Absence of Salivary Invertase in Queen and Drone Honeybees

Worker honeybees' hypopharyngeal glands contain much invertase, and there is little or none in the other glands (post-cerebral, thoracic and mandibular) that discharge on their mouthparts[1,2]. Queen and drone honeybees have no hypopharyngeal glands, so the possibility that their other salivary glands might produce invertase has now been investigated. As drones' postcerebral and mandibular glands are vestigial, only their thoracic glands were tested.

Each pair of glands was removed under distilled water and ground up in sucrose solution buffered to pH 6.5. The amounts of glucose present before and after 24 h incubation at 35°C were estimated by the iodine oxidation method. The procedure was calibrated with a standard glucose solution and its reliability tested by titrations of buffered sucrose before and after incubation.

The amounts, if any, of invertase in the glands of queens and drones were small compared with those in the hypopharyngeal glands of workers (Table). The large standard error of the latter results from the great variation in the amount of invertase in individual bees.

When bees convert nectar into honey, its sucrose is inverted in their honey-stomachs, which can receive invertase only from the salivary glands. As queens and drones do not participate in honey storage they probably do not need salivary invertase. Drones, at least, eat large quantities of honey or nectar directly from the cells[3] but the invertase required to digest any sucrose this food contains is supplied by the mid-gut cells[4].

Invertase in salivary glands

Bees	Glands	Number of observations	Mean amount (mg) of glucose produced in 24 h	Standard error of mean
Worker	Hypopharyngeal	5	35.30	5.99
Queen	Mandibular	5	0.72	0.36
Queen	Post-cerebral	6	0.25	0.86
Queen	Thoracic	6	0.32	0.21
Drone	Thoracic	5	0.36	0.43

Zusammenfassung. Untersuchungen an Mandibel-, Hinterkopf- und Thoraxdrüsen der Bienenkönigin sowie an Thoraxdrüsen der Drohne haben gezeigt, dass sie, verglichen mit den Hypopharynxdrüsen der Arbeiterin, nur sehr wenig oder gar keine Invertase enthalten. Da weder Königin noch Drohne an der Honigspeicherung teilnehmen, brauchen sie keine Speicheldrüsen-Invertase.

I. B. Riedel and J. Simpson

Bee Department, Rothamsted Experimental Station, Harpenden (Herts, England), May 2, 1961.

1 E. Kratky, Z. wiss. Zool. *139*, 120 (1931).
2 J. Simpson, J. ins. Physiol. *4*, 107 (1960).
3 J. B. Free, Brit. J. anim. Behav. *5*, 7 (1957).
4 E. N. Pavlovsky and E. J. Zarin, Quart. J. micr. Sci. *66*, 509 (1922).

Similar Pattern of Fine Structure in the Basement Lamella of the Skin and the External Sheath of the Notochord in *Xenopus* larvae[1]

Electron microscopical observations on the larval skin of various Amphibian species revealed a common pattern of fine structure in the basement lamella of the epidermis[2–7]. As already suggested by Rosin[4] from studies by light microscopy, the basement lamella was found to consist of orthogonally arranged plies of collagenous fibrils which gradually appear during embryonic development[2].

In the course of electron microscopical observations on tissues of *Xenopus* larvae, we found a very striking similarity in the texture of the basement lamella of the epidermis and in the external sheath of the notochord. The present findings refer to tail tissue of *Xenopus* larvae, measuring about 30 mm in length. The tissue samples were fixed in a mixture of 1% OsO_4 and 0.5% neutralized potassium-di-chromate for 3 h and then immediately transferred into 65% alcohol containing 0.1% uranium nitrate. For dehydration, embedding and sectioning, the conventional procedures were followed.

Figure 1 represents the basement lamella which is located between the epidermal and the pigment cells. The boundary of the epidermal cells is marked by the so-called 'hyaline layer' which is about 0.2 μ thick and contains fibrillar and presumably also granular components. The basement membrane proper is composed of about 12 orthogonally arranged plies, some of which reveal at least two layers of collagenous fibrils. In *Xenopus* larvae the collagenous fibrils are on an average 200 Å thick, and show a periodicity of about 400 Å; it is also noteworthy that open communications may be found between the intralamellar space and the underlying pigment cells. The total thickness of the basement membrane, including the hyaline layer, is about 1.5 μ.

