



Anglian Lepidopterist Supplies

Specialising in
moth traps and
related equipment

MOTH DISSECTION GUIDE

GENITALIA EXAMINATION

The beginner usually concentrates on the macro-moths (larger moths) as illustrated in Bernard Skinner or Paul Waring's moth guides. With the aid of these 'lepidopterist's bibles', the majority of moths caught will be readily identifiable. However, even within this well studied group there remain a few species or species groups which can not be easily identified, these we will term the **aggregate species**. The Minor's, Ear's, Grey/Dark Dagger, Lesser Common/Common Rustic and November, Pale November and Autumnal Moth are the classic examples. For these species it is necessary to resort to genitalia examination, usually via a full dissection, to firmly establish the identity of the moth. Although many people will be happy to record the moths as Common Rustic agg. for example, some people may wish to positively identify specimens. Moths can be kept alive for periods of several days if placed in the refrigerator, allowing second opinions to be taken. However, for the species mentioned above, the only sure way to identify the moth is to kill it, and perform a dissection. Usually the moths will die quickly if placed in the freezer. Alternatively a simple killing jar can be constructed from an old coffer jar. Place cotton wool in the bottom of the jar, add a small quantity of ethyl acetate, and then place a round cardboard disc over the cotton wool to prevent moths coming into contact with it. Moths placed in the jar will quickly become stationary. However, they should be left for at least 20 minutes to ensure they are dead (longer with larger specimens). The moths can be removed from the jar and should be sufficiently relaxed to set immediately. Ethyl acetate will dehydrate moths, and hence specimens should not be left in the jar for too long or they will become stiff, particularly so with the pugs and the micro-moths. Alternatively, moths can be killed using 0.88 ammonia. This does not stiffen moths to the same extent as ethyl acetate, and is a better killing agent for most micros and geometers, but not as effective for the larger macro-moths. However, it does tend to discolor green or yellow moths by turning them brown with time! Hopefully, by retaining positively identified specimens, new field marks allowing identification of live moths may be found and referral to your reference collection will make the future identification much easier for those 'tricky' species.

The reproductive organs are contained in the lower half of the insect's abdomen, the males usually in the 8th or 9th segment and females in the 7th, 8th and 9th segments. The features of the female genitalia are more difficult to work from, but the features of the male genitalia are much larger and hence easier to work with. To inspect the genitalia, you should begin by removing the abdomen of the specimen. This can be done with freshly dead specimens or with long dead, dry specimens. The abdomen is removed by applying upward pressure to the tip of the abdomen which should then snap off. Occasionally, the hind wings of set specimen will also be removed. These can be glued back into place with wood glue (PVA), which dries clear.



Place the removed abdomen in 10% Potassium Hydroxide solution (KOH). The solution can then be placed on a 40W light bulb to provide bottom heat. The abdomen should be left in the warm solution until soft, which can take

anything up to half an hour for larger specimens, but typically only about 10 to 20 minutes for the smaller moths. Do not attempt to provide any more heat than that provided by a 40W bulb. This will result in the solution boiling, and spitting which can be dangerous. With the abdomen now softened, it can be transferred to a watch glass or petri dish filled with a little water. The addition of a few drops of alcohol will lower the surface tension of the water and make working with the plate easier. The genitalia plate can then be gently stroked or 'teased' out. This is achieved by holding the top (open end) of the abdomen with either a dissection needle or a seeker. A seeker is then moved down the abdomen towards the tip with gently stroking motions. You should try and keep the specimen covered with fluid while extracting the genitalia plate to prevent air bubbles entering the organs. As you stroke the abdomen, the genitalia plate will pop out of the tip of the abdomen.



The plate is fairly robust and can then be gently held in place while remaining debris is cleaned away. The ideal tool for cleaning the genitalia and abdomen can be formed from the pinfeather of either a snipe or woodcock. (Have a word with a local game dealer and you can sometimes obtain them).

