



Imaging techniques and troubleshooting

Hanna Royals | Entomologist
Hanna.h.royls@usda.gov

Topics:

- Intro to stacking
- Specimen preparation
- Choosing a background
- Microscope objectives
- Imaging larvae in ethanol



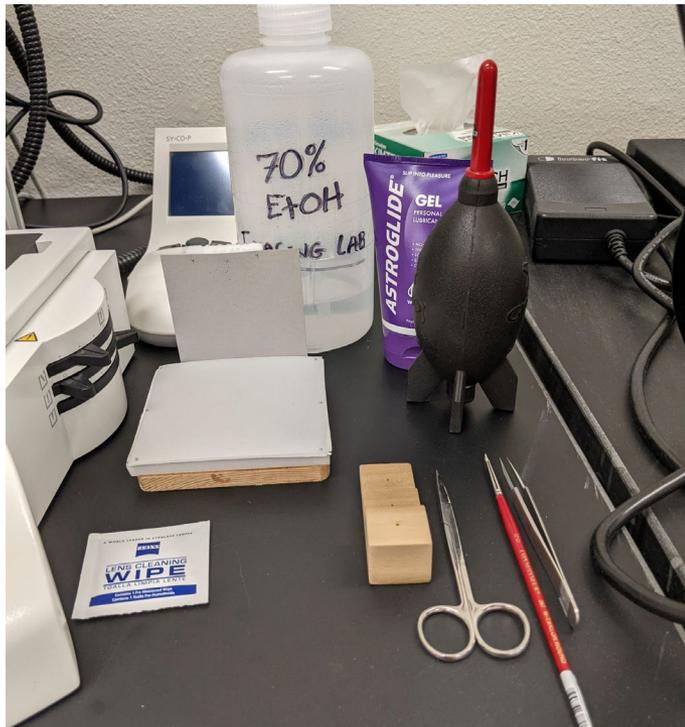


What is
image
stacking?



