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CODEX - Poly-L-Lysine Cover-Slip Preparation V.2

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Works for me

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Human BioMolecular Atlas Program (HuBMAP) Method Development Community

ABSTRACT

Poly-Lysine Coverslip Preparation

This section describes the process of creating Poly-lysine-coated coverslips that are used for the tissue sections in 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

- Be sure to gather all the appropriate materials listed in this protocol before starting.
- You will need a sanitized or acid-washed beaker for this procedure.

- 1 Remove the EMS cover-slips from box.

1m



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

- 2 Gently place the desired amount of cover-slips that you wish to coat in solution at the bottom of a sterilized glass beaker.

1m

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

30s

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

2m

7 ml Poly-Lysine

- 5 Cover the beaker with plastic wrap or parafilm, and secure tightly with a rubber band to prevent evaporation.

2m

- 6 

12h

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

 **12:00:00 or up to one week.**

- 7 After waiting the required incubation period, carefully remove the rubber band and parafilm from around the neck of the beaker.








2m

- 8 Gradually pour the remaining poly-lysine solution into the proper waste container.

2m

- 9 Fill the same beaker containing the cover-slips to half volume with double-distilled water(ddH2O) or purified water.

2m

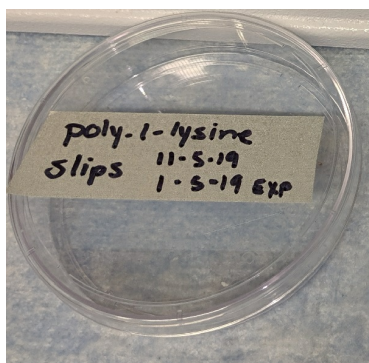
- 10 Swirl the contents to mix the solution. 1m
- 11  30s
Let the beaker and cover-slips sit for 30 seconds.
- To prevent removal of poly-lysine, do not soak in water for >1 minute during each washing step.**
- 12 Slowly pour off the water into the sink. This completes **Wash #1** 30s
- 13  go to step #9 and repeat steps 9-12 (Wash #2) 1m 30s
- 14  go to step #9 and repeat steps 9-12 (Wash #3) 1m 30s
- 15  go to step #9 and repeat steps 9-12 (Wash #4) 1m 30s
- 16  go to step #9 and repeat steps 9-12 (Wash #5) 1m 30s
- 17  go to step #9 and repeat steps 9-12 (Wash #6) 1m 30s
- 18  go to step #9 and repeat steps 9-12 (Wash #7) 1m 30s
- 19 After completing the 7 washes, place 2 Wypall towels or lint-free paper towels on the bench top. 30s
- 20 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
- 21 Using the specialty forceps indicated for use by CODEX,
flip over each cover-slip onto the second clean towel to dry the reverse side.



Specialty forceps indicated in the materials section.

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

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



Sanitized Petri Dish containing the finished Poly-L-Lysine coated cover-slips for CODEX processing.



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