

the 3 doses chosen of amphetamine. Doses of reserpine ranged from 1×10^{-6} to 1×10^{-4} g/ml.

As can be seen, independently of the doses of amphetamine previously used, reserpine response increases in a fashion related to the dose. In the dosage levels of amphetamine used, the maximal response for reserpine corresponds to a concentration of 2.5×10^{-5} g/ml. A higher concentration of reserpine produces a smaller response which may even disappear at a concentration of 1×10^{-4} g/ml. This fact could be explained on the basis that higher concentrations of reserpine produce a blockade of alpha receptors or determine a non-specific effect. The height of the contraction elicited by reserpine has in all cases been smaller than that obtained by amphetamine in the same experiment. Each point of the graph represents the mean \pm the standard error of a minimum of 7 experiments and a maximum of 12.

Further experiments are necessary to elucidate the mechanism involved in the phenomenon described. However, if we take into account the results shown above and some unpublished results, several conclusions may be drawn: (1) The phenomenon appears when amphetamine or amphetamine-like drugs (ephedrine, tranylcipromine) are added before reserpine^{8,9}. (2) Nor-

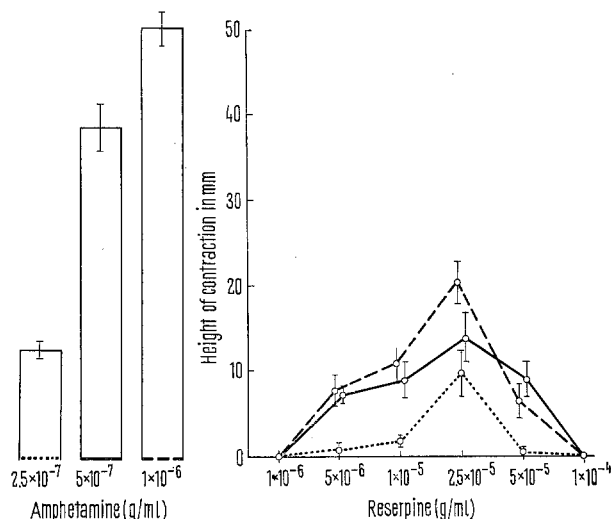
adrenaline and tyramine do not behave like amphetamine⁹. (3) It does not appear when the animals are pretreated with reserpine (1 mg/kg i.p. 24 h before the experiment). (4) The contraction produced by reserpine appears generally once. In some cases a second or even a third dose of reserpine elicit a response, which is always smaller than the previous one. In any case, further doses of reserpine fail to show any response. The weight of the animals and their corresponding vasa deferentia nor-adrenaline content could explain the different number of the responses of the preparations to reserpine. (5) It is blocked by alpha-type adrenolitics (phentolamine).

The abolition of the reserpine response by pretreatment in vivo with the alkaloid and/or pretreatment in vitro with phentolamine, points to an adrenergic mechanism. The possibility that amphetamine modifies the reserpine response through an inhibition of catecholamine uptake or a facilitation of catecholamine release will be investigated¹⁰.

Resumen. Se estudia en el conducto deferente aislado de rata la respuesta a la reserpina después de la estimulación del preparado con anfetamina. En estas condiciones, la reserpina produce una contracción que es proporcional a las concentraciones de anfetamina y reserpina utilizadas, y es máxima para una concentración de reserpina de $2,5 \times 10^{-5}$ g/ml, para ser luego decreciente cualquiera que sea la concentración de anfetamina que se haya empleado previamente. Se discuten los posibles mecanismos que regulan la respuesta a la reserpina después de la anfetamina.

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In vivo interactions between amphetamine and reserpine., after amphetamine (2.5×10^{-7} g/ml); [—], after amphetamine (5×10^{-7} g/ml); - - - - -, after amphetamine (1×10^{-6} g/ml). Each point represents the mean \pm S.E.

