Fluid preserved Invertebrate Imaging

- Overview.
- •Preparations.
- •Methods and Techniques.
- •Ethanol, Water, Oil. Glycerin

This is not rocket science.



Imaging in fluid is quiet simple and by following just a couple basic laws of physics you can image like a pro!

Preparation.

Understand your goal and prepare your specimen (s) accordingly. The failure to correctly prepare will result in a poor image.

Some real basic stuff! Determine what you are using the image for,

- Clean the specimen if needed before you set it up for imaging.
- •Make sure the solution you use is clean.
- Select your background before you start shooting
- •Color check with a gray card if you want correct color as a reference in a series.
- •Maintain as little fluid over the specimen as is practical. (The more <u>stuff</u> the optics have to image through to get to the subject the less resolution you will have.)
- •Select the proper lighting to yield the desired effect (lighting determines what you will see in the image)
- •Just use common sense!

What I have learned while shooting in Ethanol.....

Cut the ethanol to 70% Hi proof alcohol is too volatile and evaporates too quickly.

I use clear round containment vessels generally because they afford the most illumination variants. Corners of a vessel can produce shadows and reflect / refract light

Small vessels produce a smaller meniscus. The focal point changes faster as the solution evaporates so use a larger diameter containment vessel.

The more crud (fluid, glass etc.) you have on the top of the subject, the less resolution you will have in the final image.

Lighting is Key! Invest the time to get it right, it will pay off in the long haul.

Don't be afraid to to step\out of the box and experiment. Film is cheap these days.

Everyone likes to image genitalia.

3 dimensional Normally very small Never stays where you want it. Often a mix of translucent and opaque.

Positioning the material:

Type or not?

KY

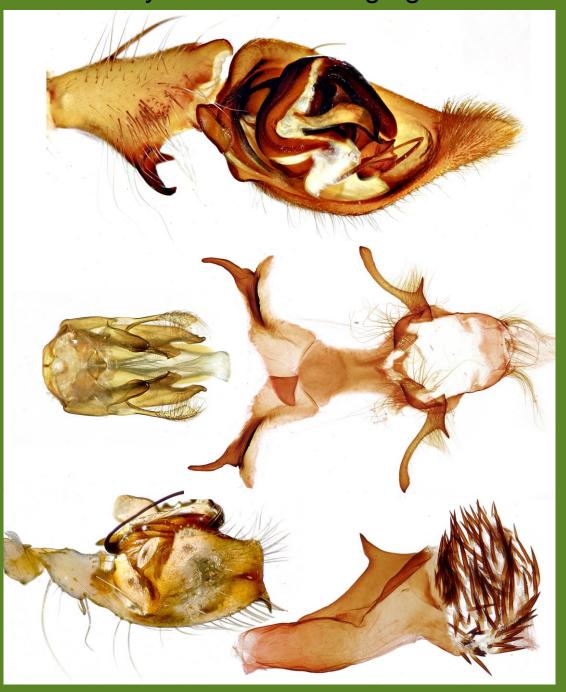
Hand Sanitizer

Water based gels

Glycerin (jelly)

Glass mounted (types)

Put the specimen<u>on</u> the fixative not in it!



Select the correct mounting material for the specimen.

Dolomedes Palp AMNH 1999

Single exposure, 70% solution, transmitted and oblique illumination (flash) KY sticky mount.



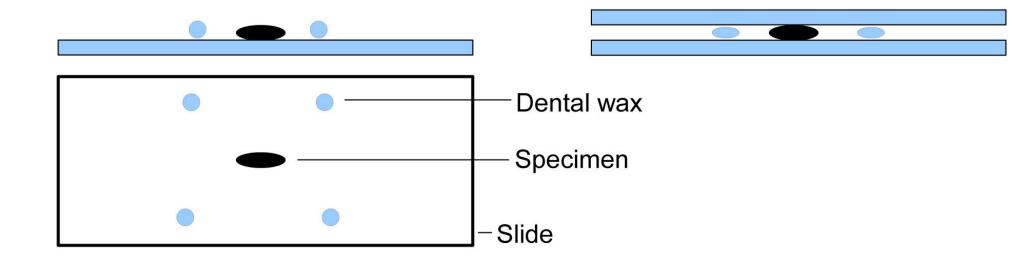
Reproductive pouch



Jon Coddington SI 2010



Very simple wax and glass prep typically for types.



Stained Colombolla Type in ethanol. Wax and glass temporary mount Adelaide Australia



Stained Colombolla Type at 100%

Through 2mm of 70% ethanol and a microscope slide



KY jelly, fun for the whole family but use with caution.

