

Pre-hybridizing Polylysine Slides

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In our hands, polylysine slides require a pre-hybridization step to lower background fluorescence.

Pre-hyb solution:

25 ml 20X SSC

1 ml 10% SDS

1 gram BSA

74 ml dH₂O

Dissolve BSA thoroughly

Filter through a 0.2 micron filter

Pre-warm at 45° C in coplin jar for at least 2 hours

Slide Preparation:

- **Mark location of spots and label slide with a diamond tip pencil**
- **Rehydrate in a humid chamber of 0.3 M potassium phosphate for 30 min.**
- **UV crosslink in Sralinker on 1,000 energy setting = 1,000 X100 ujoules**
- **Wash 30 sec. in 0.1% SDS**
- **Boil in water 3 min.**
- **Snap cool in cold dH₂O**
- **Place in 45° C pre-hyb solution**
- **Pre-hyb at 45° C for 1 hour**
- **Wash by hand dipping slides several times in a large volume (500 ml) of room temperature dH₂O**
- **Shake slides dry**
- **Set up hybridization immediately**