Plate Pouring

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Working

Behavioural Genomics

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ABSTRACT

C. elegans is maintained in the laboratory on Nematode Growth Medium (NGM) agar which has been aseptically poured into petri plates. Smaller plates (35 mm diameter) are useful for mating or when using expensive drugs. Medium size plates (60 mm diameter) are useful for general strain maintenance, and larger plates (100 mm diameter) are useful for growing larger quantities of worms, such as for certain mutant screens. The NGM agar medium can be poured into petri plates easily and aseptically using a peristaltic pump. This pump can be adjusted so that a constant amount of NGM agar is dispensed into each petri plate. A constant amount of agar in the plates reduces the need for refocusing the microscope when you switch from one plate to another.

PROTOCOL STATUS

Working

We use this protocol in our group and it is working

MATERIALS TEXT

Nematode Growth Medium (NGM) agar

Preparation of the Hood

1

- Set the flow rate to 2 in the hood
- Turn on the lights and Plug switches
- Take the Pump and plug it in
- Set up the pump for the desired volume:
  a) Setup -> Options -> Volume (15ml for medium plates) -> Accept
  b) Setup -> Next -> Tube -> 8mm (Usually 8mm for big and medium plates and 3mm for imaging plates)
- Place the tubing around the pump making sure both ends are inserted into the agar bottle (Make sure the tubing is placed clockwise as the pump moves that way)
- Prime the tubes by: Setup-> Prime -> Accept (Leave it on for few mins till the tubes get warm), Press Accept to stop Prime

Actual Pouring

2

- Take plates in stack of 5/10 (as per convenience)
- Place the nozzle on top of the centre of the plate and press the foot pedal letting the agar to flow
- If some agar drops on the surface wipe it off with a paper towel after it solidifies
- When taking a pause of more than 1min put the pump back to prime
- When one bottle finishes, place the nozzle in the new bottle and prime to suck out the residual agar to prevent wastage (Wash the empty bottle and get some hot water in it)
- Put both openings into the new bottle and prime again

Post Pouring

3

- After pouring, wash the tubing (Prime) by passing the hot water from the clean bottle to the other one (to remove any agar in the tube)
- Followed by priming air into the tubing making sure no liquid is remaining
- Discard the agar in the agar container and wash the bottles and put them in the grey box in the lab
- Close all switches in the hood
- Return the pump
- Wrap and label the tube 2x in aluminium foil and take it to the media kitchen to put in the autoclave basket (Collect the tube the

following day)

- Order agar in the media kitchen for the next pouring day (i.e. Every other Monday)
- Let the plates solidify under the hood about an hour and then store it in the cold room