When using KY as a stabilizing mount, do not sink the specimen in the KY. Place it on top.

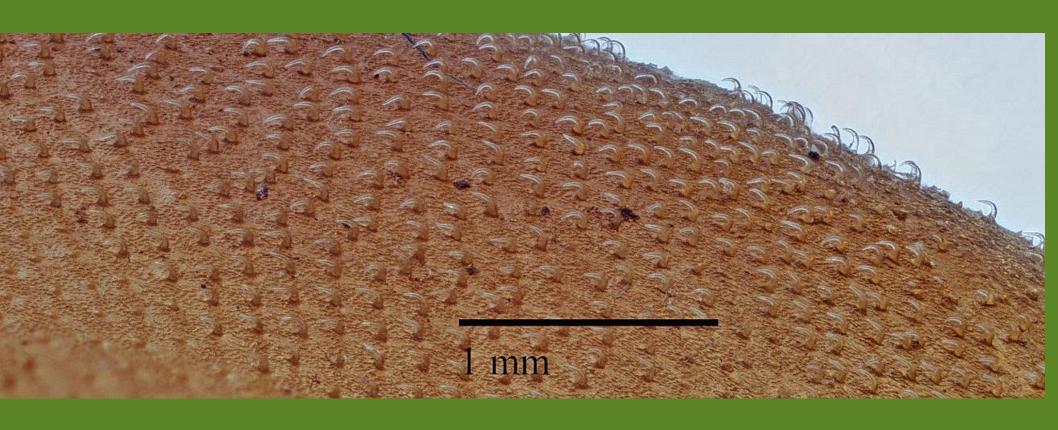
You can let the KY tack up before mounting and Use a small amount

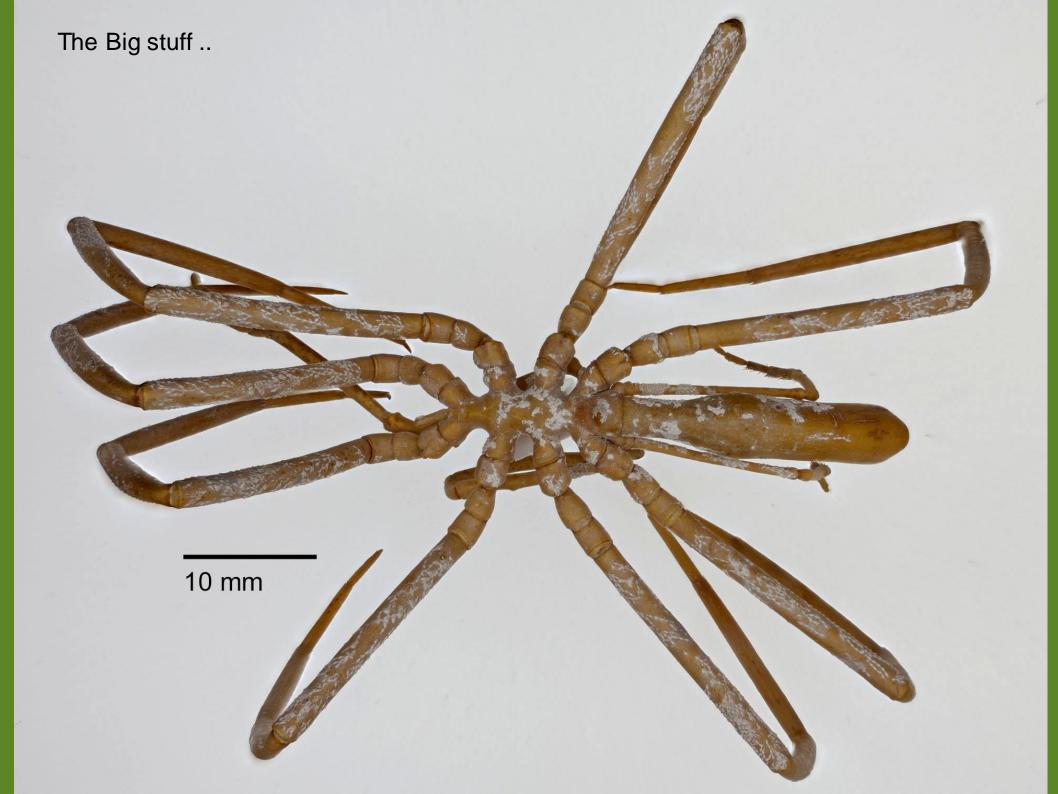
Don't let your specimen dry up in the KY it will be a sticky mess.

