



## CHIRONOMID PREPARATION PROCEDURE

From BROOKS, S. J., LANGDON, P. G. & HEIRI, O. (2007) *The identification and use of palaeartic Chironomidae larvae in palaeoecology*, London, Quaternary Research Association.

**SAFETY:** See associated COSHH form. Always wear PPE (lab coat, safety glasses and nitrile gloves). Inform safety officer of any spills.

KOH – Highly toxic, severe burns, irritant

Ethanol – severe burns, highly flammable, irritant

Euporal essence – highly flammable, irritant

First aid – run with water for ~20 minutes, seek medical help for eye incidents.

### CHEMICALS REQUIRED:

10% potassium hydroxide (KOH)

80% and 100% ethanol

Euporal Essence

Euporal mountant

### EQUIPMENT REQUIRED:

Binocular dissecting microscope for collecting head capsules

Light microscope (x10, x20 and x40 magnification) for identification

Sonic bath

Scales / balance

Hotplate

Fine forceps

Fine dissecting needle

Bogorov sorter (order from Steve Brooks, NHM London)

6mm diameter round cover slips (1 per head capsule; 50-100 head capsules per sample)

4x glass cavity blocks (for cleaning head capsules)

90 and 212µm sieves

2x Petri dishes

Glass beakers

90 µm and 212 µm sieves

### PROCEDURE:

1. Weigh the sample (0.5g to 2g depending on head capsule concentration)
2. Place in small glass beaker with about 50ml of 10% KOH on hotplate for 5 minutes (stir)
3. Transfer to ultrasonic bath for 3-5 seconds

4. Rinse into stacked sieves (212  $\mu$ m and 90  $\mu$ m )
5. Transfer into petri dishes (one for each size fraction)
6. Pipette some of the sample into a Borgorov sorting tray and pick out head capsules under 10-20x magnification using a stereo microscope and place each capsule into a glass container (with snap-on lid) of 80% ethanol
7. Once you've collected all of the head capsules for a sample, you will mount each head capsule on a slide under a small, specialized cover slip.
8. Empty the contents of your tub into a glass cavity block.
9. Place the dish under the dissecting microscope so that you can see the head capsules; pick out each head capsule and place it into another glass cavity dish filled with 100% ethanol.
10. Once you have placed all of the head capsules from of the 80% ethanol into the 100% ethanol, you will now transfer all head capsules into Euporal Essence using the same method. Try to keep the glass covers on the cavity blocks to keep the smells to a minimum.
11. Lastly, you will transfer the head capsules from the Euporal Essence to a slide. Each slide can contain 10 head capsules.
12. Place a tiny amount of Euporal on the slide in 10 places (using the model slide as a guide).
13. With the glass cavity block containing the head capsules under the dissecting microscope, pick out one head capsule and place it on a blob of Euporal.
14. Using your forceps, pick out a cover slip and carefully place it on over the blob containing the head capsule.
15. Continue doing this until all head capsules are removed from Euporal Essence.