⁸ F. G. VALDECASAS, E. CUENCA and L. RODRÍGUEZ, Actas IX Reunión Nac. Soc. Esp. Ciencias Fisiológicas, Pamplona 1965, p. 177.

⁹ F. G. VALDECASAS, J. LAPORTE and F. JANÉ, Actas X Reunión Nac. Soc. Esp. Ciencias Fisiológicas, Valencia 1967, p. 121.

¹⁰ The authors wish to thank CIBA (Spain) and Laboratorios Miquel S.A. (Spain) for their generous supply of Serpasol® and amphetamine respectively.

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A Neurite-Receptor Complex in the Avian Lung: Electron Microscopical Observations

Electron microscopic observations have demonstrated that the glomus cells of the carotid body¹⁻³ contain many dense-cored granular vesicles and are closely associated with unmyelinated axons. Certain cells in epidermal structures of vertebrates have very similar ultrastructural characteristics, notably Merkel cells in mammalian skin^{4,5} and in the avian hard palate⁶. We have found very similar specialized cells in the epithelium of the intrapulmonary primary bronchus of the domestic fowl (*Gallus domesticus*). Comparable cells have also been described in the human bronchial epithelium⁷.

Specimens of *G. domesticus*, known to be free of certain respiratory diseases, were perfused under barbiturate

anaesthesia with 2.5% glutaraldehyde in cacodylate buffer (pH 7.3). The required tissue was removed, post-fixed in osmium tetroxide and embedded in Maraglas.

¹ J. D. LEVER, P. R. LEWIS and J. D. BOYD, J. Anat. 93, 478 (1959).

² F. AL-LAMI and R. G. MURRAY, J. Ultrastruct. Res. 24, 465 (1968).

³ T. J. BISCOE and W. E. STEHBENS, J. Cell Biol. 30, 563 (1966).

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⁶ A. E. ANDERSEN and P. H. J. NAFSTAD, Z. Zellforsch. mikrosk. Anat. 97, 391 (1968).

⁷ K. G. BENSCH, G. B. GORDON and L. R. MILLER, J. Ultrastruct. Res. 12, 668 (1965).



Fig. 1. Single specialized cell between epithelial cells. The cytoplasm is invaginated (I) into the nucleus. Desmosomes (→). Lead citrate and uranyl acetate. $\times 8300$.

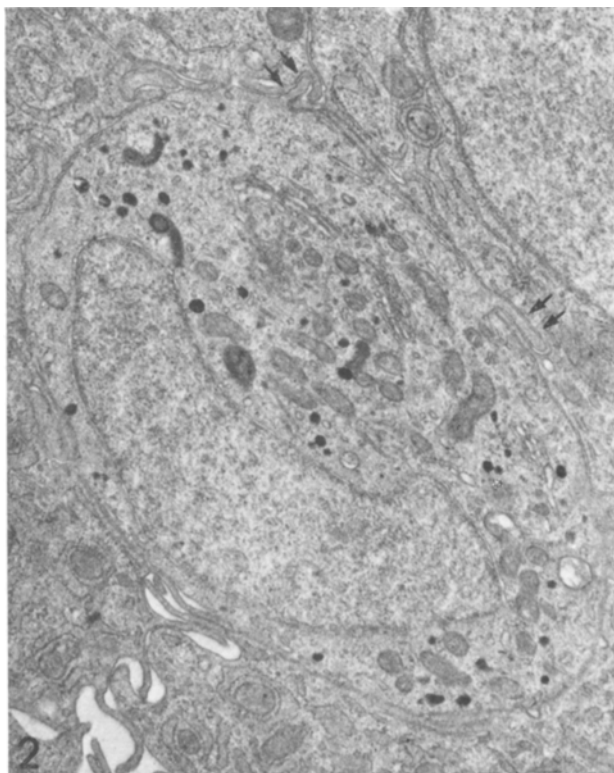


Fig. 2. Specialized cell showing the Golgi body. Cytoplasmic processes are indicated by \rightleftharpoons . Lead citrate. $\times 15,000$.