KY Contains lanolin and other things meant for human skin it It will contaminate types!

Millipede Ventral 70% Ethanol. Photo Mike Medrano at Kelly Millers lab NM (KY)











Why we need high resolution images....



Imaging trough a containment vial.

1999-2000 AMNH imaging a spider in the vial experiment.

A specimen was selected. The vial was placed in a clear container propped up on one end with wax and covered with 70% ethanol. No diffusers used only a combination of transmitted and oblique flash illumination.

These images are single exposures and the backgrounds have <u>not</u> been knocked out.



This image was taken at 2X through the ethanol bath and the vial and the ethanol inside the vial



Same spider, same vial set up now at 4X mag.



Imaging Amber in Solution. or Dancing to The Masochism Tango

At times, imaging inclusions in amber is like trying to get a clear image through yellow mud within a house of mirrors.

If the amber is not fractured you can simply use J&J baby oil as submersion solution. You can affix the amber with dental wax so that it won't show in the image.

If the amber is fractured you want to use glycerin as it can be dissolved easily in clean water.

If you cut a flat directly beneath the inclusion, light can be transmitted right through the piece back lighting the subject cleanly and evenly..

Lightning is <u>the</u> single most critical component when it comes to imaging inclusions in amber.

All images are single exposures Taken with a 6.0 MP Kodak DCS 660 camera 1999. Fiber Optic flash illumination.

Pollen grains drifting from flower.

Illuminated from the back, top right and a raking light from the upper left



Legume flower with pollen back light



Fungus gnat with eggs.



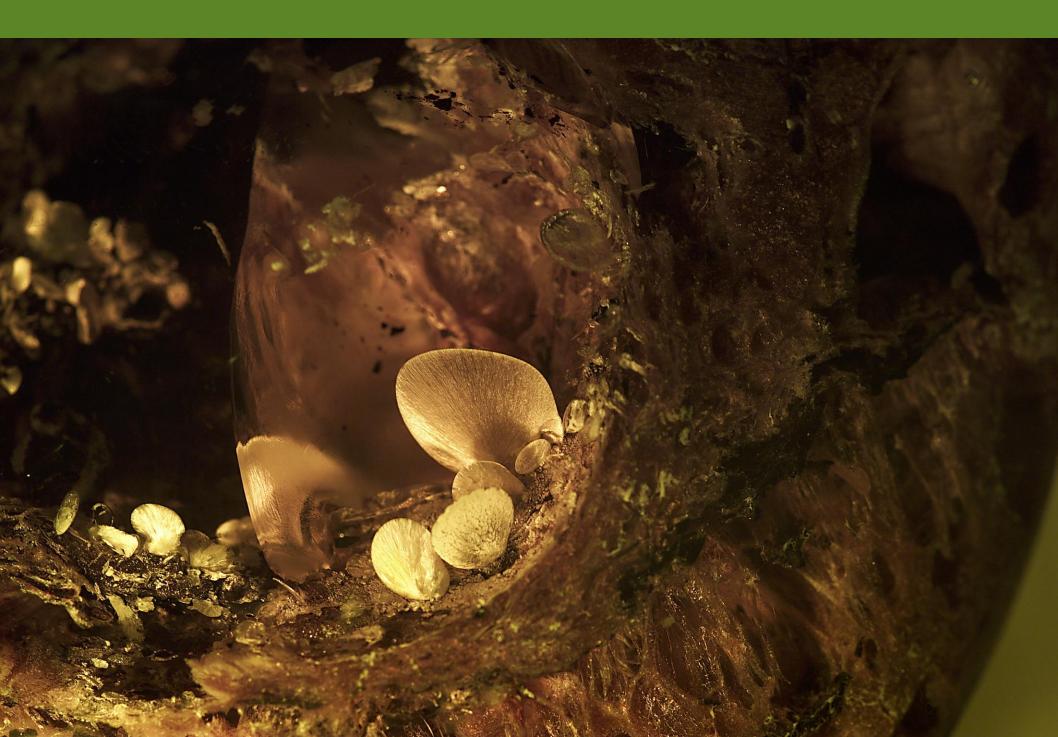




Farting beetle



2.0mm bracket fungus on wood. (Fun with lighting)



Imaging in solution can be very simple. Sometimes it is little more than point and shoot.

Nikon 60mm macro, D1X, Fiber Optic back light flash only. 1999 (f8 and be there)



The following are examples of imaging on black with only directional illumination.

Proper preparation of the material and fluid allows for a very clean image without the need to knock out the background.

