

SOP 14.0: Pre-Hybridization of Ultra GAPS (Corning)

1.0 PURPOSE: To provide a standard operating procedure (SOP) for pre-hybridization of Ultra GAPS (Corning) slides.

2.0 SPECIAL NOTES: The pre-hybridization should occur immediately prior to the hybridization of the slides.

3.0 AUTHOR: Chris Sawyer

4.0 RELATED DOCUMENTS: A7.0, A13.0, H3.1

5.0 REQUIRED MATERIALS AND EQUIPMENT:

Slide rack	Pyrex container
Milli Q water	Microfuge with slide adaptors
Oven	Forceps
Coplin jar	

6.0 DEFINITIONS:

6.1 Element: A single spot of DNA, SSC, and Betaine.

6.2 Arrayer: Robot for printing arrays.

6.3 SSC: SSC or saline-sodium citrate buffer is used as a buffer for spotting elements and cleaning the pins of an array.

7.0 PROCEDURES:

7.1 Preparation of pre-hybridization solution:

7.1.1 Make a 50ml solution containing in a small container: 12.5ml of Formamide, 12.5ml of 20X SSC, 250 μ l of 20mg/ml of BSA from, 500 μ l of 10% SDS, and 24.25ml of Milli Q water. It may be necessary to make a larger volume depending on the number of slides being processed.

7.1.2 Warm the solution in the oven at 42° Celsius. Allow about an hour for the solution to come up to the appropriate temperature.

7.1.3 After the solution has warmed to 42° Celsius, place the slides flat in the container with the elements up. Make sure the slides are not stacked and are covered by the solution.

7.1.4 Place the container back into the oven at 42° Celsius and allow the slides to incubate for 30-60 minutes.

7.1.5 Fill a coplin jar marked H₂O with Milli Q water for later use.

7.1.6 Take the container out of the oven after its incubation time is complete.

7.1.7 With forceps, take one slide at a time out of the solution and dunk it in the water inside the coplin jar 3-4 times to get off any excess solution and salts.

- 7.1.8 Immediately spin the slide, 15 seconds or until it is dry, with elements up in a microfuge equipped with a slide adaptor.
- 7.1.9 The slide is now ready for hybridization.

8.0 REVISION HISTORY:

Date	Version	Author	Reason
8/28/03	1.0	Chris Sawyer	Initial creation of protocol