Stripping Method 1: Denaturation & Neutralization (Den/Neutr) - 38-54% recovery

A 0.4 M NaOH solution was heated to 45C, and the blots were placed in it for 30 minutes with gentle agitation. The blots were then neutralized in 0.1 x SSC, 0.1% (w/v) SDS, 0.2 M Tris-HCl, pH 7.5, for 15 minutes.

Stripping Method 2: 0.5% SDS (0.5% SDS) - 59-67% recovery

A solution of 0.5% SDS was heated to boiling. The blots were placed in a glass baking dish and the boiling SDS solution was poured onto the blots. The membranes were kept in the SDS solution until it cooled to room temperature.

Stripping Method 3: Tris/EDTA/Denhardt's (TE+Denhardt's) - 23-44% recovery

An oven was pre-heated to 75 deg C, and a solution of 1 mM TrisCl (pH 8.0), 1 mM EDTA (pH 8.0), 0.1 x Denhardt's reagent, was heated in a glass beaker to 75 deg C. The blots were placed in the 75 deg C solution, and the beaker was put in the oven for 2 hours. The blots were removed from the solution and rinsed briefly with 0.1 x SSPE at room temperature.

Stripping Method 4: 50% Formamide (50% Formamide) - 20-37% recovery

A 50% formamide, 2 x SSPE solution was heated to 65 deg C in a covered glass container in a fume hood. The blots were placed in the formamide solution at 65 deg C for 1 hour. The blots were removed from the formamide and rinsed briefly with 0.1 x SSPE at room temperature.

DNA Blot Stripping

1. Prepare the solutions as follows:

• Stripping: 0.4 M NaOH
• Neutralization: 0.1 x SSC, 0.1% (w/v) SDS, 0.2 M Tris·Cl, pH 7.5

2. Heat the Stripping solution in a glass beaker on a hot plate to 45°C. Heat enough
solution to fully cover the blot.

3. Remove the blot from its plastic bag with forceps and gently place it in the heated
Stripping solution. Let the blot float into the solution. Avoid pushing it into the
solution with forceps to prevent damaging the membrane.

4. Incubate the blot in the Stripping solution at 45°C for 30 minutes. Then transfer the
blot to the Neutralization solution and incubate for 15 minutes.

5. Seal the blot in a plastic bag. For radioactive probes, expose the blot to film for the
normal exposure time to check for the removal of the probe. For non-radioactive
probes, repeat your specific detection protocol. If the probe has not been completely
removed from the blot, repeat the stripping process. When all probe has been removed from the blot, you can reprobe it beginning with the pre-hybridization step.

DNA or RNA Blot Stripping

1. Prepare this stripping solution: 0.1% (w/v) SDS for RNA blots or 0.5% (w/v) SDS for
DNA blots.

2. Heat the SDS solution in a glass baking dish until boiling. Remove it from the heat
after boiling.

CAUTION: Do not continue to heat the SDS solution containing radio

active membranes while stripping. Radioactive contamination can occur by aerosolization or boiling over.

3. Place the blot with forceps in the SDS solution. Incubate the blot for 15 minutes with gentle agitation. Repeat this step once.

4. Seal the blot in a plastic bag. For radio active probes, expose the blot to film for the normal exposure time to check for the removal of the probe. For non-radioactive probes, repeat your specific detection protocol. If the probe has not been completely removed from the blot, repeat the stripping process. When all probe has been removed from the blot, you can re-probe it beginning with the pre-hybridization