## A Replica Technique for the Study of Psyllid Wing Microstructure

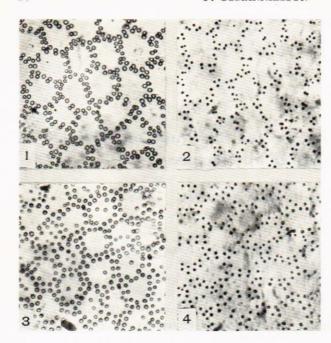
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The fore wing microstructure in *Psyllidae* has been used by many workers as a valuable specific character. In their study of this detail, however, most authors have not kept the microstructure of the under wing surface apart from that of the upper surface. On the other hand, Karel Šulc († 1952) in his important Psyllid works described the microstructure of both wing surfaces separately, by which he gained one additional character for the separation of species. Recently e.g. Schaefer, Vondráček and Klimaszewski have made use of this character in their papers.

The separate study of the microstructure of the two wing surfaces is not an easy matter, however. Though the under surface spinules are generally on an average distinctly smaller than those of the upper surface, variation in the size of individual spinules makes this criterion unreliable. Further, the difference in level of the two surfaces can be made use of, but since the wing membrane is far from plane, this means of distinction is very limited in application. If the wing is made to float on a drop of a highly refractive medium, e.g. Canada balsam, the spinules on the surface being in touch with the latter will be made less visible than those on the opposite, free surface. However, the difference is not always sufficiently marked to be relied on. My attempts to stain the spinules of one wing surface only by various dyes have been unsuccessful so far.

For electron microscope examination of surface structures of objects which cannot be penetrated by the electron beam, various techniques of making surface replicas have been developed. (See any handbook on electron microscopy, e.g. Bradley 1961). The essential idea is coating the specimen by a film which after removal from the surface to be studied will constitute an exact negative replica of the latter. This replica can be examined in the electron microscope, and also in the light microscope, within the range of its possibilities.

For some time I have made use of a similar technique for the purpose here discussed. The procedure is as follows. By aid of a brush or a glass rod a large drop of some suitable colourless plastic solution is painted on the center of one side of a clean glass slide. After evaporation of the solvent, the solution used should make a film attaching to the glass and with a permanent



Figs. 1—4. Replicas of fore wing microstructure. (1) Craspedolepta latior W. Wagner, \$\frac{3}{2}\$, paratype from Germany, Holstein, upper side of wing. (2) same individual, under side of wing. (3) Craspedolepta malachitica (Dahlbom) \$\frac{3}{2}\$, specimen from Oland, Mörbylånga, upper side of wing. (4) same individual, under side of wing. \$\times 450\$.

surface structure. Therefore, e.g. Canada balsam is unsuitable since impressions made on its surface will later smooth down. I use "Becker's Cellulose Fixative", a Swedish commercial product on nitrocellulose basis made for fixing pencil and charcoal drawings. (Probably collodium will also be satisfactory). The solution should cover a large proportion of the slide, at least 1/3 of its area, to prevent the film obtained from loosening from the slide at a later stage of the procedure here described. After a few minutes the film will be dry. Then one more drop of the fluid is added on the center of the slide. After two or three minutes this drop will have got the right viscosity. In the meantime the wing to be used should be cleaned in xylene or ethyl acetate, if necessary, and dried on a piece of filter paper. The wing is then carefully placed on the center of the still fluid drop of solution by aid of a pointed pair of watchmaker's forceps. The wing should float on the surface of the drop. If the fluid is still too thin, the wing will tend to submerge more or less, which should be avoided. If this happens, the wing can be cleaned in ethyl acetate and dried before another attempt is done. Air bubbles under the wing should be carefully removed by aid of a fine needle. Then the preparation should be left to dry for a few hours. Now the wing is to be removed. This can be done by pressing a narrow strip of adhesive tape firmly against the wing and pulling it away. Care should be taken not to attach the tape to a large part of the film surface, or the film will also be detached from the glass. If adhesive tape is used as above described, the wing can be released from the tape by dissolving the glue in xylene. However, the wing will often be torn into pieces by using adhesive tape. If one is dealing with unique specimens which should be spared as much as possible, the wing can often be

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loosened from the film by covering it with a large drop of water containing a small proportion of some substance reducing surface tension, e.g. soap or any synthetic washing material. After a few minutes the wing can be loosened by aid of pair of very fine needles, e.g. micro-pins attached to holders. The soap solution is then rinsed away with distilled water.

The photographs reproduced in Figs 1—4 showing details of the microstructure of the upper and under surfaces of fore wings of two Craspedolepta

species were taken from replicas made as above described.

## Reference

Bradley, D. E., 1961. Replica and shadowing techniques. In: Kay, D.: Techniques for Electron Microscopy. Oxford 1961. Chapter V, pp. 82—137.