

Three-dimensional thin film coating

Nexterion® Slide H



Overview

Type of coating	Immobilization method	Typical probes	Ordering information			
			Nexterion® product	Barcode option	Item number	Slides per pack
Thin film 3-D polymer surface	Amine reactive chemistry Covalent binding	<ul style="list-style-type: none">• Proteins• Amino-linked glycans• Amino-linked oligonucleotides	Slide H	Laser	1070936	25

Key product features

- Ideal substrate for printing protein, amino-modified glycans or oligonucleotide microarrays
- High probe loading capacity
- Exceptionally wide dynamic range
- Very low non-specific binding characteristics
- Optimal preservation of native structure and biological activity of protein probes
- Compatible with all common microarray printers and scanners

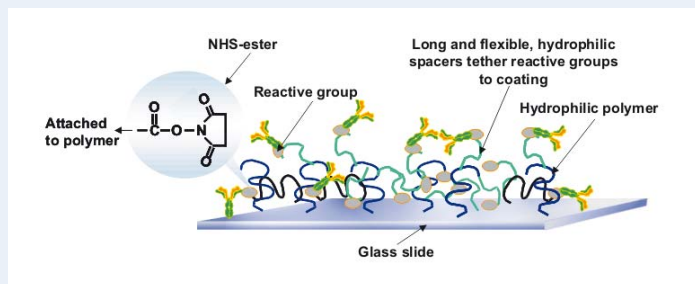
Introduction

SCHOTT launched Nexterion® Slide H as a dedicated slide surface for printing protein microarrays, as it is ideally suited for the covalent immobilization of peptides and proteins such as antibodies, antibody fragments, enzymes, or receptors. For many protein microarray applications Nexterion® Slide H has proven to be a very attractive alternative to the commonly used nitrocellulose coated slide, especially where low background, or slide transparency are important considerations. Since its introduction, the slide coating has also been successfully used with amino-modified oligonucleotides, and has become the slide of choice for printing amino-linked glycan microarrays. Carbohydrate arrays are a rapidly growing area of microarray research and Nexterion® Slide H is an excellent choice for use in the rapid screening of carbohydrate-protein interactions. The permeable, polymer coating has a large immobilization capacity, and helps to preserve the native three-dimensional structure of complex bio-molecules, thus maintaining conformation and functionality. Nexterion® Slide H produces excellent signal-to-background ratios and an exceptionally wide dynamic range compared to conventional “two-dimensional” coatings through a unique combination of low non-specific binding characteristics, and high probe loading capacity. Even very low intensity signals, such as those obtained from low-abundance analytes, or weakly expressed

genes can be reliably detected and quantified on Nexterion® Slide H. The robust coating matrix is fully compatible with commercial microarray printers and scanners. Simple and robust protocols are available making Nexterion® Slide H easy-to-use.

Immobilization chemistry

The coating on the SCHOTT Nexterion® Slide H consists of a cross-linked, multi-component polymer layer activated with N-Hydroxysuccinimide (NHS) esters to provide covalent immobilization of amine groups. All Nexterion® microarray slides are manufactured from a high quality, low-fluorescence glass coated with low-fluorescence coatings. However, the non-specific binding of assay components still remains an important contributor to the off feature background for many microarray applications. For most types of slide coatings the post-print processing protocol involves a method of adsorptive blocking to reduce non-specific binding, however, these procedures are difficult to perform in a consistent manner.



The Nexterion® Slide H coating has been engineered to exhibit a very low intrinsic non-specific background without the need for blocking. This is achieved by using a polymer based layer that is extremely resistant to non-specific binding. The polymer coating has a three-part structure; NHS-ester reactive groups are attached to the cross-linked hydrophilic polymer layer via long, flexible spacers. The terminal amino group of amino-modified nucleic acids and glycans react immediately and irreversibly with the NHS-ester groups to form a covalent bond. Proteins and other bio-molecules bind via surface-exposed amine-groups. The flexible spacers tether the immobilized bio-molecules in a quasi-liquid environment that maintains the protein specificity and chemical conformation. The high accessibility of the tethered bio-molecules facilitates interactions with their binding targets in solution.

Typical applications

- Proteome expression profiling
- Functional protein arrays
- Transcriptional profiling
- Biomarker discovery
- Detection of protein modifications
- Carbohydrate-protein interactions
- Bacteria serotyping
- Immunoassays

Suitable probe types

- Antibodies and antibody fragments
- Functional proteins such as enzymes or receptors
- Small protein fragments such as peptides
- Amino-modified oligonucleotides 16 to 70 mers
- Amino-linked glycans
- Cells

Highly reproducible coating

Nexterion® Slide H is fabricated using a proprietary thin-film deposition process optimized by SCHOTT to produce a uniform and reproducible polymer coating on one side of a high quality borosilicate glass slide. All slides are individually examined for physical defects and the presence of particles before and after coating. The surface is applied in a tightly controlled, class 100 clean room facility, resulting in coated slides with highly uniform surface properties and low auto-fluorescence.

