

Preparing, labeling, and shipping Hexapoda

Supplies and Equipment

Some of the equipment needed to preserve and observe insects will be provided by us:

- vials
- ethanol
- shellac
- points for point mounting (we have point punches and Bristol boards for students to use in lab)
- size 2 pins
- label paper, 3×5 cards and cellophane envelopes for Odonata
- Schmitt box(es)
- barcode labels
- clay block
- stereo- and compound microscopes (cannot leave the classroom)
- soft-tipped forceps (available from Bioquip as product #'s 4748 or 4750)
- waterproof pen for labels; a good option from Bioquip is the size 005 Pigma pen (black) # 1154E. You can probably find these or their equivalent at an art supply store or even the PSU bookstore.
- scissors for cutting labels

Slide-mounting and double-mounting supplies are available upon request, as is acetone for Odonata. We have collecting equipment that students can check out for short periods of time. This gear will also be available to everyone during field trips. Sharing is encouraged with this equipment:

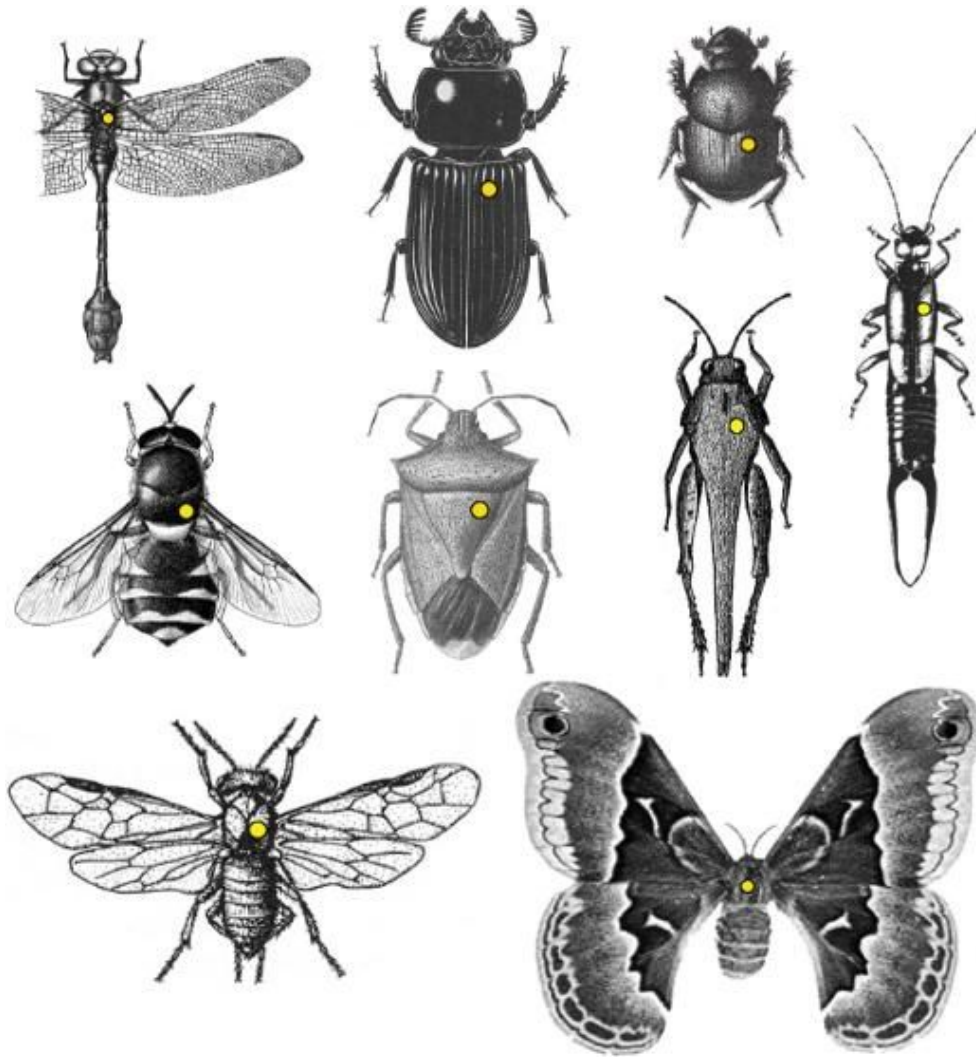
- UV/Hg-vapor lights with white sheet
- Malaise trap
- aquatic net
- Winkler extractors, litter sifters
- Berlese funnel (cannot leave Headhouse III)
- sweep net
- ethyl acetate for kill jar
- spreading board

