

Etching glass coverslips

(from Matt Lang lab)

Materials

100 g KOH

Ethanol

MilliQ DI H₂O

Corning Coverglass (22 x 40 mm) No. 1.5

1. Mix 100g of KOH in 300 mL of Ethanol during 30 min in a 1 L beaker with magnetic bar
2. Load teflon coverslip racks with coverglass
3. Fill 2 more 1 L beakers with 300 mL DI H₂O and degas for 5 minutes
4. Fill one more 1 L beaker with 300 mL ethanol and degas for 5 minutes
5. Degas the ethanol/KOH beaker for 5 minutes
6. Submerge one coverslip rack in the KOH/ethanol solution and sonicate for 5 minutes
7. Wash coverslips by dipping the rack up and down and spinning it in the ethanol beaker
8. Wash coverslips by dipping the rack up and down and spinning it in the DI H₂O beaker
9. Submerge coverslip rack in the second DI H₂O beaker and sonicate for 5 minutes
10. Spritz coverslips with DI H₂O bottle - use lots of H₂O and wash every single slip twice using lots of pressure
11. Spritz coverslips with ethanol bottle - use lots of ethanol
12. Repeat steps 6-11 with each rack of coverslips
13. Dry all coverslips in the oven for at least 15 minutes
14. Store coverslips in racks inside sealed plastic nalgene containers

Peptide-DNA tethering assay

(Marie-Eve Aubin-Tam)

Materials

20 μ L of \sim 0.1ng/ μ L A08-3500bpDNA-biotin (see note for A08 sequence)

1.26 μ m streptavidin coated polystyrene beads (Spherotech, SVP-10-5)

Casein

PBST (PBS with 0.01% tween-20)

Vacuum grease

1. Make a flow chamber with etched glass coverslips
2. Flow \sim 0.1ng/ μ L A08-3500bpDNA-biotin, incubate 1 hour at room temperature
(do steps 3-7 in the meantime)
3. Make a 1mg/ml casein solution in PBST, filter with 0.2 μ m syringe filter
4. Mix 3 μ L of 1.26 μ m streptavidin coated beads with 150 μ L PBST
5. Spin down at 10000rpm during \sim 3 min, remove supernatant, resuspend in 150 μ L PBST
6. Spin down at 10000rpm during \sim 3 min, remove supernatant, resuspend in 150 μ L PBST
7. Spin down at 10000rpm during \sim 3 min, remove supernatant, resuspend in 150 μ L of the casein solution, sonicate beads, keep on ice
8. Flow 100 μ L of casein solution in flow chamber, incubate 30 minutes
9. Flow 50 μ L of beads solution in flow chamber, incubate 30 minutes
10. Wash channel with 150 μ L of casein solution
11. Seal chamber with vacuum grease.

Note: The sequence of the glass binding peptide (A08) is CGGRSGRRRSHHHRL.