Excellent spot morphology and signal-to-background ratios

Nexterion® Slide H provides excellent spot morphologies and reproducible spot sizes over a wide range of probe concentrations for protein, oligonucleotide and other bio-molecule microarray applications.

Product details

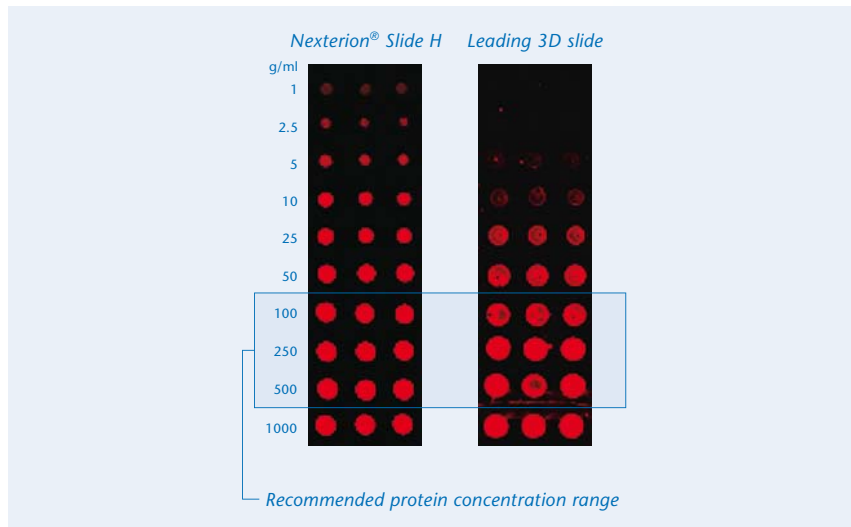


Figure shows scanned images of Nexterion® Slide H when evaluated against the market leading three-dimensional competitor slide in an anti-IgG/IgG interaction study using a range of probe concentrations.

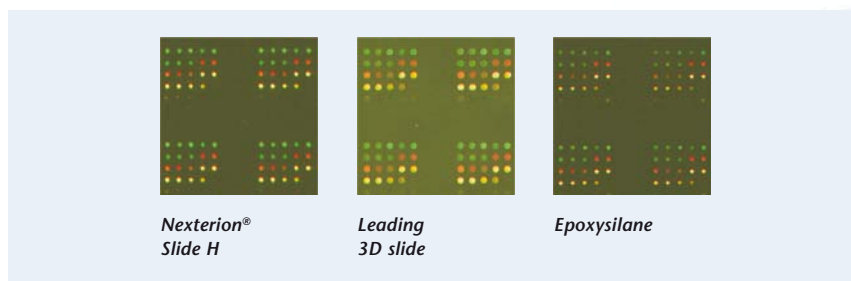


Figure illustrates the superior signal-to-background ratios on Nexterion® Slide H when compared to the market leading three-dimensional slide, and a conventional two-dimensional coating. This was demonstrated in two-color hybridization experiments following the recommended protocols, using amino-modified oligonucleotide probes.

Packaging and storage

Nexterion® Slide H are packaged in chemically stable plastic boxes and sealed under an inert atmosphere. The slides are ready-to-use from the box, and are stable for 12 months in the sealed packaging when stored at –20 °C.

Format

Nexterion® Slide H is available in packs of 25 slides with code 128 barcodes enabling automated sample tracking. The identical coating is also available in a 16-well slide format. For further information refer to the section on “Multi-well formats”.

Protocols

Separate Nexterion® Slide H protocols are available for DNA and protein microarray applications.

Compatible reagents

Process step	SCHOTT products	Alternatives	Recommended concentrations
Spotting (Protein)	Nexterion® Spot PB (1178050)	150 mM phosphate, pH 8.5, 0.01 % sarcosyl or Tween20®	Protein concentration 0.1 to 1 mg/mL
Spotting (DNA)	Nexterion® Spot (1066029)	300 mM sodium phosphate (pH 8.5) containing 0.005 % Tween20® and 0.001 % sarcosyl 150 mM sodium phosphate (pH 8.5) containing 0.001 % Tween20®	Oligonucleotides: 20 µM
Chemical deactivation of unreacted NHS-esters	–	25 mM ethanolamine in 100 mM sodium borate buffer (final pH 8.5)	
Incubation (Protein)	–	137 mM NaCl, 2.7 mM KCl, 4.3 mM Na ₂ HPO ₄ , 1.4 mM KH ₂ PO ₄ , pH 7.5 with 0.05 % Tween20®	
Hybridization (DNA)	Nexterion® Hyb (1066075) Nexterion® Oligo Hyb (1116890)	2x SSC containing 0.1 % SDS and 0.1 % salmon sperm DNA (formamide can be added if required)	

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.

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