We'll show you make your very own:

- aspirator
- kill jar

Preparing Hexapoda

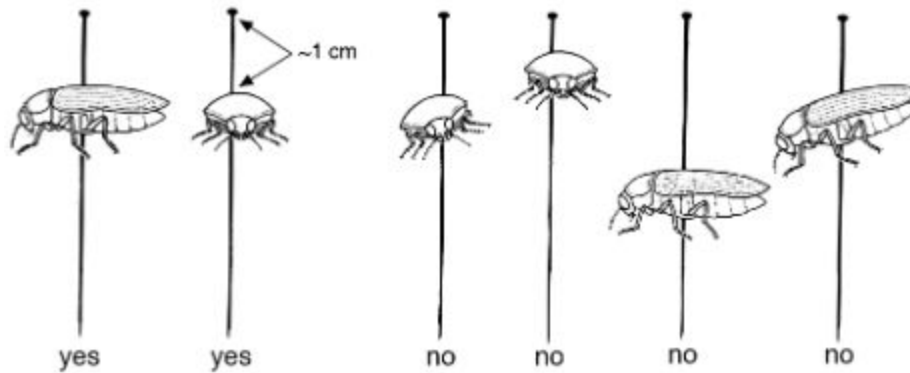
Preamble.— A rough guide to preservation preferences—which taxa are pinned vs. slide-mounted, *etc.*—is provided in the Appendix. See the end of this document.



Pinning.— Pinning is the best way to preserve hard-bodied, medium to large hexapods. One should use specially made insect pins, rather than common pins used in sewing and other crafts. Insect pins range in size from 000 (VERY thin and mostly unmanageable) to 7 (very thick and longer than most pins). We recommend sizes 1, 2 (especially), and 3 for use in this class. The best way to pin an insect is to:

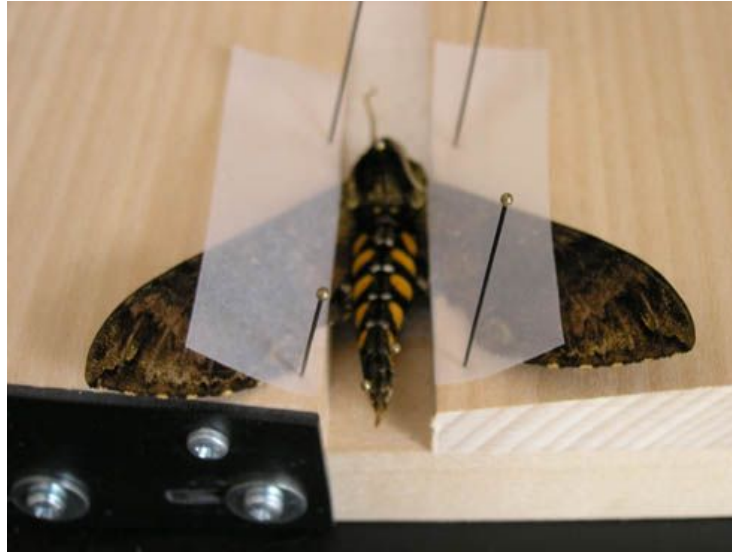
1. hold the dead insect between your index finger and thumb
2. pass the pin vertically through the mesonotum, slightly to the right of center (except in Lepidoptera, which gets the pin through the center of the mesonotum), such that it emerges near the right mid coxa

- slide the pin far enough through the body such that approximately 1 cm of pin is left between the insect and the pin head – this forms the “handle” for manipulating specimens. Pin placement often varies slightly by taxon (see above).

