*References Cited*

Dyar, H. G. and Shannon, R. C. 1927. The North American two-winged flies of the family Simuliidae. Proc. U. S. Nat. Mus. 69(10):1–54.

Fredeen, F. J. H. 1977. A review of the economic importance of black flies (Simuliidae) in Canada. Quaes. Entomol. 13:219–229.

Peterson, B. V. 1959. Notes on the biology of some species of Utah black flies (Diptera: Simuliidae). Mosq. News 19:86–90.

Peterson, B. V. 1970. The *Prosimulium* of Canada and Alaska (Diptera: Simuliidae). Mem. Entomol. Soc. Can. 69:1–216.

Sommerman, K. M., Sailer, R. I. and Esselbaugh, C. O. 1955. Biology of Alaskan black flies (Diptera: Simuliidae). Ecol. Monog. 25:345–385.

Stone, A. and Peterson, B. V. 1958. *Simulium defoliarti*, a new black fly from the western United States (Diptera, Simuliidae). Bull. Brooklyn Entomol. Soc. 53:1–6.

Wirth, W. W. and Stone, A. 1956. Aquatic Diptera. *In*: Aquatic Insects of California, by R. C. Usinger. pp. 372–482.

## A RAPID TECHNIQUE FOR EXTRACTING SALIVARY GLANDS FROM LIVE ADULT BLACK FLIES (DIPTERA: SIMULIIDAE)

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Our investigation of the salivary gland proteins of some blood-sucking black flies of the dipterous family Simuliidae required us to devise a technique for the clean, rapid, extraction of large numbers of salivary glands from living flies. Rapidity of procedure was required for completing the dissection of large series in each sample, as well as for minimizing autolytic deterioration of the material during the process prior to quick-freezing storage. Rapid dissection was required also to maintain uniformity of age class in any given sample, because the condition and composition of the salivary glands of these flies undergo changes during the first few days of adult life. We believe that our procedure may be helpful to other investigators of salivary glands of flies.

Several workers have referred to the location of black fly salivary glands (Cox 1938, Krafchick 1942, Smart 1935, Wenk, 1962), some with detailed anatomical descriptions (Gosbee et al. 1969, Wachtler et al. 1971, Welsch et al. 1968). However, removal of these glands is not so well documented. Bennett (1963) described a technique for gland removal in which he first cut the thorax with a pin, pulled on the head, and then extruded the glands through the severed thorax. Poehling (1976) used a similar technique for his study of salivary gland proteins. Here we describe a faster and simpler technique.

*Simulium vittatum* and *S. decorum* adults, reared in the laboratory from larvae, were maintained at 13°C (55°F) and 85% R.H., prior to dissection. Although any number of flies could have been prepared for dissection at one time, we found fifty to be a suitable number to handle easily.

The selected flies were immobilized with a stream of carbon dioxide, enabling us to sex them and to transfer them to an 8 cm × 1 cm glass tube. The test-tube, which had been previously chilled in a freezer, was then dropped into a custom-made ice jacket. This receptacle was made by freezing a styrofoam mug of water around an empty test-tube, and then removing the latter to leave an equal sized cavity in the ice. While the fly-filled test-tube was held in this ice cavity, the flies remained completely immobilized.

For the dissection 1 fly at a time was removed with forceps from the holding tube, and plunged under insect ringer's solution contained within a raised-ring calibration slide. The slide and contained specimen were then placed under a stereoscopic miscroscope at a magnification of approximately 50. For a right-handed worker we found it easiest to secure the fly by the upper abdomen by using forceps held in the left hand. A pair of extra-fine forceps (Dummont #5 Biologie) in the right hand, was used to grasp the head and pull it clear of the thorax at the neck membrance. In this way the salivary glands were drawn out of the thorax and remained attached to the head, still held by forceps in the right hand. The remaining carcass was discarded, freeing the left hand to make the final break between the glands and the head.

The entire dissection process took only 10 sec per fly after it was lifted from the ice jacket.

For our purposes the dissected salivary glands were removed from the slide with a minuten pin mounted on a tooth-pick. The glands were then frozen for later protein analysis.

*References Cited*

Bennett, G. F. 1963. The salivary gland as an aid to identification of some simuliids. Can. J. Zool. 41:947–952.

Cox, J. A. 1938. Morphology of the digestive tract of the blackfly *Simulium nigroparvum*. J. Agric. Res. Washington, D.C. 57:443–448.

Gosbee, J., J. R. Allen and A. S. West. 1969. The salivary glands of adult blackflies. Can. J. Zool. 47:1341–1344.

Krafchick, B. 1942. The mouthparts of blackflies with special reference to *Eusimulium lascivum* Twinn. Annals Ent. Soc. Amer. 35:426–434.

Poehling, H. M., D. I. Wolfrum and V. Neuhoff. 1976. Mikroelektrophorese von Proteinen aus Speicheldrüsen und Hämolymphe verschiedener Simuliidenarten in Polyacrylamidgradientengelen. Ent. Exp. Appl. 19:271–286.

Smart, J. 1935. The internal anatomy of the blackfly *Simulium ornatum* Annal. Trop. Med. Parasit. 29:161–170.

Wächtler, V. K., W. Rühm and U. Welsch. 1971. Histologische und histochemische Untersuchungen an den Speicheldrüsen der Imagines von *Boophthora erythrocephala* de Geer und *Odagmia ornata ornata* (Meig.) (Dipt., Simuliidae). Z. für angew. Eng. 67:189–201.

Welsch, U., K. Wächtler and W. Rühm. 1968. Die Feinstruktur der Speicheldrüse von *Boophthora erythrocephala* de Geer (Simuliidae, Diptera) vor und nach der Blutaufnahme. Z. für Zellforschung 88:340–352.

Wenk, P. 1962. Anatomie des Kopfes von *Wilhelmia equina* L. Zool. Jb. Anat. und Ontog. 80:81–134.

# FIRST RECORDS OF *COELOMOMYCES* AND MERMITHIDS IN MOSQUITOES OF ANGOLA, AFRICA[1]

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The following is a brief account of our present knowledge of the occurrence in Angola of fungi of the genus *Coelomomyces* Keilin and nematodes of the family Mermithidae Braun, important potential biocontrol agents of mosquitoes.

*Coelomomyces africanus* Walker is recorded from southern and central Angola (near Calai and Huambo, respectively) parasitizing larvae of the following anophelines: *An. argenteolobatus* (Gough); *An. cydippis* De Meillon; *An. distinctus* (Newstead & Carter); and *An. squamosus* Theobald. So far as we know, all these 4 mosquito species are new hosts for *C. africanus*.

In southern Angola, *Coelomomyces indicus* Yengar was also found parasitizing larvae and one adult female of a member of the *An. gambiae* Giles complex (most probably *An. arabiensis* Patton), as well as larval *An. squamosus* Theobald.

*An. squamosus* was also found infected by another undetermined species of *Coelomomyces* in southern Angola.

Lastly, a 4th undetermined *Coelomomyces* species was also recorded as a parasite of *Culex argenteopunctatus kingii* (Theobald) and *Cx. guiarti* Blanchard in northern Angola and Cabinda. So far as we know, also both these culicines are new host species for *Coelomomyces*.

Nematodes of the family Mermithidae belonging to yet undetermined genera and species, were also found in southern Angola (ca. Calai) as parasites of the following species of mosquitoes: *An. squamosus* Theobald, *An. theileri* Edwards, *Cx. poicilipes* (Theobald), and *Cx. simpsoni* Theobald. So far as we know, also these 4 mosquito species had not yet been recorded as hosts of mermithid nematodes.

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