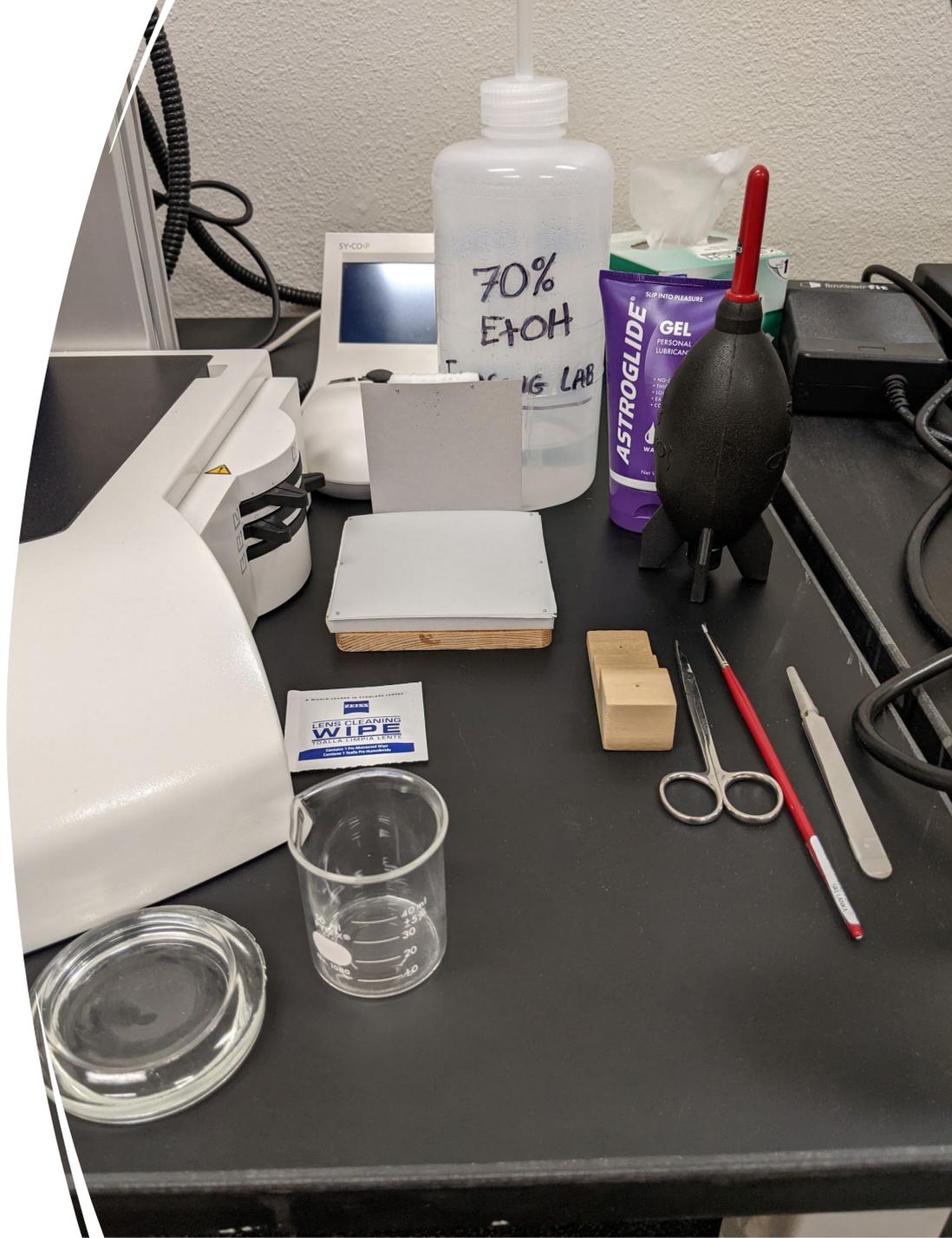
Specimen cleaning

- Don't nit-pick over tiny dust
- Minimize touching with hands
- Limit cleaning on type material



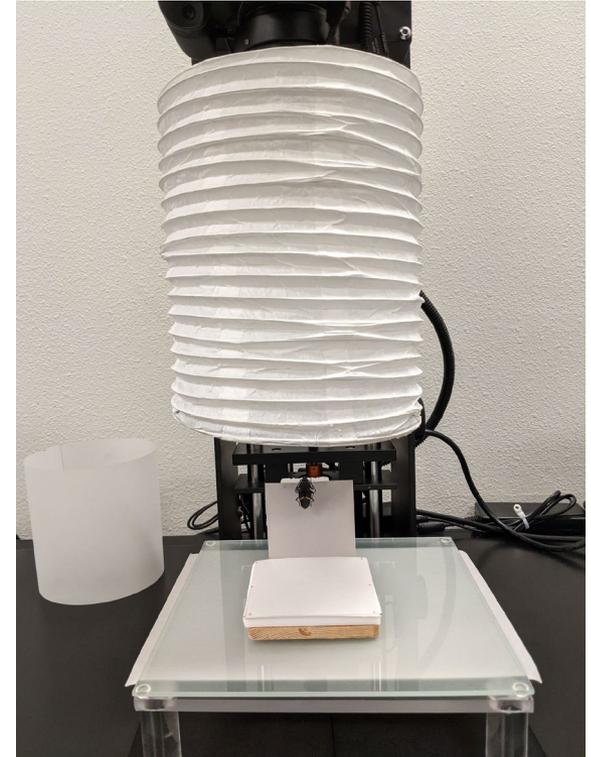
Cleaning supplies

- Stage for specimen
- Fine-tip forceps (removing pin labels, hairs, etc.)
- Paintbrush for gentle cleaning
- “Rocket” duster to blow off particles
- Diluted ethanol to remove dust and clean off grease



Imaging setup and light diffusers

- Stage with neutral background (18% grey cardstock)
- Frosted acrylic sheets
- Modified paper lantern
- Your imagination





Lighting

- Flash bulbs are essential for crisp, high-resolution images
- Many different flash setups are available for a range of budgets
- White-balance each time your lighting changes using a neutral spot in your image
- White backgrounds can wash out colors



Background comparisons



Background comparisons



Grey



White



Blue



Black

Microscope lenses

- Minimizing specimen movement is critical
- 5x, 10x, and 20x objectives can capture tiniest details.