Ultrathin sections were stained with lead citrate and uranyl acetate or with lead citrate only.

These specialized cells occur in small groups, but each individual cell is isolated (Figure 1) by the ciliated columnar epithelial cells which line the primary bronchus. The cells have an irregular outline. Although they do not possess the extensive foldings of the surface membrane which characterize the adjacent epithelial cells, they do occasionally show small processes which penetrate a short distance between the adjoining epithelial cells (Figure 2). Some of these small processes bifurcate. In some sections, desmosomes are seen between these specialized cells and the surrounding epithelial cells (Figure 1). The cytoplasm contains numerous free ribosomes, many dense-cored granular vesicles (about 750 to 1300 Å diameter, with a core about 600–1000 Å diameter), sparse rough endoplasmic reticulum, and occasional fine filaments. The mitochondria are small and round or narrow and elongated. Their arrangement in the cell usually appears to be random, though sometimes they seem to be accumulated at the basal region. The Golgi complex is well-developed (Figure 2) and occasionally 1 or 2 dense-cored granular vesicles are associated with it. The nucleus, which seems to contain 2 nucleoli, appears lobulated and irregular, or oval and regular, depending on the plane of the section. The chromatin is evenly dispersed throughout the nucleus. No evidence of sustentacular cells or supporting structures has been observed.

There are usually 1 (Figure 3) or sometimes 2 unmyelinated axons in close contact with the basal region of

these cells. These axons are typically bare of Schwann cell cytoplasm, and appear to have a greater diameter than those commonly seen in the nerve bundles in the lamina propria beneath the bronchial epithelium. The axons associated with the specialized cells are randomly packed with small round, or elongated mitochondria and many agranular vesicles (450–950 Å diameter). Dense-cored granular vesicles are not usually present. The gap between the axonal and cell membranes is very narrow, about 100–150 Å, and in some cases small interlocked interdigitations have been seen.

These cells resemble the glomus cells and Merkel cells in having granular vesicles, small processes penetrating between adjacent epithelial cells, and close association with apparently enlarged axons full of mitochondria and agranular vesicles. The glomus cells are widely accepted as chemoreceptors, whilst Merkel cells are thought to be mechanoreceptors. It therefore seems reasonable, on the morphological evidence, to consider the possibility that these cells in the bronchial epithelium of *G. domesticus* are another example of a neurite-receptor cell complex, either chemoreceptor or mechanoreceptor. There is experimental evidence for the presence of carbon dioxide receptors in the intrapulmonary primary bronchus of this species of bird⁸. It is also known that afferent vagal

⁸ D. F. PETERSON and M. R. FEDDE, *Science* 162, 1499 (1968).

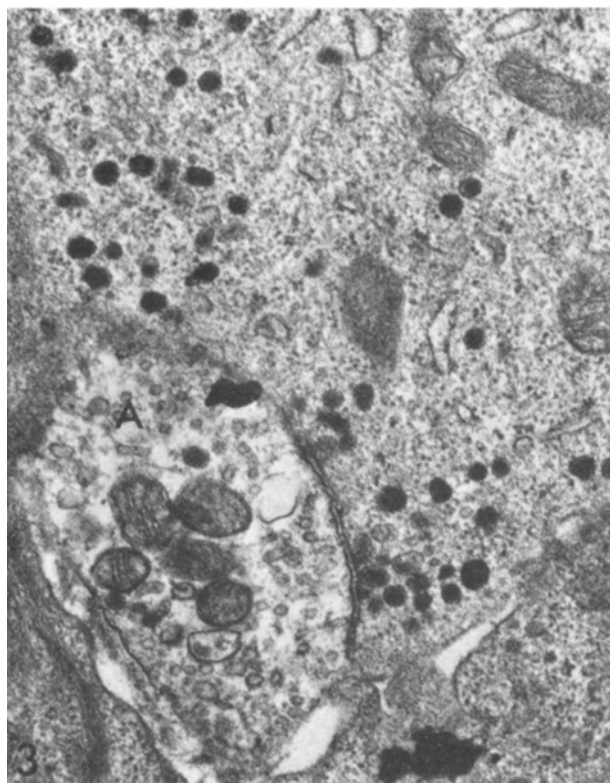


Fig. 3. Shows the close relationship between the axon (A) and the specialized cell. Note the mitochondria and agranular vesicles in the axon and the concentration of granular vesicles in the specialized cell near the axon. Lead citrate and uranyl acetate. $\times 32,000$.