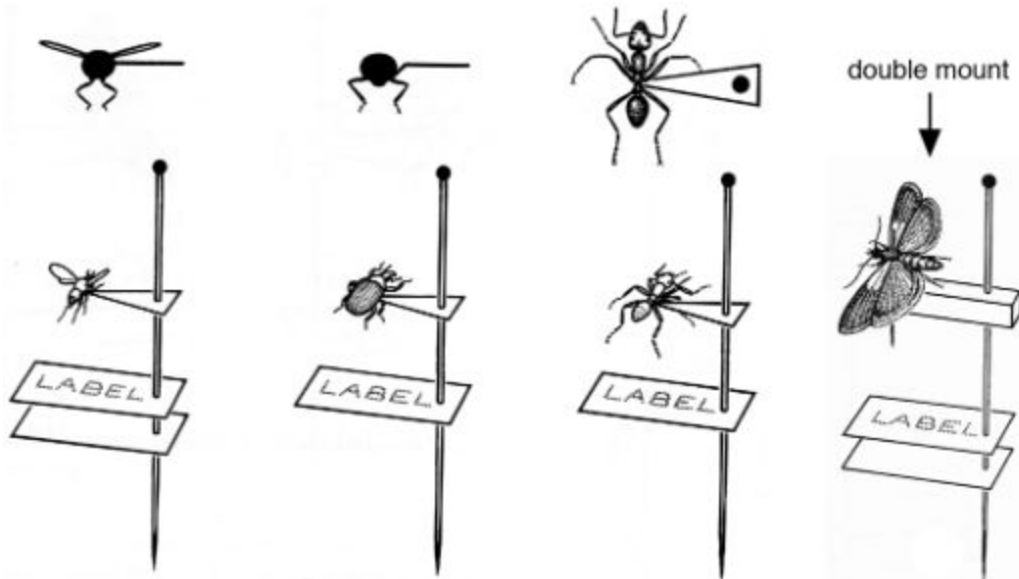


Lepidopterans, especially the larger species, need to have their wings spread by using a spreading board (image source: [Goliathus](http://utahbugclub.org/layout/pdf/thebigshow.pdf)) (see also this demonstration: <http://utahbugclub.org/layout/pdf/thebigshow.pdf>):





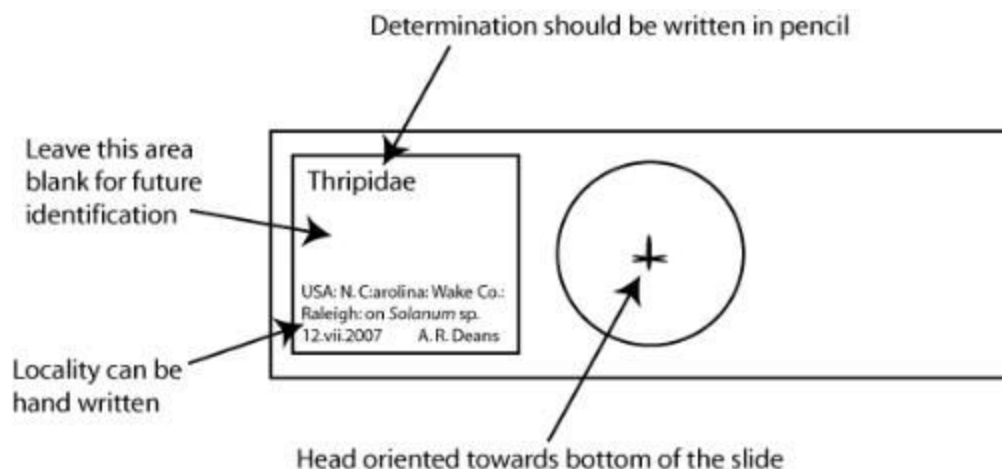
Pointing.— Smaller insects are usually pointed on a piece of acid-free cardstock or Bristol board. Place a drop of alcohol-soluble glue, such as Gelva or shellac, on the tip of a point that's already been pinned. Touch the point tip to the mesosternum of the insect, usually between the fore and mid coxae. The pointed insect should be oriented in a similar position to that of a pinned insect:



Double mounts.— Microlepidopterans need to be double mounted using minuten pins. Essentially this is the same as pinning, except one uses a very small pin (the minuten) to mount the lepidopteran. That mount is then affixed to a normal-sized insect pin via a small piece of foam (see above, right). A more detailed description is provided by [Chris Grinter](#).

Alcohol.— Hexapods preserved in >95% alcohol are best for DNA extraction, especially if they are kept cold. Unfortunately, high concentrations of alcohol also tend to make specimens a bit fragile, by dehydrating them. Concentrations below 70% are generally not recommended, as specimens have a better chance of rotting. We use 80% ethanol in the lab, which works well for nearly any kind of hexapod. Soft-bodied hexapods must *always* be preserved in alcohol, rather than being pinned or pointed. Adult Lepidoptera should *never* be preserved in ethanol, as the scales will detach.

