

SOME NEW PREPARATION TECHNIQUES USED IN
LEAFHOPPER IDENTIFICATION

DOUGLAS E. BARNETT

Department of Entomology, University of Kentucky
Lexington Kentucky 40506

ABSTRACT

Construction of a microsaw for disjoining the abdomen from the thorax of leafhoppers is described. A method is given for clearing the abdomen by use of a mixture of KOH, ethyl alcohol, and ethyl ether. A method is also described for flushing softened abdominal tissue using either a syringe and/or ultrasonic cleaner.

Leafhopper identification was based almost entirely on external characters in the initial period of leafhopper taxonomy. During the late 1920's and the 1930's the internal male genitalia were recognized to be of considerable taxonomic importance (Lawson 1920, DeLong 1931). The methods used in handling and preparing specimens for taxonomic study have generally been transmitted from one generation of workers to another with little change (Eltringham 1930, Beirne 1956). Efficiency in handling and preparing study material should be improved but not at the sacrifice of the material, and any change in methods or procedure should leave the study material in a state which will render it of maximum benefit to the current worker and subsequent workers.

Most leafhopper taxonomists usually begin preparing a specimen by disjoining the abdomen from the thorax (Beirne 1956, Morrison 1973). In the majority of cases for most leafhopper groups this technique works admirably. Occasionally, however, the muscle and visceral tissues become very hard and firmly connect the abdomen to the thorax. When this condition exists even the most careful dissection cannot disjoin the 2 without considerable or total damage to the specimen. Destruction of these specimens can be prevented by using a genitalia microsaw. This device can easily be constructed from a doorbell (Fig. 1) which can be purchased at most hardware stores. A piece of plastic wire insulation is attached with epoxy glue between the vibrating armature of the doorbell and the center tube of 2 closely fitting concentric metal tubes. The plastic tubing transmits the motion of the vibrating arm and acts as a differential. A minuten pin glued in the opposite end of the center tube acts as the cutting point. The outer metal tube may be glued inside a pencil casing for comfort in handling and for added strength. The doorbell circuit is connected via a foot switch to a 6-volt power source. This device can cut or saw, a facility which is easily controlled by finger tip pressure. The depth of stroke can be controlled at the lever arm.

Leafhopper systematists commonly use a 10% KOH solution to soften muscle tissues and clear the darkly tanned areas of the abdomen and/or genital capsule (Knight 1965). However, in reality the strength of the KOH solution is frequently much stronger, having been increased rather inadvertently in an attempt to maintain the strength of the solution.

Often systematists unknowingly use a KOH solution as high as 50%-80%. Even at these concentrations, it may require 24, 48 or more hours to complete the clearing process for some species; hence some faster technique is desirable.

A part of the problem of clearing specimens lies in getting the clearing solution in contact with the tissues to be cleared. Natural waxes and oils of the exoskeleton present a barrier to contact and entrance of the KOH solution. Ethyl alcohol is a good solvent for the waxes and oils and, when used as the solvent to make the KOH solution, it was found to wet and penetrate rapidly. In an ordinary 10% KOH solution the genitalia frequently float, but in an alcoholic KOH solution they sink, and clearing is immediately initiated on all sides. One can further speed clearing process by adding a little ethyl ether to the alcoholic KOH clearing solution. Various combinations of water, ethyl alcohol and ethyl ether were tested. The one that cleared the best and maintained its relative integrity was a clearing solution made by combining 5 ml of ethyl ether, 50 ml of 70% alcohol, and 50g of potassium hydroxide. More than 5 ml of ether can be added to further speed the process, but the stock solution has a shorter shelf life. On more difficult groups such as certain species of *Scaphoideus* which require 2-3 days in ordinary 10% KOH, it takes only 1-2 hr. with the alcohol-ether-KOH solution. On light yellow or green leafhopper species, such as species of *Draculacephalia*, clearing begins instantaneously, with the forewings completely clear within 40 sec and the genitalia clear within 1-3 min. When necessary to maintain the specimens in the clearing solution for more than a few minutes, it is best to cover the genitalia tray to prevent evaporation of the clearing solution. On all species compared thus far, this procedure causes no reorientation of the genitalia or armature.

