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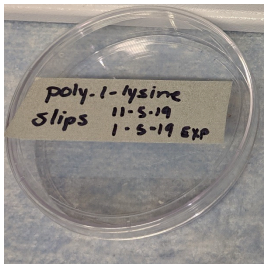
CODEX - Poly-l-Lysine Cover-Slip Preparation V.3



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We use this protocol and it's working

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Abstract

Poly-Lysine Coverslip Preparation

This protocol describes the process of creating Poly-lysine-coated coverslips that are required for mounting tissue sections for the CODEX® experiment workflow.

Guidelines

- Managers and supervisors - are responsible for making sure that technicians are properly trained and equipment and facility are maintained in good working order.

- Laboratory personnel - are responsible for reading and understanding this SOP and related documents and to perform these tasks in accordance with the SOPs.

Materials

MATERIALS

- WypAll™ L30 Multipurpose Wipers **Thermo Fisher Catalog #**19168203
- Parafilm Wrap PM996, 4 in. Wide, 125 ft./Roll **Thermo Fisher Catalog #**CP0672040
- Dumont Forceps (Cover Slip Forceps) **Fine Science Tools Catalog #**11251-33
- EMS Glass Cover Slips 22mm x 22mm **Electron Microscopy Sciences Catalog #**72204-01
- Poly-L-lysine 0.1% (w / v) **Merck MilliporeSigma (Sigma-Aldrich) Catalog #**P8920

Troubleshooting

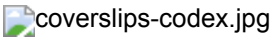
Safety warnings

- Use physical safety precautions when working with sharps (disposable blades, coverslips, etc).

Before start

- Wear gloves when handling cover slips to avoid depositing epithelial cells or residues on surfaces.
- You will need a sanitized or acid-washed beaker for this procedure.

1 Remove 12-15 cover-slips from box. 1m
Cover-Slip brand and type are REQUIRED, not suggested.


coverslips-codex.jpg

CODEX requires use of these specific cover-slips for their diagnostic process.

2 Pour enough poly-l-lysine solution to cover the bottom of the clean beaker.

3 Gently place the desired amount of cover-slips into the beaker, spreading them out and allowing both surfaces to have contact with the solution. 1m

4 Slowly swirl the beaker to spread the cover-slips around the base. 30s

5 Add 7 mL of poly-lysine solution above the cover-slips to ensure that all are fully covered. 2m

7 mL Poly-Lysine

6 Cover the beaker with plastic wrap or parafilm, and secure tightly with a rubber band to prevent evaporation. 2m

7 Leave cover-slips in poly-lysine solution for a **minimum** of 12 hours and up to one week **at room temperature**. 12h

12:00:00 or up to one week.

8 After waiting the required incubation period, carefully remove the rubber band and parafilm from around the neck of the beaker. 2m

9 Gradually pour the remaining poly-lysine solution into the proper waste container. 2m

10 Fill the same beaker containing the cover-slips to half volume with double-distilled water(ddH20) or purified water. 2m

11 Swirl the contents to mix the solution. 1m

12 Let the beaker and cover-slips sit for 30 seconds. 30s

To prevent removal of poly-lysine, do not soak in water for >1 minute during each washing step.

13 Slowly pour off the water into the sink. This completes **Wash #1** 30s

14 go to step #10 and repeat steps 9-12 (Wash #2) 1m 30s

15 go to step #10 and repeat steps 9-12 (Wash #3) 1m 30s

16 go to step #10 and repeat steps 9-12 (Wash #4) 1m 30s

17 go to step #10 and repeat steps 9-12 (Wash #5) 1m 30s

18 go to step #10 and repeat steps 9-12 (Wash #6) 1m 30s

19 go to step #10 and repeat steps 9-12 (Wash #7) 1m 30s

20 After completing the 7 washes, place 2 lint-free Wypall towels on the bench top. 30s

21 Remove the cover-slips from the water, placing them on top of the first set of towels. Ensure the cover-slips are not overlapping to allow proper drying. 2m

22 Using the specialty forceps indicated for use by CODEX, flip over each cover-slip onto the second clean towel to dry the reverse side. 20m

Specialty forceps indicated in the materials section.

23 Leave the cover-slips on the Wypall towel to dry. 1d

24 When the cover-slips are dry, the Poly-Lysine-coated cover-slips can be stored in a sterile petri dish or similarly covered container for up to 2 (two) months. 30m

Sterile Petri Dish containing the finished Poly-l-Lysine coated cover-slips for CODEX processing.