When imaging dark field any particulate matter will reflect light and appear as a speck of light.

These images have the backgrounds untouched. Very little post shot editing is needed to clean them up.

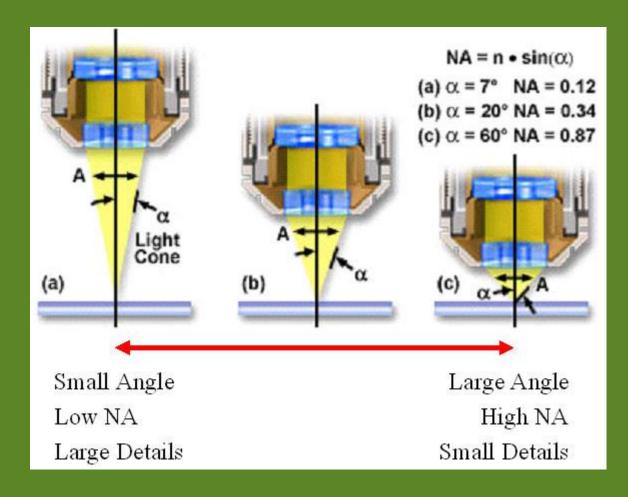


Small sea horse @ 2X





Translucent slide mounted or permanent mounts may best be imaged with the aid of a variable N.A. transmitted light source



Matching the Numerical aperture of the transmitted illumination to the objective brings it all into focus.

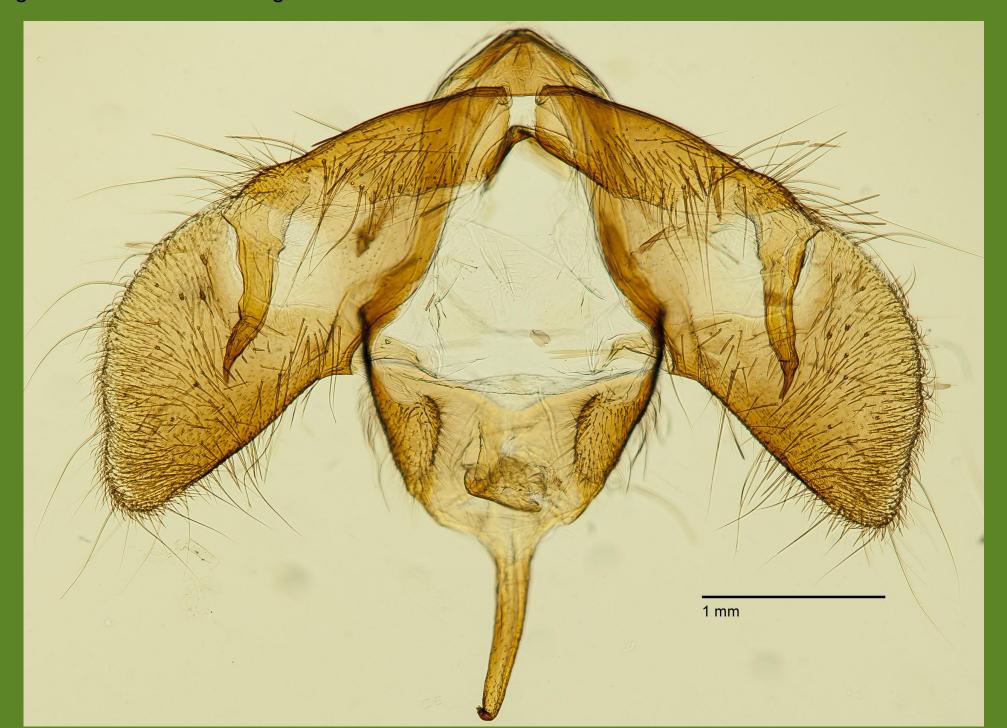
THIS CAN ALSO BE DONE WITH MACRO LENSES!!

Translucent slide mounted material.

1947. Moth genitalia mounted in balsam Single shot diffused back light. 65Mm Canon MPE @3.5X f4.0 Canon 5D Mark II



1947. Moth genitalia mounted in balsam VDE SolMate variable N.A. Trans Illuminator Single shot Focused back light. 65Mm Canon MPE @3.5X f4.0 Canon 5D Mark II







In Conclusion:

- •Proper preparation, cleaning and choosing the best mounting material/method.
- •Select the proper containment vessel and background based on desired results.
- •Keep as little material over the top of the specimen as is possible and use very clean solution.
- •Develop the lighting solution that works for you. Lighting is everything
- •Keep exploring what works!

Roy Larimer Visionary Digital Enterprises

END.