pathways fire in phase with breathing in this species^{9,10}, and this is consistent with mechanoreceptor activity.

Using the light microscope, FRÖLICH described 'light cells' in the bronchial epithelium of several mammals¹¹. These cells, which were sometimes present in clumps, appeared to be innervated by fine nerve fibres extending deeply into their cytoplasm, and he suggested that they

were chemoreceptors. The presence of these cells in the normal human lung has been confirmed¹², but apparently their innervation has not yet been corroborated. BENSCH et al.⁷ suggested that their new cell type probably corresponded to FRÖLICH's¹¹ 'light cells', but did not report a close contact between these cells and nerve endings. The specialized cells which we have described very probably correspond to FRÖLICH's 'light cells', and they do have a close relationship to axons.

If our cells are chemoreceptors they suggest the possibility of a device for the rapid and direct monitoring of gases within the airway, as opposed to the relatively indirect monitoring of gases in the blood. If they are mechanoreceptors they are of interest in that it has been difficult hitherto to demonstrate convincingly any definite receptor structures within the vertebrate lung which might be sensitive to mechanical stimuli¹³.

Résumé. L'examen au microscope électronique du bronchus primaire intrapulmonaire de *Gallus domesticus* a révélé la présence d'un complexe neurite-récepteur très semblable aux cellules du glomus carotidien des mammifères, et aux cellules de Merkel qui se trouvent dans les structures épidermiques des vertébrés. La fonction possible chémorécepteur ou mécanorécepteur de ces cellules est discutée.

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¹¹ F. FRÖLICH, *Frankfurt. Z. Path.* 60, 517 (1949).

¹² H. VON HAYEK, *The Human Lung* (Hafner, New York 1960), p. 130.

¹³ The electron microscope was presented by the Wellcome Foundation and the project was sponsored by the British Egg Marketing Board.

Effect of Temperature on the Neurosecretory Activity in *Nezara viridula* Linn. (Heteroptera; Pentatomidae)

The role of temperature on the activity of neurosecretory cells has not so far been studied in detail in insects (NOVAK¹). The only report is that of CLARKE², who has studied the histological changes in the neuroendocrine system of *Locusta migratoria* at different temperature regimes. In the present work an attempt has been made to study the role of temperature on the activity of the neurosecretory cells of the brain of green cotton bug, *Nezara viridula*.

Material and method. The adults of *Nezara viridula* of about equal age group were collected from the plants of *Althaea rosea*. For each replicate experiment they were sorted out in 4 lots. The first lot was kept in an incubator maintained at a temperature of 35 °C. The second and third lots were kept in a refrigerator maintained at 10 and 0 °C for 21 days and 4 days respectively. One lot was always kept at the room temperature (28 °C) as control.

The insects kept at 35 and 10 °C lived for several days, but those at 0 °C survived for 3–4 days only. The experimental and controlled insects were studied after staining with paraldehyde fuchsin³ (PF) and performic acid victoria blue (PAVB)^{4,5} techniques.

Results and discussion. In the pars intercerebralis medialis of the protocerebrum of *Nezara viridula*, there are 2 distinct groups of neurosecretory cells, situated

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² K. U. CLARKE, *J. Insect Physiol.* 12, 163 (1966).

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⁵ G. S. DOGRA and B. K. TANDAN, *Q. Jl. microsc. Sci.* 105, 455 (1964).