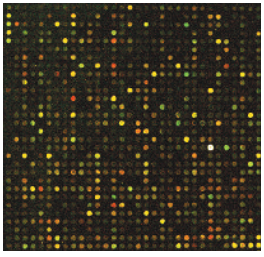
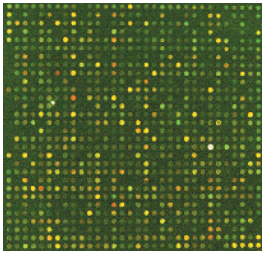
Pronto!™ Background Reduction Kit Quick Reference Guide

Treament with Pronto! Background Reduction reagents effectively eliminates both substrate and spotted background autofluorescence and prepares microarrays for hybridization. The strong reducing effect of this treatment leads to increased assay sensitivity and specificity by removing colored compounds and oxidation precursors. Although the beneficial effects of this treatment are fully realized when carried out as part of the Pronto! Universal Hybridization Kit (Cat. No. 40026), it may also be performed as the final step in the process of array fabrication.

The Pronto! Background Reduction Kit (Cat. No. 40029) is designed for treating 50 microarrays. Reagents are used most efficiently by simultaneously processing multiple arrays in staining jars. For example, 10 arrays can be processed simultaneously in glass staining dishes (Fisher Cat. No. 08-812) using 200 mL of solution for each step. Larger numbers of arrays and containers of greater capacity will require proportionally larger volumes.





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Pre-Soak Protocol

1. Heat required volumes of Pre-Soak Solution to 42ºC for 30 minutes.
2. Dilute 2 mL of Liquid Borohydride solution into 198 mL (1:100 dilution) of 42ºC Pre-Soak Solution. Swirl gently to mix.

   Note: Open bottle of Liquid Borohydride under an exhausting hood and keep closed when not in use. Wear appropriate Personal Protective Eyewear when working with this material, as indicated in MSDS. Consult MSDS and the appropriate local authority regarding disposal of this material.
3. Immerse arrays in solution from Step 2 and incubate at 42ºC for 20 minutes.
4. Transfer arrays to nuclease-free, high purity water and incubate at ambient temperature (22 to 25°C) for 30 seconds.
5. Repeat Step 4 twice, for a total of 3 washes.
6. Proceed according to one of either alternative steps a or b:
   a. If presoaking was done in preparation for hybridization, transfer arrays to the Pronto!™ Pre-Hybridization Solution and continue through the hybridization process as indicated in laboratory protocols.
   b. If presoaking was done as the final step of the fabrication process, dry arrays by centrifugation at 1,600 xg for 2 minutes. Place arrays in a Corning® 25 Slide Holder (Cat. No. 40081). Place holder containing arrays in a Corning Microarray Storage Pouch (Cat. No. 40086) and heat-seal the pouch. Hybridize arrays within 6 months of fabrication.

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Corning Incorporated
Life Sciences
Tower 2, 4th Floor
900 Chelmsford St.
Lowell, MA 01851
t 800.492.1110
t 978.442.2200
f 978.442.2476
www.corning.com/lifesciences

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