

## Shedding cercariae from patent *B. glabrata*

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### Introduction

After cercariae mature within the snail (usually 4- 5 weeks post infection), they are then liberated (shed) into the surrounding water. In field conditions, cercariae typically emerge in greatest numbers in the daytime. In the laboratory and for experimental purposes, investigators can adjust lighting conditions to take advantage of maximal release of cercariae at a time of the investigator's choosing. Some cercariae will still emerge in the dark. The following procedure works well for both *B. glabrata* and *B. truncatus*

### Equipment

- Incubator or modified room that can maintain a consistent temperature around 26°C
- Incubator fitted with a strong light source (e.g. 60 watt fluorescent bulb)

### Materials and reagents

- Featherweight forceps (photo, right)
- 100-200mL glass beakers
- Artificial pond water
- Filtration screen apparatus (if available) consisting of a 300 mL funnel with glass support (Kontes Glass) and a 47 mm diameter stainless steel support screen



Filtration screen apparatus



Assembled cercarial screen

### Procedure

- Using featherweight forceps, remove snails from their maintenance aquarium or pan, and place 10 snails into a glass beaker with enough water to cover the snails, ~50mL
- Allow the snails to sit in the water for five minutes as a gentle wash step. Discard the water and replace it with fresh water
- Place the beaker(s) under the light source (e.g. 26°C incubator) for 60-80 minutes, taking care not to overheat the snails.
- Pour the cercarial suspension through the filter apparatus into a clean beaker. Give the snails an additional rinse, with a squirt bottle containing water, to collect any left over cercariae.

- View the beaker under a microscope and look for healthy cercarial movement
- Remove snails, and place them back into the maintenance aquarium or snail pan
- Pour the cercarial suspension into a 50 cc tube

**To quantitate:** pipet 200 $\mu$ L of the cercarial suspension onto a gridded watch glass. Add 10-20 $\mu$ L of iodine to fix (kill) and stain the cercariae. Staining with iodine also aids in visualizing the cercariae with microscopy.

Count the number of cercariae to determine the number in 0.2mL of the cercarial suspension. After this is completed, calculate the number in 1.0mL of the suspension.

### **Caution**

*Schistosoma* spp. cercariae are infectious to humans and can penetrate exposed skin. Always use personal protective equipment (lab-coat, gloves, plastic sleeve covers, and face shield when working with cercariae. If exposed, immediately wash the skin with 70% EtOH and monitor for irritation. Report the exposure to your supervisor and/ or medical doctor.

### **References**

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