Once the muscle and visceral tissue is softened, it must be removed. The technique used by most taxonomists is to prod the abdomen and genital capsule until the contents are expelled. This technique is often laborious, requires several repeated sessions, and has the disadvantage of reorienting the genital structures or breaking off pieces of the genital armature and/or

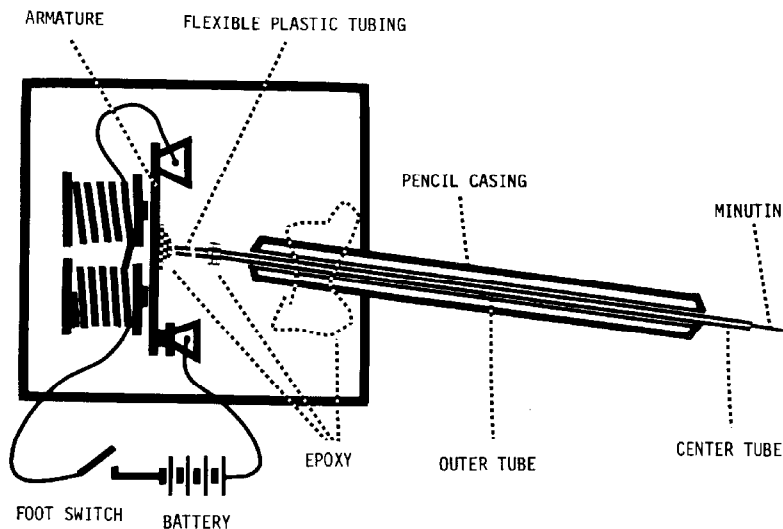


Fig. 1. Genitalia microsaw.

setae. To circumvent these difficulties, a device (Fig. 2) was constructed from a hypodermic syringe that could be inserted into the anterior opening of the abdomen and a fine stream of 70% alcohol used to flush out the contents of the abdominal cavity. The abdomen can be held in place with the heel of the crook near the point of the needle. The clearing solution discussed earlier leaves the visceral contents in a granular state, and the contents flush out easily. Insect material that has been recently collected or has been maintained in alcohol for a long time is more difficult to soften. With this material the pressure on the syringe may be increased, and the fine stream of jetted water can be used to cut away clinging material.

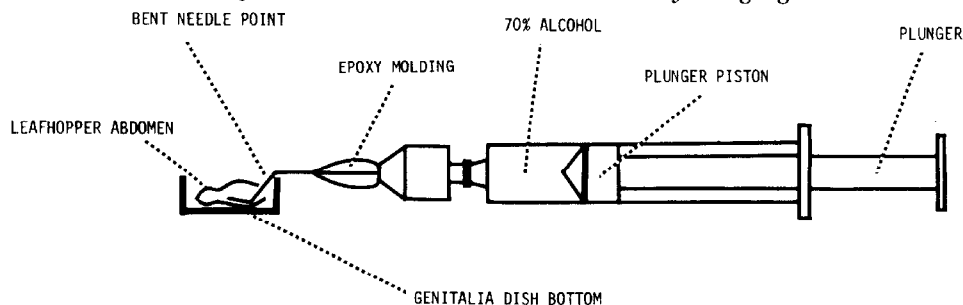


Fig. 2. Modified hypodermic syringe shown with needle inserted into leafhopper abdomen.

The time required to remove the abdominal contents can be shortened by placing the KOH processed material in the tank bottom of a device which produces ultrasonic sound such as the Branson Ultrasonic device. A quarter-inch of water should be added to facilitate vibration conduction. Most of the contents will be removed from the abdomen and genital capsule after 5-10 sec of operation. Setae and all structures remain intact. In a few specimens, some debris may remain in the abdomen or genital capsule. This debris can be removed with the syringe method discussed earlier. Treatment with ultrasonic sound saves considerable time when masses of material need to be processed.

LITERATURE CITED

- BEIRNE, B. P. 1956. Leafhoppers (Homoptera: Cicadellidae) of Canada and Alaska. *Can. Ent.* 88:(supl. 2) 1-180.
- DELONG, D. M. 1931. A revision of the American species of *Empoasca* known to occur north of Mexico. *USDA Tech. Bul.* 231:1-59, 11 illus.
- ELTRINGHAM, H. 1930. *Histological and illustrative methods for entomologists.* Oxford, Clarendon Press. 139p.
- KNIGHT, W. J. 1965. Techniques for use in the identification of leafhoppers (Homoptera: Cicadellidae). *Ent. Gazette* 16:129-36.
- LAWSON, P. B. 1920. The Cicadellidae of Kansas. *Kans. Univ. Sci. Bul.* 12:5-306, Pl. II-XVII.
- MORRISON, W. P. 1973. A revision of the Hecalinae (Homoptera: Cicadellidae) of the Oriental region. *Pacific Insects* 15:(3-4) 379-438.