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Agar coated slide protocol

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1) Prepare 50 ml of 0.75% agar dissolved in media (same media that your organism grows in). Alternatively, media can be substituted for buffer.
    a. For example, 0.375g of agar is mixed with 50 ml of algal growth media in a 500 ml flask.
    b. Microwave on low power (~20%) for 4 minutes. NOTE: microwave power settings vary by manufacturer so watch the flask to make sure it does not boil over.
    c. Check that agar is completely dissolved and microwave again if solids remain.
2) Pour hot agar into a petri dish (or any other small vessel that a microscope slide fits in). Allow the agar to cool a bit or agar will not adhere to slide
3) While wearing gloved, hold microscope slide so that one side can be dipped into agar. I pinch the sides of the slide. Place slide agar side up onto a paper towel.
4) Finish coating the rest of the slides. Briefly examine slides to ensure agar pad is developed on slide. It does not have to be overly thick.
5) Load slides into slide rack and store refrigerator until use.
6) Allow slides to come to room temperature before loading cells. Use appropriate cover glass and seal with nail polish.
7) Allow remaining agar to solidify and then dispose according to your institutions requirements.

As prepared, the slides will last for about 24 hours but then the agar will dry out and new slides are needed. I usually make 10-12 slides at a time.

Some notes: Excess agar can easily be removed with a razor blade. Before using a slide that was stored in the refrigerator, allow it warm to room temperature for about 2-3 minutes or poor cell adhesion can occur.

Lastly, media or buffers with high salt contents (e.g. 0.5M NaCl and higher) may not be suitable for this technique. I have noticed that the agar does not uniformly solidify and the cells are exposed to high salt conditions. This leads to cell lysis.

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