Last Update: 22 March 2023

DNA/RNA Surface

1. Expose the coverslips in Piranha solution for 1 hr.
2. Discard the Piranha solution and rinse the coverslips in DI water three times.
3. Dry coverslips with N2 or air stream.
4. Place coverslips and a 200 mL beaker in oven for 30 min under 110C.
5. Take the coverslips and beaker out of oven and let cool to room temperature.
6. Add 142.5 mL hexane and 7.5 mL GPTES into the beaker. Immerse the coverslips in the beaker overnight. (Note: salinization on the coverslip surface relies on the thin water layer on glass surface, thus the reaction takes much longer time compared to ethanol/acid based methods)
7. Rinse the coverslips with hexane several times to remove unbound GPTES.
8. Place the coverslips in the over for 30 min under 110C. This step allow crosslinking of silane molecules. (Curing step)
9. Prepare 100 uM DNA solution in 3X SCC buffer. Add 100 uL per 2 coverslips. Incubate in dark and humid environment for 10 hrs.
10. Rinse the coverslips with DI water.
11. Immerse the coverslips in 50 mM ethanolamine/0.1 % SDS/0.1M Tris pH 9.0 for 30 min.
12. Additional surface passivation with BSA for 10 min before experiment.

DSSTween-20 Surface

1. Scrub off tape residues using acetone from slides.
2. Rinse with DI water, acetone, methanol, Alconox until the slide looks clean.
3. Put slides in a staining jar. Sonicate with 1:1 acetone/methanol to remove tape residues.
4. Rinse with DI water. Dry with N2 gas.
5. Expose the coverslips in Piranha solution for 1 hr.
6. Discard the Piranha solution and rinse the coverslips in DI water three times.
7. Add 5 M KOH to the staining jars and sonicate for 1 hour.
8. Rinse PP staining jar with DI, hexane three times. Dry with N2 gas.
9. Rinse the coverslips with DI water three times. Dry with N2 gas. Put dried coverslips in to staining jar.
10. Add 50 ml hexane and 50ul DDS to the staining jar. Make sure DSS has minimal contact with the air. Tighten the cap.
11. Gently shake the staining jar at room temperature for 1.5 hour.
12. Dump hexane-DSS. Rinse and sonicate the coverslips in fresh hexane for 1min. Repeat this step for 3 times.
13. Dry the coverslips using N2 gas.

PEG Surface

1. Rinse the coverslips thoroughly in DI water.
2. Add acetone to the staining jars. Sonicate for 20 minutes.
3. Discard the acetone and rinse the coverslips in DI water three times.
4. Add 1 M KOH to the staining jars and sonicate for 20 minutes.

5. Rinse the coverslips with DI water three times.
6. Add coverslips to a large beaker. Add sulfuric acid to the beaker containing the coverslips.
7. Add hydrogen peroxide to the sulfuric acid in an acid: hydrogen peroxide ratio of 3:1.
8. Gently stir the solution and leave undisturbed for ~30 minutes.
9. Remove the coverslips and rinse with DI water before putting them into the staining jars.
10. Rinse the coverslips thoroughly with DI water.
11. Prepare 2% v/v solution of silane reagent in methanol. (50ml Methanol, 1.5ml APTES, 2.5 ml acetic acid)
12. Add salinization solution to staining jar and agitate for 20 mins.
13. Rinse with excess amount of water.
14. Dry Coverslips in vacuum desiccator.
15. Prepare a 50:1 methoxy: biotin PEG-NHS mixture in pH7.4 PBS buffer. (100mg PEG, 5mg bioPEG in 500ul Buffer)
16. Pipette 100uL of PEG mixture onto a dry salinized coverslip and place another coverslip on top. Use a spacer between two coverslips. Make sure the entire side of the coverslip is coated.
17. Incubate for 3 hrs. (At least for 2 hr).
18. Wash with excess water. Blow dry with filtered air/Oven dry.
19. Prepare (0.01 mg/ml avidin solution), (1mg/ml BSA), (0.01mg/ml biotin-BSA and 1mg/ml BSA mixture) in Tris-HCl buffer.
20. Flow BSA/biotin-BSA solution through microchannel and incubate for 5 min.
21. Flow Tris-HCl through to wash away excess BSA.
22. Flow avidin solution through and incubate for 1 min.
23. Flow Tris-HCl through to wash away excess avidin.
24. Flow BSA solution through and incubate for another 5 min.
25. Flow Tris-HCl through to wash away excess BSA.
26. Prepare 2.5% v/v (5ul SWCNTs in 195ul TAE) DNA-SWCNTs sample TAE buffer. Flow through microchannel and incubate for 30 mins.
27. Flow TAE buffer through to wash out unbound SWCNTs. (200 flow cell volume)
28. Prepare 2.5% v/v (5ul QD in 195 TAE) DNA-QD sample in TAE buffer. Flow through microchannel and incubate for 30min.
29. Flow TAE buffer through to wash out unbound SWCNTs. (200 flow cell volume)
30. Flow reaction buffer through the channel.