

## **Protocol for egg disinfection**

### ***Materials for the disinfection process***

- Prepare several 1L measuring cups or graduated cylinders
- Also prepare several 100 mL plastic vials with several holes (with Ø 1-2 mm) drilled in them to function as sieves (Figure 1)
- While disinfecting the eggs wear disposable gloves suitable for buffodine (check MSDS and gloves specifications).



*Figure 1: Plastic vials of 100 mL and with holes of Ø 1-2 mm drilled in the bottom. These holes will function as sieves and are effective in evenly distributing the disinfectant solution throughout the incubator.*

### ***General guidelines***

#### ***Precautions***

- The eggs must not be disinfected until 12 hours post-fertilization
- Disinfect the eggs once a week (with buffodine).
- Eggs should not be disinfected after 700 degree days, to prevent premature hatching
- For eyed eggs, use a ratio of 1:100; 100mL Buffodine per 10 L of water
- If the incubator holds 16 L of water, use 160 mL of buffodine
- Disinfect each incubator with eggs for 10 minutes

#### ***Disinfection steps***

- Measure 160 mL of buffodine in a 1 L measuring cup or graduated cylinder; this is for one incubator
- Dilute the buffodine with 500 mL of seawater at ambient temperature
- Close the waterflow of the incubator
- Lower the plastic 100 mL vials with holes into the water in the incubator, just deep enough for the holes to be submerged, but avoid touching the eggs

- Pour the previously prepared diluted buffodine solution slowly into the vial while holding the vial steady; this strategy will help to disperse the buffodine solution gently and evenly into the incubator (Figure 2).
- After the solution has been completely drained, remove the vial from the incubator and allow the buffodine to act for 10 minutes
- Turn on the water into the incubator and check the flowrate every 10-20 minutes and adjust as needed until a stable flow rate of 160 L/h is reached



*Figure 2: Disinfecting the eggs in the incubator with the aid of a vial with several small holes in the bottom. The holes in the bottom will help the distribution of the solution more evenly.*

### **Remarks**

Establishing a steady workflow is a good practice while disinfecting the eggs. This can be achieved by distributing the solution in one incubator at a time, with 2-minute interval between each incubator. Such a strategy is essential to disinfect every incubator efficiently. Furthermore, for consistency and to minimize stress, all incubators in a room should be disinfected consecutively. In addition, if several batches of eggs are fertilized on different weekdays the disinfection schedule can be adjusted for each batch through a 6- or 8-day shift in the beginning to obtain a shared weekday for disinfecting all the eggs. For example, a Monday and Thursday fertilized batch can have a shared disinfecting day on Wednesday by running a two 8-day shift for the Monday batch and one 6-day shift for the Thursday batch, meaning that both will be done on Wednesdays in tandem after the first two weeks.

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