Slide Mounting.— See [separate slide mounting handout](#).



Locality Labels

Labels can take many forms, but you generally want to adhere to the following formats:

Pinned Specimens

A	B	C
USA: NC: Wake Co.: Raleigh: 35.788,-78.632 UV light in residential area 12.vi.2012 A.R. Deans	USA: N. Carolina: Wake Co.: Raleigh: Schenk Forest: 35.81627, -78.70760 coll. 1.v.2012 A.R. Deans ex: <i>Spodoptera</i> sp. on <i>Digitaria</i> sp. (Poaceae) eclosed 5.v.2012	USA: NC: Durham Co.: Hebron Rd. at Old Oxford Rd.: FIT in fallow field: 36.060, -78.869, 98 m 12–15.vii.2012 A. R. Deans

- Labels always begin with the country (sometimes in **bold** or ALL CAPS) and continue with finer scale details of the locality: **USA:** PA: Centre Co.: *etc.* One should always include the lat/long, which can now be estimated in programs like Google Earth. Using decimal based degrees is preferred over minutes and/or seconds, as they are easier to database.
- The date should be formatted such that the month is in lowercase Roman numerals (e.g., October would be 'x'): 12.x.2010 or 11–12.x.2010 or 11.x–12.xi.2010
- The collector's name(s) should also be included, as should the collecting method. Common abbreviations include: MT=Malaise trap, YPT=yellow pan trap, FIT=flight intercept trap, SS=screen sweep. Collectors' names are sometimes followed by "leg.", which is short for the Latin 'lego', to gather or collect.
- A sans-serif font (like Arial Narrow) makes the label more readable when the size gets small. Most people use 4 pt for the font size. The finished label should be informative, with a minimal amount of abbreviations, but also reasonably small in size. The information should also be presented in a symmetrical label (*i.e.*, minimize whitespace).
- Labels that seem to be excessively large (label B) can be cut into two labels. Specimens are prone to multiple labeling from future studies (voucher label, determination label, accession numbers, barcodes, *etc.*), so it's desirable to keep the label number to a minimum.
- Use cotton rag, acid free cardstock for printing labels.

Fluid-preserved Specimens

D	E	F
USA: NC: Wake Co.:Raleigh: UV in residential area 35.788,-78.632 12.vi.2012 A.R. Deans	USA: N. Carolina: Wake Co.: Raleigh: Schenk Forest: 35.81627, -78.70760 coll. 1.v.2012, eclosed 5.v.2012 A.R. Deans ex: <i>Spodoptera</i> sp. on <i>Digitaria</i> sp. (Poaceae)	USA: NC: Durham Co.: Hebron Rd. at Old Oxford Rd.: FIT in fallow field: 36.060, -78.869, 98m 12–15.vii.2012 A. R. Deans

- Same suggestions apply to labels for fluid-preserved specimens, but one can and should make his/her labels slightly larger (5 or 6 pt) and more elongate. Small labels act like blades and chop up wet-preserved specimens, which are usually relatively soft-bodied.

Determination Labels

Determination (“det”) labels can also vary in their appearance, but it’s important that they include the taxon name(s), the name of the determiner, and the year (or date) the determination was made. Again, det labels for fluid preserved specimens (label H) should be slightly larger and more elongate:

G	H
Evaniidae	Evaniidae (Hymenoptera)
det. AR Deans 2013	det. AR Deans 2013

Preparing Specimens for Shipment

Adapted from a summary by L. L. Deitz, NC State University.

Schmitt boxes

1. select strong, tight fitting box with suitable pinning substrate – not soft Styrofoam
2. remove fumigant, slides, vials
3. place cardboard on top of Schmitt box (under the lid)
4. make sure pins are well distributed in empty areas of box and are fixed well in substrate
5. brace genitalia vials, large insects, and rotating insects
6. use some type of filler- foam, cotton, newspaper – on top of the cardboard
7. place Schmitt box(es) within a larger package
8. fasten address labels on outside Schmitt box as well as on outside of package
9. ship only 1 box in a larger package to prevent 2 boxes from hitting each other or ship 2 boxes together only if they are securely tied together
10. package with 2 inches on each side between Schmitt box and bigger package, filled with packing material







