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Comparative Ultrastructure of Corneal Surface Topography in Insects with Aspects on Phylogenesis and Function

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Summary. The corneas of some nocturnal Lepidoptera carry an array of surface protuberances (nipples), about 200 mμ high, that acts as an impedance transformer equalizing by gradual transition the refractive index of air to that of the cornea. A screening of the insect class has been carried out in the present study with respect to the variation in corneal topography seen in previous observations.

361 species in most insect orders were prepared for EM by thin sectioning or platinum replication. Using the amplitude of the surface protuberances as parameter, a grouping of the cornea types was made on the basis of the well-defined variation of nipple heights observed between individuals of certain species. Thus, the corneas of one extreme group were either smooth (Fig. 4A) or possessed *protrusions* (Fig. 4B and C) less than about 50 mμ high, arranged either irregularly or in a regular, hexagonal array (group I). At the other extreme, there was a group with "*full-sized*" *nipples* (Figs. 1 and 3) ranging in amplitude around 250 mμ (group III). An intermediate group of nipple heights (group II) comprised corneas with "*low-sized*" *nipples* (Fig. 6) between 50 and about 200 mμ high. *Regularity* in the arrangement of the various types of protuberances was observed both in groups I and II (compare Figs. 4C and 8). *Irregularity* tended to be associated with low amplitudes (*e.g.* Fig. 4B), being found only in group I corneas.

Full-sized nipples (group III corneas) were found only among the anagenetically highest orders, Trichoptera and Lepidoptera (Tables 3 and 4), which, however, also had corneas with low protrusions (group I) and low-sized nipples (group II). In one of the two mecopteroid orders, Diptera (Table 2), protrusions and low-sized nipples were found (the latter only in Culicomorpha). The corneas of all other orders (Table 1) had only group I corneas with one noteworthy exception, Thysanura, the most primitive order investigated. All four thysanuran species examined had corneas with low-sized nipples (Fig. 8), thus belonging to group II.

The morphological findings demonstrated in the present study can be taken to illustrate the following trends.

1. a) The higher relative number of group II and III corneas (low- and full-sized nipples) in the anagenetically highest orders and the exclusive occurrence of group III corneas in the most advanced orders, Trichoptera and Lepidoptera, may indicate a progressive development of nipples during phylogenesis. Thus, the full-sized nipples may represent an apomorphous condition. Parallelisms — leading to the appearance of low- and full-sized nipples — may have occurred several times during insect phylogenetic development, thus possibly reflecting an evolutionary potential (with or without a selective pressure for this character).

b) Low-sized nipples were found in the most primitive insect order, Thysanura. This circumstance may point to regression from full-sized nipples that were present in an ancestor common to all insects. In such a case, the full-sized nipples in Trichoptera and Lepidoptera are to be considered plesiomorphous.

At present nothing supports conclusively a preference for one of these two alternatives.

2. Among the Lepidoptera, a greater relative number of butterflies than moths lack full-sized nipples. This may be taken to indicate a regression of nipples in the day-flying group of insects in comparison with their possible moth-like, nocturnal ancestors. That in many species there are low-sized nipples (the regression thus being incomplete) may indicate that a function of the nipples in ranges of shorter wavelengths has brought about an arrest of the regression at lower amplitudes.

During an electron microscopic work on photoreceptor structures in a night moth, *Prodenia eridiana* (Sphingidae), Bernhard and Miller (1962) discovered that the corneal surface carried cone-shaped protuberances, about 200 mμ in height, which were termed nipples (see Fig. 1). It could be inferred from sections tangential to the corneal surface that the nipples were placed close together in a hexagonal array. This arrangement was confirmed in carbon replicas and in scanning electron micrographs (Miller, Møller and Bernhard, 1966).

The action of the nipple array was shown in microwave experiments on dielectric lens models scaled to the frequency of the microwaves (Bernhard, Miller and Møller, 1963, 1965) as well as in comparative spectrophotometric measurements on corneal fragments from insects with nippled and non-nippled facets (Miller, Møller and Bernhard, 1966). These experiments demonstrated that the nipple array decreases the reflection and increases the transmission of visible light. A mathematical model was also set up to describe the mechanism of the nipple array (Bernhard, Miller and Møller, 1963, 1965). The calculations were made for radiation normal to the surface of the cornea. The curve for the decrease in reflection was calculated for a model with nipples formed as ideal sharp-tip cones and illustrated a broad band anti-reflection effect with a maximum for wavelengths corresponding to about twice the nipple height. The nipple array can thus be regarded as an impedance transformer matching the characteristic impedance of air to that of the lens. Correspondingly, there is, in the insect, a gradual transition of the refractive index from that of air to that of the lens material. This transition results from the form of the nipples, the proportion of corneal material gradually changing from 0% at the tips to 100% at the bases of the nipples. Since the nipple height is smaller than the wavelengths of visible light, the effect of the nipples is achieved without interferring with the image-forming capacity of the lens.

The biological significance of the nipple pattern described above has been discussed (Bernhard, Miller and Møller, 1965; see also Bernhard, 1967; Bernhard, Gemne and Møller, 1968; Miller, Bernard and Allen,