## **PEG** coating procedure

This is an alternate PEG coating protocol, which appears to passivate the surface more completely than the other. It is more time consuming, so is appropriate if the alternate procedure causes sticking.

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Adapted from: Gidi, et. al. "Efficient one-step PEG-silane passivation of glass surfaces for single-molecule fluorescence studies." ACS applied materials & interfaces 10, no. 46 (2018)

## **Initial Cleaning**

- 1) Clean slides by immersing in  $\sim$ 1% Hellmanex solution.
- 2) Heat in the microwave and sonicate for 10 mins.
- 3) Rinse 3-5 times in DI water.
- 4) Immerse slides in 0.1 M NaOH for 10 minutes. Rinse 3-5 times and store in DI water.

## Coating

The goal in this procedure is to minimize the water concentration in solution during the coating. Water hydrolyzes the PEG-silane, making the coating procedure less efficient.

- 1) Take cleaned slides and coverslips out of the water and dry with nitrogen. Rinse slides with acetone and dry again with nitrogen.
- 2) Place slides in glass petri dish containing a desiccant like Drierite. Place in 90°C oven for 10 minutes.
- 3) Dissolve PEG silane at 5% by weight in anhydrous DMSO. Briefly sonicating the solution disperses the PEG silane more efficiently.
- 4) Drop 15  $\mu$ L of silane solution on the slide and place the coverslip on top of the drop. Leave at 90°C for 20 minutes.
- 5) On the underside of the slide, mark the corners of the coverslip using a permanent marker. You will need to place the coverslip back in nearly the same location.
- 6) Separate the coverslip and glass slide using a razor and rinse both the coverslip and slide thoroughly with DI water. Dry with nitrogen.
- 7) Place spacers and put the coverslip back in nearly the same location as it was when coating.