Slides





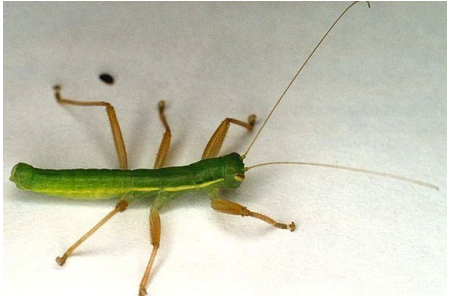
1. make sure they are double boxed similar to Schmitt box
2. pack slides with coverslips facing each other, form a “brick”
3. place cardboard between opposing coverslips
4. if in slide box, place packing material between top of slides and lid to prevent breakage







Vials





1. avoid air bubbles and “burp” vials
2. wrap each vial in paper towel or bubble wrap to separate from other vials
3. shipping ethanol is now heavily regulated and often illegal; consult with safety expert(s) and use UPS (possibly) or FedEx





Appendix. Adult Hexapod Preparation Recommendations





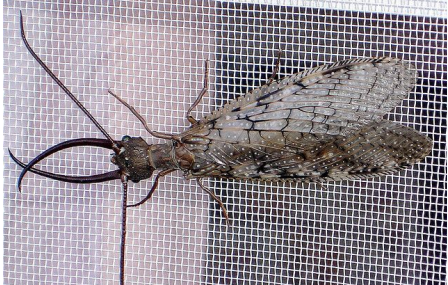
Taxon (common name)	Typical habitus	How to preserve them
Protura (coneheads)	 Andy Murray	Traditionally these hexapods are slide-mounted. For this class, they can be preserved in vials with ethanol, propylene glycol, or glycerol.
Diplura	 Marshal Hedin	Traditionally these hexapods are slide-mounted. For this class, they can be preserved in vials with ethanol, propylene glycol, or glycerol.
Collembola (springtails)	 Andy Murray	Traditionally these hexapods are slide-mounted. For this class, they can be preserved in vials with ethanol, propylene glycol, or glycerol.
Archaeognatha (bristletails)	 Henry Lydecker	These insects are preserved in vials with ethanol, propylene glycol, or glycerol.
Zygentoma (silverfish, firebrats)	 Jean-Raphaël Guillaumin	These insects are preserved in vials with ethanol, propylene glycol, or glycerol.
Ephemeroptera (mayflies)	 Jean-Jacques Boujot	These insects are preserved in vials with ethanol, propylene glycol, or glycerol.





<p>Odonata (dragonflies, damselflies)</p>	 <p>Frost Entomological Museum</p>	<p>Odonata should be kept alive in glassine envelope or paper triangle, until they can be euthanized and subsequently soaked with acetone. See the Frost SOP on Odonata Preparation.</p>
<p>Phasmatodea (walking stick, stick insects, leaf insects)</p>	 <p>Norman Walsh</p>	<p>These insects should be pinned in a way that minimizes their size (legs and antennae close to body). Never spread their wings.</p>
<p>Orthoptera (grasshoppers, crickets, katydids)</p>	 <p>Andreas Kay</p>	<p>These insects should be pinned in a way that minimizes their size (legs and antennae close to body). The gut of large specimens should be removed prior to pinning (sharp forceps under the posterior edge of pronotum). Left wings could be spread. Crickets (Gryllidae) go in ethanol.</p>
<p>Grylloblattodea (ice crawlers)</p>	 <p>Joyce Gross</p>	<p>These insects are preserved in vials with ethanol, propylene glycol, or glycerol.</p>
<p>Mantophasmatodea (heel walkers)</p>	 <p>P. E. Bragg</p>	<p>These insects are preserved in vials with ethanol, propylene glycol, or glycerol.</p>

Dermaptera (earwigs)	 Mick E. Talbot	<p>These insects should be pinned/pointed. Never spread their wings.</p>
Mantodea (mantises)	 Karyn Christner	<p>These insects should be pinned/pointed in a way that minimizes their size (legs and antennae close to body). Never spread their wings.</p>
Blattodea (cockroaches, termites)	 Martin Grimm  Stevenw12339	<p>Non-isopteran Blattodea (cockroaches): These insects should be pinned/pointed in a way that minimizes their size (legs and antennae close to body). Never spread their wings. Genitalia are important for determination, so make sure they're exposed.</p> <p>Isopterans (termites; bottom image): These insects are preserved in vials with ethanol, propylene glycol, or glycerol.</p>
Embioptera (webspinners)	 Bill & Mark Bell	<p>These insects are preserved in vials with ethanol, propylene glycol, or glycerol.</p>
Zoraptera	 David Maddison	<p>These insects are preserved in vials with ethanol, propylene glycol, or glycerol.</p>