Figure 2 represents a small portion of the external sheath of the notochord in cross-section. It clearly reveals the pattern of orthogonally arranged plies in which the collagenous fibrils are rather tightly packed. On closer inspection, there are some differences as compared with the basement lamella. Thus, the outer boundary (500 Å) of the sheath is composed of very thin circular fibrils. Its body shows at least 13 crossed plies which often comprise several layers of fibrils, having similar dimensions as those of the basement lamella. At the inner boundary, the sheath communicates with the epithelial cells of the notochord.

In summary, our observations on *Xenopus* larvae reveal the occurrence of a common texture in both the epidermal basement lamella and the external sheath of the notochord, being composed of orthogonally arranged collagenous fibrils of approximately 200 Å in diameter. In the notochord sheath, the collagenous fibrils are more tightly packed than in the basement lamella[8].

1 Supported by the 'Swiss National Science Foundation for the Promotion of Scientific Research'.
2 N. E. Kemp, Developmental Biol. *1*, 459 (1959).
3 K. R. Porter, Anat. Rec. *118*, 433 (1954).
4 S. Rosin, Rev. Suisse Zool. *53*, 133 (1946).
5 M. M. Salpeter and M. Singer, J. biophys. biochem. Cytol. *6*, 35 (1959).
6 P. Weiss and W. Ferris, Exp. Cell Res. *6*, 546 (1954).
7 P. Weiss and W. Ferris, Proc. Natl. Acad. Sci. U.S. *40*, 528 (1954).
8 The author is greatly indebted to the Laboratory of Electron Microscopy at the Institute of Inorganic Chemistry in the University of Bern for all facilities that were put at his disposal.

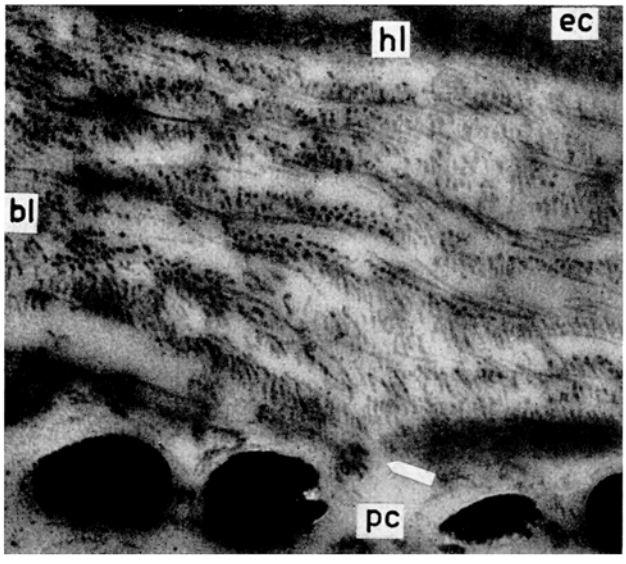

Fig. 1. *Basement lamella* (32 000 : 1): ec = epidermal cells, hl = hyaline layer, bl = basement lamella with crosswise arranged collagenous fibrils at (↓) communicating with pigment cell (pc).

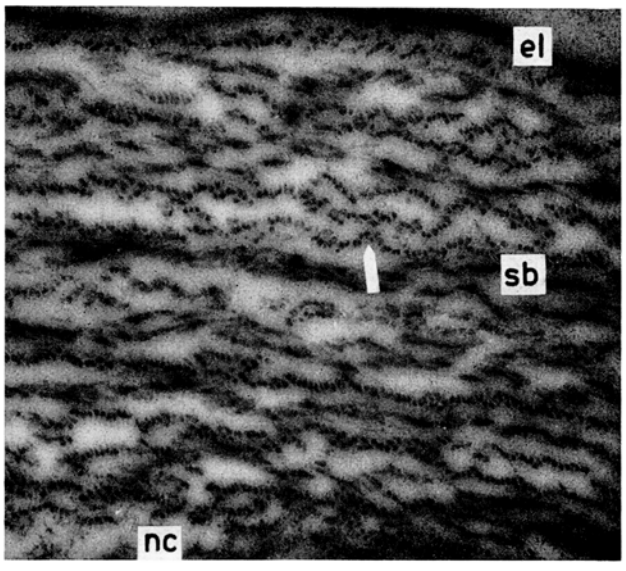

Fig. 2. *External sheath of the notochord* (32 000 : 1): el = external layer with circular fibrils. The plies of the sheath body (sb) appear slightly folded (↓) and are composed of several layers of fibrils. The internal boundary is marked by epithelial cells of the notochord (nc).