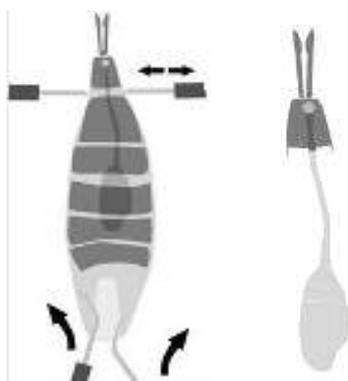
Mounted on a suitable piece of dowel, this forms a superb brush for cleaning debris away. However, make sure you have rinsed the plate in a petri dish of alcohol/water before using your brush. Potassium hydroxide will dissolve it! Cleaning and examination can be done by eye for larger moths, but a binocular stereo microscope will make life very much easier and is essential for the smaller specimens.

The cleaned plate can now be transferred to a petri dish of water and given a final rinse before examination. A number of standard guides give details of the genitalia of difficult species such as the Pierce & Metcalfe guides and the Moths of Great Britain and Ireland series published by Harley Books. In addition we have set up the Dissection Web Site which covers a large percentage of the British moths, male and female. See: <http://www.dissectiongroup.co.uk/>

For female moths a slightly different technique is required. The tip of the genitalia plate should be examined in the first instance to ascertain which sex of moth you have. You will either see the claspers of the male moth or the tip of the ovipositor in female specimens.

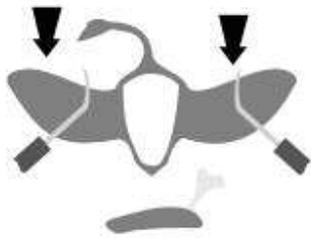


If the moth is a female, the skin of the abdomen should be carefully cut all the way around the abdomen about three segments back from the tip. This can be done with a dissecting needle or extra fine tweezers. The whole tip of the abdomen is then removed. The remaining collar of skin can then be cut length wise and peeled away from the genital plate. If you try and squeeze the female genitalia out of the tip of the abdomen, usually the ductus bursae will break off from the rest of the genitalia making determination much harder and in some cases, impossible.



If you wish to preserve the genitalia as a reference sample, then you should prepare a permanent mount.. Firstly, air bubbles will invariably be present somewhere within the genitalia structure. This should carefully be removed by use of the dissecting needles. The structure consists of a set of hollow tubes, and the air bubbles can be maneuvered around within these tubes until they arrive at an opening when they can be removed. If smaller bubbles cannot be removed don't persist as damage could occur, leave them there as in most cases they will disappear once in the mounting fluid.

Transfer the plate into isopropyl alcohol (IPA). This is a dehydrating agent. The plate should be held in position (e.g. with the claspers apart for male moths) and the plate will gradually harden and set.



At this stage the plate is fragile and care should therefore be exercised when moving the plate around. Next, the plate can be stained in an alcohol based stain (if required)-cholorazal black or mercurochrome being the most widely used. Staining will often enhance subtle, difficult to see features. However, caution is urged since over staining will obliterate those same features.

The plate is then transferred into Euparal essence and left to 'wet'. A few drops of Euparal can then be placed on a clean slide (give them a wipe with Isopropyl alcohol to remove grease first) and the plate is lowered into position and sealed with a cover glass. When placing cover slips, the rear edge should first be placed in contact with the slide. The front edge is then gently lowered. This helps to prevent air bubbles forming in the mountant. The slide and the specimen should be clearly labeled with a unique number that ties the two together. A data label giving the species details should also be attached to the specimen.

QUICK GUIDE:

1. Remove the abdomen and place in 10% KOH
2. Gently heat for required time.
3. Remove abdomen to Petri dish with a small amount of water
4. Gently remove scales with tweezers, paint brush or Woodcock/Snipe pin feather if available. Stain if required, don't forget to wash off excess stain in water.
5. If a male, tease out the genital capsule.
6. If a female, gently cut away the abdominal skin.
7. Place genitalia in Isopropyl Alcohol to dehydrate. Don't leave too long.
8. Place in Euparal Essence ('wetting')
9. Transfer to Euparal on microscope slide.
10. Apply microscope cover slip.
11. Make a data label.
12. Keep flat to dry out.

MORPHOLOGY

