VERSION 3

JAN 03, 2020

OPEN ACCESS

DOI:
dx.doi.org/10.17504/protocols.io.baxyifpw

Protocol Citation: Leigh Propper, Franchesca Farris, Marda Jorgensen 2020. CODEX - Poly-L-Lysine Cover-Slip Preparation. **protocols.io** <https://dx.doi.org/10.17504/protocols.io.baxyifpw>

License: This is an open access protocol distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

Protocol status: Working
 We use this protocol and it's working

Created: Jan 03, 2020

Last Modified: Oct 02, 2020

PROTOCOL integer ID:
 31448

Keywords: CODEX, HuBMAP, poly-lysine, coverslips, histology, staining, prep

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

Leigh Propper¹, Franchesca Farris¹, Marda Jorgensen¹

¹University of Florida

Human BioMolecular Atlas Program (HuBMAP) Method Development Community
 Tech. support email: Jeff.spraggins@vanderbilt.edu

Marda Jorgensen

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 #11251-3**

EMS Glass Cover Slips 22mm x 22mm **Electron Microscopy Sciences Catalog #72204-01**

Poly-L-lysine 0.1% (w / v) **Sigma Aldrich Catalog #P8920**

SAFETY WARNINGS

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

BEFORE START INSTRUCTIONS

- 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.

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(ddH2O) 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	and repeat steps 9-12 (Wash #2)	1m 30s
15	and repeat steps 9-12 (Wash #3)	1m 30s
16	and repeat steps 9-12 (Wash #4)	1m 30s
17	and repeat steps 9-12 (Wash #5)	1m 30s
18	and repeat steps 9-12 (Wash #6)	1m 30s
19	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,	20m

flip over each cover-slip onto the second clean towel to dry the reverse side.

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.