Zusammenfassung. Auf Grund von elektronenmikroskopischen Beobachtungen wird nachgewiesen, dass die Basalmembran der Epidermis und die äussere Chordascheide im Schwanz von *Xenopus*larven eine übereinstimmende Feinstruktur besitzen; sie ist gekennzeichnet durch orthogonal angeordnete Kollagenfibrillen.

R. WEBER

Zoologisches Institut der Universität, Bern (Switzerland), May 5, 1961.

Beat to Beat Analysis of the Radiocardiogram

In radiocardiography the impulses from the precordial detector are usually fed into a ratemeter with a direct-writer (PRINZMETAL, CORDAY, SPRITZLER, and FLIEG[1], VEALL, PEARSON, HANLEY, and LOWE[2], PIETILÄ and HAKKILA[3]). The inertia of this type of recording makes it difficult to register correctly rapid changes in radiation intensity such as the dilution of a radioactive indicator in determinations of cardiac output. The inertia of the ratemeter makes it impossible to record correctly the changes in the radioactivity of the radiocardiogram during individual heart cycles. The disadvantages of the ratemeter can be avoided if the impulses from the detector are recorded on a tape recorder running at a high speed (BERNE, HALLBERG, and LINDELL[4]). The magnetic tape can then be analyzed when running at low speed, using an ordinary scaler. In this way the activity, during time intervals corresponding to 0.075 sec at the original recording, may be correctly counted. This type of recording, which allows a 16-fold stretching of the time axis, is free of inertia and will accept up to 10^7 impulses/min. The record may be analyzed repeatedly using different counting intervals, if this is desirable. Moreover, using a double printer, recording alternate intervals, the system can be fully automatized.

Such a tape recorder was used in the present experiments. It has 3 channels, one of which was used to record the pulses from a precordial scintillation detector. Another channel was used to register the pulses from a scintillation detector through which arterial blood was sucked at constant speed. This record was used to calculate cardiac output (BERNE et al.[4]). The R waves of the electrocardiogram were fed into the third channel of the tape recorder. The experiments were carried out in anaesthetized dogs, lying on their right side. The precordial detector was placed over the heart from above with the help of fluoroscopy. The collimation was such that the detector 'saw' the whole heart and as little as possible of the surrounding tissues. Arterial blood, for the determination of cardiac output according to the indicator dilution technique, was drawn from a femoral artery. Radioactive potassium iodide (I^{131}) in saline solution was injected intravenously in doses of 10 μC.

In the semiautomatic analysis of the record in this study, the first R wave after the injection started the counter. As many 0.075 sec intervals as possible were counted between the R waves. An indetermined time interval (< 0.075 sec) preceding each R wave was omitted in the record (Figure 1).

The records from an experiment are shown in Figure 1. It may be seen that there is a decrease in the radioactivity recorded by the precordial detector about 0.1 sec after the R wave in the electrocardiogram. This decrease is a regular phenomenon and it is fairly constant for 3 heart cycles, during the passage of the radioactive bolus through

[1] M. PRINZMETAL, E. CORDAY, R. J. SPRITZLER, and W. FLIEG, J. Amer. med. Assoc. *139*, 617 (1949).
[2] N. VEALL, J. D. PEARSON, T. HANLEY, and A. E. LOWE, Radioisotope Conf. *I*, 183 (1954).
[3] K. A. PIETILÄ and J. HAKKILA, Cardiologia *36*, 97 (1960).
[4] E. BERNE, L. HALLBERG, and S. E. LINDELL, Scand. J. lab. clin. Invest., in press (1961).