

# SOP-M018

Rectangle Plate Procedure on back of sheet

## Making Blood Agar for Bacteria Growth Round Plates

**Objective:** To prepare Agar for growing bacteria for the Bardot Project

**Getting Plates from Biology:** Email Amber Chase (achase@purdue.edu ) and ask for TSA plates with 25 mls = we normally ask for 200 or 400 at a time. Amber will email you when they are ready.

**Procedure:** Use the Brain Heart Infusion Agar Catalog #241830 BD (500g)

1. Weigh out 52gm of the powder and suspend it in to 950ml of Millipore Water.
2. Mix thoroughly. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder. Do not overheat. Aliquot 475ml into each of two 1L pyrex bottles, then Autoclave.
3. Place the bottles in the pre-warmed water bath, set at 55°C. Leave to equilibrate to that temperature for at least 15 minutes.
4. Place the sterile petri dishes on the flat surface in the hood to be filled with the agar.
5. Remove the autoclaved agar from the water bath and place the bottles in the laminar flow cabinet. Aseptically add 25 ml blood to each bottle. Gently swirl to mix.
5. **Pipette 25ml** (draw up 27 but use only 25ml) of the agar into the petri dishes being careful not to create bubbles. If bubbles do occur, just remove them with the pipette. Let cool and then place them with the agar side up carefully in the plastic sleeve they came in. Seal the sleeve with tape and label each stack the type of agar and the date. The next day, place the plates you poured in the 37° incubator for the sterility test overnight. If no contamination, label each stack “sterility check-good”. Then store in the cold room until needed.

**Procedure: Pouring plates of Agar without chilling first:**

1. Once autoclaved, let the Agar cool down to safe handling temperature. (Bottles will still be warm to touch.) Place the bottles containing Agar in a 60°C waterbath.
2. Repeat steps 4 and 5 above.

**Rectangle Plates Procedure on M007**

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Verified by: \_\_\_\_\_ Date: 10/22/2010 \_\_\_\_\_

Print Name

Sign Name