parathyroid glands from toads sacrificed in summer, an abundant amount of intensely DDD diazo blue B reactive granules were seen in the cytoplasm of the parenchymal cells, on the outer surface of the plasma membrane and within the connective tissue space (Figure 3)—while in those from animals killed in spring and autumn the amount and stainability of the granules were found to be moderate (Figure 4). In the parathyroid glands from toads in dormant state, the granules were minimal in amount and staining intensity, in fact occasionally absent (Figure 5).

From the physiological point of view, the result is possible to comprehend. The present evidence indicates certainly that the amount and staining intensity of DDD diazo blue B reactive granules are approximately parallel to the extent with which the parathyroid gland is physiologically functioning in both animal species studied. On this basis, it may be concluded that the granules are a reflection of the secretory activity of the glandular cells. It deserves, however, discussion whether the granules represent parathyroid hormone itself or its supporting material. According to RASMUSSEN¹¹, parathyroid hormone consists of a polypeptide which is found on analysis to contain only an exceedingly small amount of cystine. Therefore, although the hormone may be responsible, to a certain extent, for the stainability of the DDD diazo blue B reactive granules, a carrier protein containing significant amounts of sulphhydryl and disulphide groups and existing in association with the active principle appears to constitute the major moiety of the granules. This notion seems significant in view of a biochemical evidence that the biological activity of parathyroid hormone is relevant to its oxidation-reduction properties¹¹.

More detailed studies on the morphological features of DDD diazo blue B reactive granules in the parathyroid tissues will be published elsewhere.

Zusammenfassung. Granula in Parenchymzellen von Ratten und Kröten-Epithelkörperchen treten durch 2, 2'-Dihydroxy-6, 6'-dinaphthyl-disulfid (DDD) Diazo-Blau-B gefärbt, deutlich hervor. Ihre Menge, Färbbarkeit und Verteilung unter verschiedenen Bedingungen scheinen für die zelluläre Sekretionstätigkeit charakteristisch zu sein.

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⁹ R. J. BARNETT and A. M. SELIGMAN, *Science* **116**, 323 (1952).

¹⁰ R. J. BARNETT and A. M. SELIGMAN, *J. Nat. Cancer Inst.* **14**, 769 (1954).

¹¹ H. RASMUSSEN, *The Parathyroids* (Charles C. Thomas Publisher, Springfield, Illinois 1961).

Plantar Reflexes in Cat

SHERRINGTON¹ described that broad innocuous pressure of the plantar cushion in the spinal dog evoked a bilateral brief extension of the whole limb—the extensor thrust. Reflex responses, in man, to painful stimulation of different parts of the foot have been elucidated². The present investigation on acute spinal cats deals with some plantar reflex actions to muscles performing toe and ankle

movements. Reflex actions from different skin areas to the short toe muscles flexor digitorum brevis (FDB) and extensor digitorum brevis (EDB) have been studied in a few cases³ but in general reflex connections to the short muscles of the foot are unknown.

¹ C. S. SHERRINGTON, *Proc. Roy. Soc.* **76**, 161, 269 (1905).

² E. KUGELBERG, K. EKLUND, and L. GRIMBY, *Brain* **83**, 394 (1960).

³ K.-E. HAGBARTH, *Acta physiol. scand.* **26**, Suppl. 94 (1952).