

Protocol

SCHOTT

Nexterion® Aminosilane Slide Processing Kit DNA application

Dok-Nr.:	LS6-HBM-N-002
Version:	1.0
Seite:	1/4
Datum:	© Feb 2010

1	Product overview	2
2	Kit contents.....	2
3	Storage and handling.....	2
4	Pre-hybridization.....	3
5	Hybridization.....	3
6	Post-hybridization washing	4
7	Important information about patents	4

For Technical Assistance, please contact

**SCHOTT Technical Glass
Solutions GmbH**
Otto-Schott-Str. 13
07745 Jena
Germany

Phone: +49-(0)3641-681-4069
Fax: +49-(0)3641-681-4970
E-Mail: coatedsubstrate@schott.com

Additional information at:
www.schott.com/nexterion

Protocol

SCHOTT

Nexterion® Aminosilane Slide Processing Kit DNA application

Dok-Nr.:	LS6-HBM-N-002
Version:	1.0
Seite:	2/4
Datum:	© Feb 2010

1 Product overview

SCHOTT Product Group Nexterion® offers a kit with ready to use reagents for the blocking, hybridization and washing of 25, 100 or 500 printed aminosilane coated slides. The reagents in the kit are optimized for use with aminosilane coated slides (e.g. Nexterion® Slide A+ and Nexterion® Slide AStar) printed with long oligonucleotides, PCR products etc. The Nexterion® Oligo Block A offers the advantage combining some of the initial washing and blocking steps thereby reducing the overall processing time. Nexterion® Oligo Hyb contains formamide, which allows for increased hybridization stringency at decreased hybridization temperatures. The kit includes an easy to-use slide processing protocol.


2 Kit contents

Kit components	Description	Store at	25 Slide Kit	100 Slide Kit	500 Slide Kit
Nexterion® Oligo Block A	Pre-hybridization solution (add BSA)	20 - 25 °C 4 °C †	1 x 500 ml	3 x 500 ml	5 x 1000 ml
Nexterion® Oligo Hyb	Hybridization solution (ready-to-use)	4 °C	1 x 10 ml	2 x 10 ml	1 x 100 ml
Nexterion® Oligo Wash A	Washing solution component A	20 - 25 °C	1 x 1000 ml	2 x 1000 ml	6 x 1000 ml
Nexterion® Oligo Wash B	Washing solution component B	20 - 25 °C	1 x 100 ml	3 x 100 ml	2 x 1000 ml
BSA	Bovine serum albumin	4 °C	1 x 5 g	3 x 5 g	5 x 5 g

†: after adding BSA

3 Storage and handling

Store Nexterion® Oligo Hyb Solution and BSA at 4 °C. After adding BSA to the Nexterion® Oligo Block A Solution store it at 4 °C and use it within 3 - 5 days. All solutions should be used prior to the expiration date.

<h1>Protocol</h1>		
Nexterion® Aminosilane Slide Processing Kit DNA application	Dok-Nr.:	LS6-HBM-N-002
	Version:	1.0
	Seite:	3/4
	Datum:	© Feb 2010

4 Pre-hybridization

Note: Do not allow the slides to dry during the pre-hybridization and washing procedure.

1. Pre-warm an appropriate volume of Nexterion® Oligo Block A containing BSA to 42°C for several minutes until the precipitate is completely dissolved. Shake the bottle from time to time.
2. Incubate slides in Nexterion® Oligo Block A for 60 min at 42°C. Use a volume of buffer which ensures that all slides are completely covered with Nexterion® Oligo Block A.
3. During the pre-hybridization step prepare Wash Solution 1 by diluting Nexterion® Wash B 1:40 with diH₂O. Prepare 50 ml per slide (i.e. 1.25 ml Nexterion® Oligo Wash B, add diH₂O to 50 ml).
4. Wash the slides 5 min in Wash Solution 1 at 20 - 25 °C.
5. Rinse with diH₂O for 30 sec.
6. Dry the slides in either an oil-free air (or nitrogen) jet or by centrifugation (200 x g for 5 min). This will avoid leaving any drying marks on the slide surface.

5 Hybridization

1. Re-suspend the dried, labeled target to be applied to the slide in Nexterion® Oligo Hyb. If the target is already dissolved in a different buffer or water, the sample can also be diluted in the hybridization solution to get at least 90% (v/v) buffer in the final hybridization solution.
2. Denature the resuspended target by heating at 95 °C for 3 min in a water-filled well of a heat block. Perform a quick spin in a microcentrifuge. Pipette the appropriate volume of hybridization solution containing the target onto the array surface of a pre-hybridized slide under a coverslip or inside a hybridization station.
3. It is suggested to hybridize the slides at 42 °C for 16 hours. In most cases it's necessary to adjust the temperature and incubation time to the needs of the particular application.

Protocol

SCHOTT

Nexterion® Aminosilane Slide Processing Kit DNA application

Dok-Nr.:	LS6-HBM-N-002
Version:	1.0
Seite:	4/4
Datum:	© Feb 2010

6 Post-hybridization washing

Note: Do not allow the slides to dry during the hybridization and washing procedure. Wash Solution 2, 3 and 4 should be prepared in advance. Use at least 50 ml solution per slide.

Post-hybridization washing solutions	Preparation
Wash Solution 2	50 ml Nexterion® Oligo Wash A, 25 ml Nexterion® Oligo Wash B, Fill up to 500 ml with diH ₂ O
Wash Solution 3	50 ml Nexterion® Oligo Wash A, Fill up to 500 ml with diH ₂ O
Wash Solution 4	5 ml Nexterion® Oligo Wash A, Fill up to 500 ml with diH ₂ O

1. Place the slides into a slide rack and immerse it in a dish containing Wash Solution 2. Wash in the above solution 1 x 10 min at 20 - 25 °C.
2. Wash 1 x 10 min in Wash Solution 3 at 20 - 25 °C.
3. Wash 1 x 10 min in Wash Solution 4 at 20 - 25 °C.
4. Dry the slides either in an oil-free air (or nitrogen) jet or by centrifugation (200 x g for 5 min).
5. Protect the slides from light, dust and abrasion of the slide surface, until ready for scanning. Ensure that the laser and filter set of the scanner are compatible with the fluorescent labeling of the probe molecules.

7 Important information about patents

Using arrays based on SCHOTT Nexterion® products for dual color analysis on a single array in which at least two different samples are labeled with at least two different labels may require a license under one of the following patents: U.S. patent nos. 5,770,358 or 5,800,992 or 6,225,625 and U.S. patent no. 5,830,645. Manufacturing and use of probe arrays may require a license under the following patents: U.S. patent no. 6,040,138 or 5,445,934 or 5,744,305 and under the following patents owned by Oxford Gene Technology Ltd. ("OGT"): European patent no. EP 0,373,203, U.S. patent nos. 5,700,637 and 6,054,270 and Japanese patent nos. 3393528 and 3386391 ("The OGT patents"). Other patents may apply. The purchase of SCHOTT Nexterion® products does not convey any license under any of the OGT patents or any of the other patents referred to. For all applications SCHOTT North America Inc. and SCHOTT Technical Glass Solutions GmbH make no representation or warranty that the practice of its technology and products or any improvement will not infringe or violate any domestic or foreign patent of any third party. Before making or using any oligonucleotide arrays you should contact OGT to discuss a licence. To inquire about licensing under the OGT patents, please contact OGT at licensing@ogt.co.uk.