General guidelines

- Cleaning isn't essential but makes for nice photos
- A stable base is critical
- Shutter speed minimum 1/100s – can be faster with flash bulb setup
- Use neutral grey background whenever possible – no colors!
 - Some exceptions for black and white backgrounds

Imaging alcohol specimens

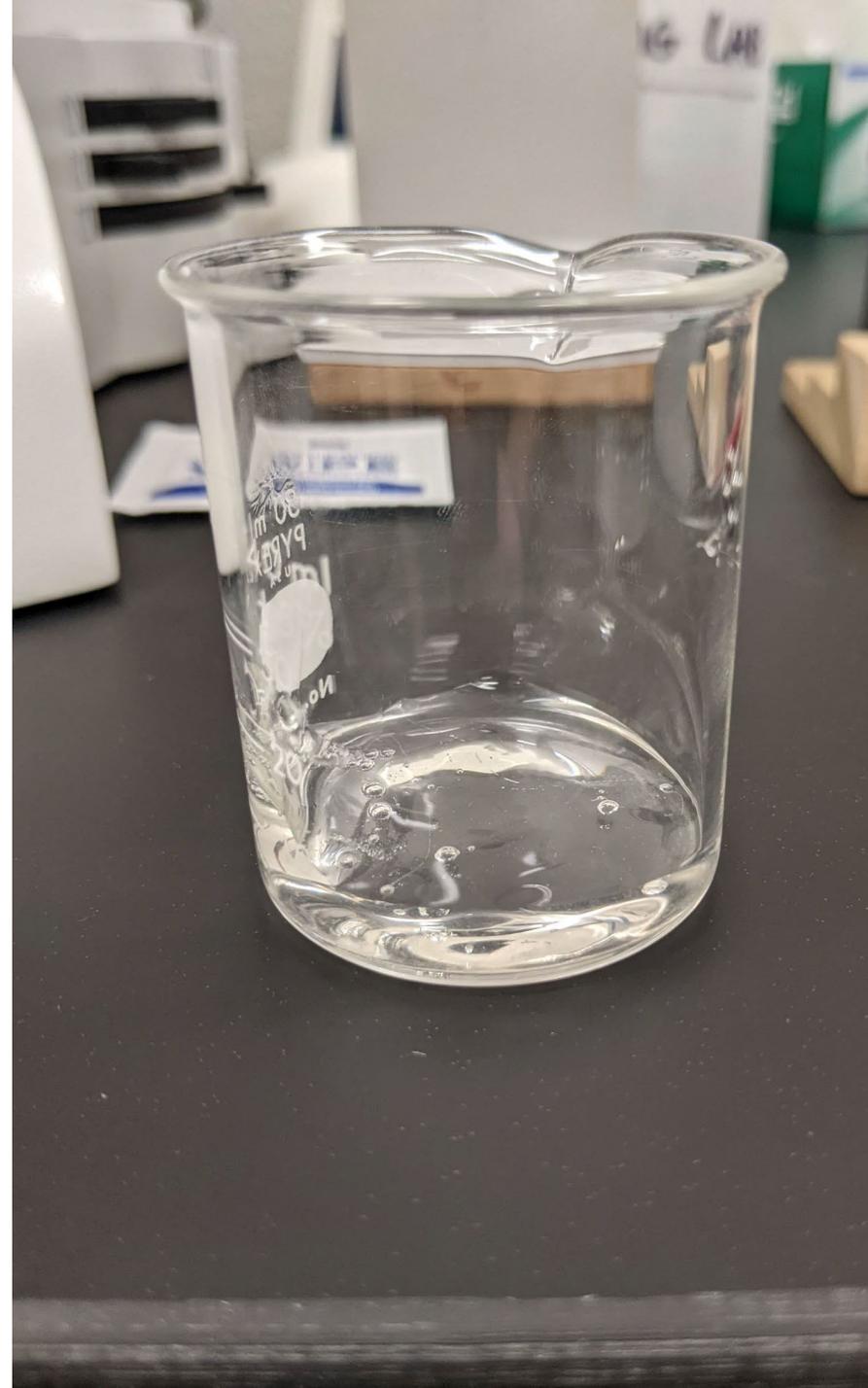
- Petri-dishes or glass beakers to fit your specimen
- 30%-70% ethanol
- Glycerin
- Specimen pins
- Forceps
- Patience



Beaker or petri dish large enough for specimen, but not wasting glycerin or ethanol

Glycerin – personal lubricant is thicker than pure glycerin and soluble in ethanol

INGREDIENTS: Purified Water, **Glycerin**, Hydroxyethylcellulose, Chlorhexidine Gluconate, Methylparaben, Glucono Delta Lactone, Sodium Hydroxide



Specimen positioning

- Settle specimen on top of glycerin
- Anterior/posterior views may require more of specimen to be submerged
- Gently pour ethanol to cover specimen
- Pins or forceps can be used to gently remove bubbles





Up next: Paul Langlois – setup, software and image editing