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

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1 Works for me

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Human BioMolecular Atlas Program (HuBMAP) Method Development Community

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

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

SAFETY WARNINGS

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

BEFORE STARTING

- 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.

Cover-Slip brand and type are REQUIRED, not suggested.



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

30s

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

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.
- 2m. After waiting the required incubation period, carefully remove the rubber band and parafilm from around the neck of the beaker.
- 9 Gradually pour the remaining poly-lysine solution into the proper waste container.

2m

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.

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

3 Slowly pour off the water into the sink. This completes **Wash #1**

30s

0 go to step #10 and repeat steps 9-12 (Wash #2)

1m 30s

5 go to step #10 and repeat steps 9-12 (Wash #3)

1m 30s

ogo to step #10 and repeat steps 9-12 (Wash #4)

1m 30s

dgo to step #10 and repeat steps 9-12 (Wash #5)

1m 30s

go to step #10 and repeat steps 9-12 (Wash #6)

1m 30s

 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



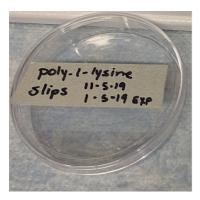
Specialty forceps indicated in the materials section.

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

24h

18

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.



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

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