Plecoptera	 Bernard DuPont	<p>These insects are preserved in vials with ethanol, propylene glycol, or glycerol.</p>
Thysanoptera (thrips)	 Mick Talbot	<p>Traditionally these hexapods are slide-mounted. For this class, they can be preserved in vials with ethanol, propylene glycol, or glycerol.</p>
Psocodea (bark lice, book lice, parasitic lice)	 Ken Schneider  Ian Boyd	<p>Traditionally these hexapods are slide-mounted. For this class, they can be preserved in vials with ethanol, propylene glycol, or glycerol.</p>

<p>Hemiptera (true bugs, scale insects, aphids, hoppers)</p>	 <p>NY State IPM Program</p>  <p>Mick E. Talbot</p>	<p>These insects should be pinned/pointed.</p> <p><i>Exception:</i> Soft-bodied hemipterans (e.g., Aphididae, Coccoidea (scales)) are traditionally slide-mounted. For this class, they can be preserved in vials with ethanol, propylene glycol, or glycerol.</p>
<p>Mecoptera (scorpionflies)</p>	 <p>Orest Shvadchak</p>	<p>These insects should be pinned/pointed. Never spread their wings.</p> <p><i>Exception:</i> Boreidae (snow scorpionflies) should be preserved in vials with ethanol, propylene glycol, or glycerol.</p>
<p>Diptera (true flies, gnats, mosquitoes)</p>	 <p>Troup Dresser</p>	<p>These insects should be pinned/pointed in a way that one can view all sclerites (<i>i.e.</i>, legs pulled away from body). Wing veins and bristle patterns are important for diagnosis.</p> <p><i>Exception:</i> Small, dainty flies, with soft cuticle (e.g., Cecidomyiidae) should be preserved in vials with ethanol, propylene glycol, or glycerol.</p>

Siphonaptera (fleas)	 <p>Mark Yokoyama</p>	<p>Traditionally these hexapods are slide-mounted. For this class, they can be preserved in vials with ethanol, propylene glycol, or glycerol.</p>
Lepidoptera (moths, butterflies)	 <p>Andy Reago & Chrissy McClarren</p>	<p>These insects should be pinned or double-mounted, depending on their sizes. Wings should be spread, as color patterns and wing venation are important.</p>
Trichoptera (caddisflies)	 <p>Macroscopic Solutions</p>	<p>These insects are preserved in vials with ethanol, propylene glycol, or glycerol.</p>
Neuroptera (lacewings, antlions, mantisflies)	 <p>Mick E. Talbot</p>	<p>These insects should be pinned/pointed in a way that minimizes their size (legs and antennae close to body). Never spread their wings.</p>
Megaloptera (dobsonflies, fishflies, alderflies)	 <p>Ronald Orosz</p>	<p>These insects should be pinned in a way that minimizes their size (legs and antennae close to body). Never spread their wings.</p>

<p>Raphidioptera (snakeflies)</p>	 <p>Tim Haye</p>	<p>These insects should be pinned in a way that minimizes their size (legs and antennae close to body). Never spread their wings.</p>
<p>Strepsiptera (twisted-wing parasites)</p>	 <p>Biodiversity Heritage Library</p>	<p>Traditionally these hexapods are slide-mounted. For this class, they can be preserved in vials with ethanol, propylene glycol, or glycerol. An infected host (e.g., a vespid wasp) should be preserved according to that taxon's suggested method.</p>
<p>Coleoptera (beetles)</p>	 <p>Gilles San Martin</p>	<p>These insects should be pinned/pointed. Ventral characteristics of the thorax are important for diagnosis, as are features of the legs, so make sure they are observable. Never spread their wings.</p>
<p>Hymenoptera (sawflies, wasps, ants, bees)</p>	 <p>Patrick K59</p>	<p>These insects should be pinned/pointed.</p> <p><i>Exception:</i> Small, dainty hymenopterans, with soft cuticle (e.g., most Chalcidoidea) should be preserved in vials with ethanol, propylene glycol, or glycerol.</p>