Association. The steroids were provided by Dr. Robert L. Craig, G. D. Searle and Co., Chicago.

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ENTOMOLOGY

Secretion of Wax through the Cuticle of Insects

ON the outside of an insect there is a layer of wax or grease which is its main protection against desiccation[1]. At moulting the wax appears as an outer layer of the epicuticle about 0·1–1μ thick[2] at the time the old skin is shed. Wax may also be secreted during an instar for the repair of abrasions to the cuticle. This poses the problem of how the wax—often hard and with a m.p. above 60° C.—can be transported from the epithelium to the outside through the hydrophilic endocuticle.

Three mechanisms have been suggested. (1) The wax may pass through the pore canals; but this cannot be a general explanation for pore canals do not reach the surface in all insects. (2) The wax may be conveyed to the surface in a solvent which later evaporates, for in cockroaches there is a grease composed of a hard wax dissolved in long chain alcohols and hydrocarbons[3]. (3) The wax may be secreted in a water-soluble form in combination with a protein as it is in the eggs of ticks[4].

In most insects these hypotheses are difficult to verify because of the small quantity of wax present and the complexity of the other events taking place at moulting. However, in the last instar larva of *Calpodes ethlius* Stoll (Lepidoptera, Hesperidae) wax secretion can be observed conveniently localized in space and time.

Three days after moulting to the final instar the larva appears as in Fig. 1 (left). During the next 48 hr., a wax (m.p. 81–82° C.) is extruded as filaments from fine pores in the epicuticle over a well-defined region of the ventral surface of abdominal segments 7 and 8 (Fig. 1, right). One or two days later the larva ceases feeding and begins to pupate.

In *Calpodes* there are no pore canals from the epithelium to the epicuticle, and the surface where wax appears is separated from the cells by a hydrophilic endocuticle about 50μ thick. Sudan stains do not show any soluble protein-bound lipid in the endocuticle. The third hypothesis is more difficult to disprove but (*a*) rearing the larvæ in the presence of decane or decanol vapour does not suppress wax formation (*b*) wax continues to be secreted in an area enclosed by paraffin wax, celamel cement, arildite, rubber latex or below a glass plate sealed in place with paraffin wax, and (*c*) the wax filaments are very slightly birefringent but not crystalline.

There remains the possibility that the wax is synthesized at the surface from its constituent fatty acids and alcohols which might themselves diffuse across the endocuticle. A search was therefore made in the epicuticle for an esterase which could effect this synthesis.

Fig. 1. Last instar larvæ of *Calpodes ethlius* from the underside. Left, larva almost fully grown; right, larva about 3 days before pupation showing wax-secreting areas

An esterase is readily demonstrated with 'Tween 80', O-acetyl-5-bromoindoxyl, α-naphthyl acetate and 'Naphthol A.S.' acetate as substrates, using whole larvæ abraded to expose a cut surface of the epicuticle. 10^{-3} *M* potassium cyanide was used to inhibit the phenolase system which would otherwise produce rapid blackening. The esterase reaction is more intense over the wax secreting areas but is not confined to them, suggesting that the formation of wax filaments is but an exaggeration of normal secretion due to a greater quantity of the wax precursor. The results with O-acetyl-5-bromoindoxyl as a substrate indicate that the esterase is on the outside of the epicuticular phenolase system. In the absence of cyanide, only the deep scratches blacken due to the phenolase although an esterase is shown in the more superficial scratches by the blue deposit of indigo. There is then an esterase at the surface of the epicuticle which could synthesize wax *in situ*, the insolubility of the end product favouring the production of wax.

The observations suggest that the efficiency of insecticides containing abrasive fillers might be improved by the addition of an esterase inhibitor to prevent repair of the wax layer.

A full account will be published elsewhere.

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New Neotropical Element (*Anoplotermes*) in Indian Termite Fauna

THE termite genus *Anoplotermes* Müller[1] is, along with *Speculitermes* Wasmann[2], unique among termites in not having a soldier caste, only the workers and the reproductives being present. The two genera have a close resemblance to each other. *Speculitermes* has been regarded by certain authorities, for example, Holmgren[3] and Grassé[4], as a subgenus of *Anoplotermes*, while others[5] regard it as a full genus. We believe the latter view to be more tenable since the differences between the two genera are considerable, though they are undoubtedly closely allied. They may be keyed as follows: