

Multi-well formats



Introduction

Multi-well microarray substrates allow the parallel analysis of multiple biological samples against focused subsets of probes. The multi-well format permits a number of versatile assay designs such as multiplexed experiments, side-by-side comparisons, or replicate experiments to be performed. Additional advantages of using multiple well formats include higher array-to-array reproducibility and lower experimental costs when compared to conventional single-array substrates.

SCHOTT can offer multi-well coated substrates in two formats: NEXTERION® Slide MPX-16 on a 75.6 mm x 25.0 mm glass slide, or the NEXTERION® Plate MPX-96 in microplate format. The MPX-16 slide is fully compatible with standard microarray slide printers and scanners, whereas the MPX-96 conforms to the SBS standard format commonly used in clinical diagnostics and drug discovery. In addition 48-wells on slide format as well as 384-wells on plate format or customized well design can be offered on special customer request.

Formats	Immobilization method	Typical applications
Multi-well slide and SBS microplate format	Various	<ul style="list-style-type: none"> • Surveillance chips • Virus or bacterial identification • Molecular diagnostics • Dose response studies

Key product features

- Simultaneous analysis of multiple biological samples in one microarray experiment
- Excellent reproducibility compared to conventional single-array applications
- Optimal use of limited sample material
- Substantial reductions in cost per experiment
- Compatible with standard commercial printing, liquid handling and scanning equipment



Immobilization chemistry

The multi-well formats are available with functional chemistries and coatings that are identical to the standard NEXTERION® slides, allowing assays to be easily transferred to the high throughput multiwell formats. Please refer to the table in the section “General information coated slides” in order to find the appropriate coating chemistry for your specific application.

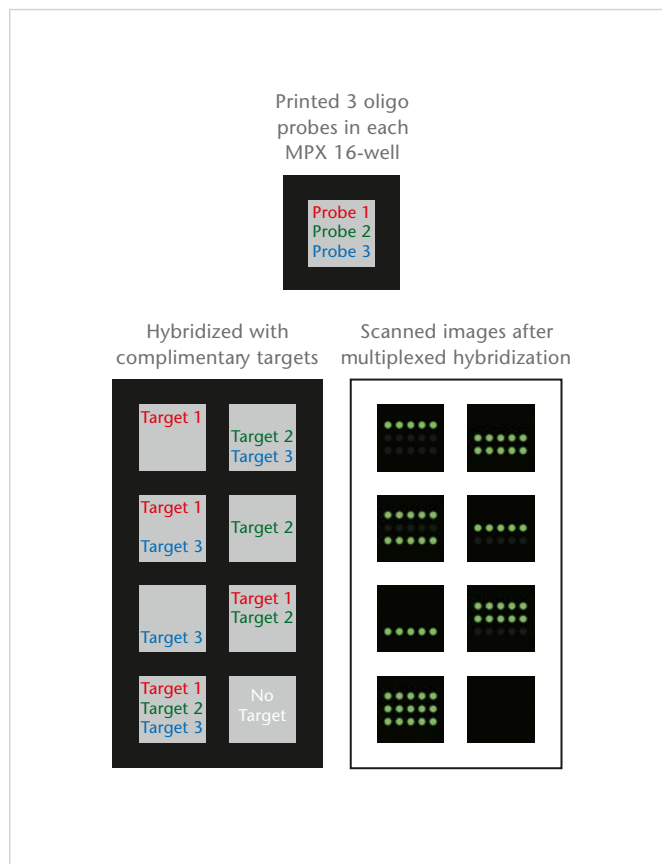
Product details

Hydrophobic multi-well patterning

The microarray quality glass substrates are printed with either a 16- or 96-well hydrophobic patterning, and are available with a number of functional coatings, making both formats suitable for most microarray applications using DNA, proteins, or other bio-molecules as probes. The black patterning material serves several important functions, one of which is to create the individual wells that act as a hydrophobic barrier preventing cross-contamination between the arrays. The well pattern also serves as a registration aid for probe deposition, and a reproducible surface for the adhesion of the silicone superstructure. The absence of well rims makes NEXTERION® multi-well substrates compatible with all microplate arrays and significantly reduces print times compared to substrates that have recessed wells. The patterned glass substrates have a corner indicator to provide an unambiguous orientation mark. Most modern robotic arrayers are equipped to handle the 96-well microplate formats, and many have pre-programmed definitions for both the NEXTERION® MPX-16 slides and NEXTERION® MTP-96 plates, simplifying the set up process for users. Information on compatible arrayers and detailed instructions for printing into the NEXTERION® multi-well formats are available on request.

Avoidance of cross-contamination

Cross-contamination is avoided during multiplexed hybridization as a result of the 16- or 96-well hydrophobic patterning, and the use of the optional self-adhesive silicone superstructures.

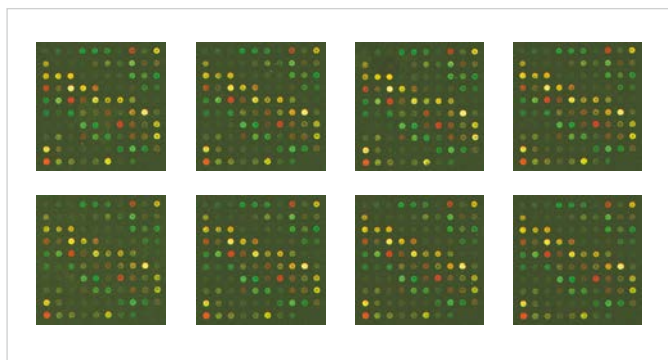


The figure shows a pictorial representation of sub-array wells from NEXTERION® Slide MPX-16 that were first printed with three distinct nucleic acid probes and then hybridized with various combinations of three complementary, fluorescent-labeled targets. No cross-contamination was observed.

Exceptional reproducibility

Intra-slide reproducibility

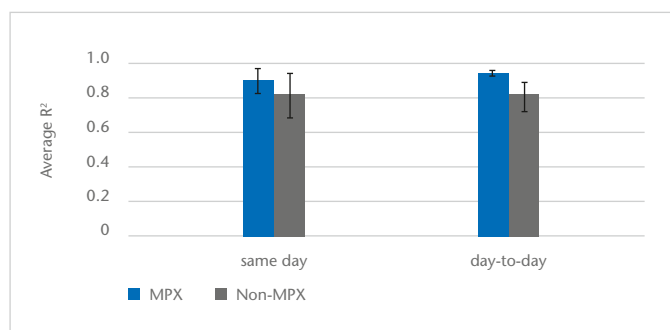
NEXTERION® multi-well coated substrates allow users to obtain microarray data with exceptionally high reproducibility. This was demonstrated by hybridizing multiple 10x10 DNA arrays with fluorescent-labeled cDNA targets using NEXTERION® Slide MPX-16. Correlation coefficients were obtained by comparing the normalized signal intensity from spots in different subarray wells. An average correlation coefficient of 0.96 was obtained after pair-wise comparison of all eight-subarray wells.



Virtually identical subarrays in each well of one NEXTERION® Slide MPX-16.

Inter-slide reproducibility

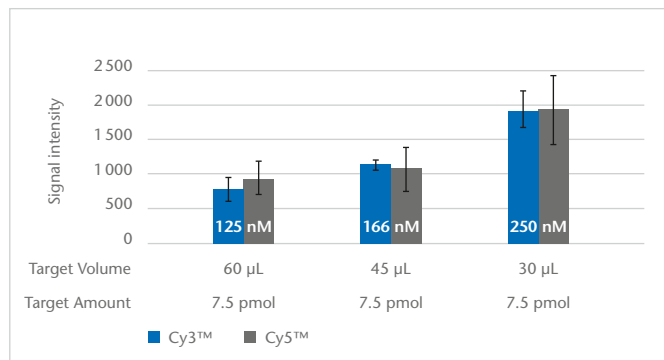
The excellent inter-slide reproducibility offered by NEXTERION® multi-well coated substrates is attributable to the simultaneous analysis of all subarray assays, and to the improved sample mixing during hybridization within the wells of the superstructure. The figure shows the average correlation coefficient (R²) obtained with NEXTERION® Slide MPX-16 (indicated as MPX) and conventional slides (indicated as Non-MPX). Experiments were conducted as described in the previous figure, but now using multiple slides for same day and day-to-day experiments.



Inter-slide reproducibility

Optimal use of limited target material

The NEXTERION® multi-well coated substrates allow lower target solution volumes to be used compared to conventional slides. The figure demonstrates that a reduction in target volume results in increased target concentration and thus in improved signal intensities. NEXTERION® Slide MPX also offers cost savings by reducing the amount of target material required without any loss of sensitivity (assuming that the target volume and target amount are reduced equally, keeping the concentration consistent).



Identical probe sets in separate subarray wells of NEXTERION® Slide MPX-16 were hybridized with 7.5 pmol of fluorescent-labeled target (house-keeping gene: GAPDH), which was dissolved in different volumes (60, 45, 30 µL) of hybridization solution.

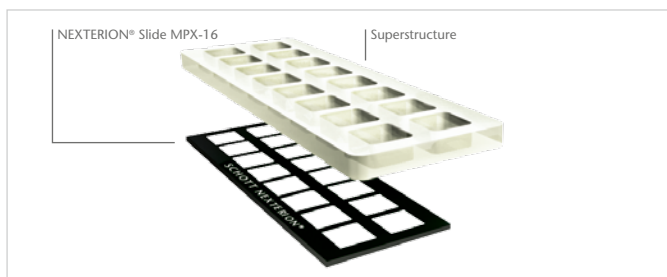
Multi-well slide format

NEXTERION® MPX Slides



Product components

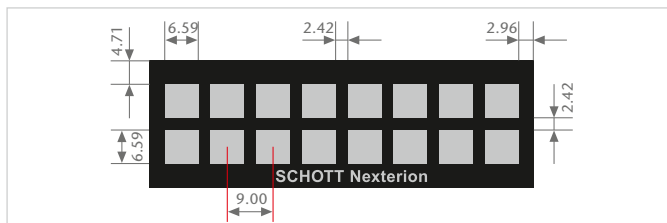
The NEXTERION® 16-well slide system consists of a 16-well patterned glass substrate and a 16-well silicon superstructure. Both components are available separately.



NEXTERION® 16-well slide system

High quality glass substrate

The ultra-flat glass slide is manufactured from low auto-fluorescent borosilicate glass with the dimensions 75.6 mm x 25.0 mm x 1.0 mm. The uniform flatness of the borosilicate glass ensures highly reproducible contact printing and scanning across the wells. The glass slides are partitioned into 16 wells by an ultra-hydrophobic patterning layer. The dimensions and locations of the wells are fully compatible with standard microarray printing parameters.



Dimensions of NEXTERION® Slide MPX-48. All dimensions are given in mm.

Introduction

The special 16-or 48-well architecture of NEXTERION® Slide MPX allows the parallel analysis of multiple biological samples against focused subsets of probes on a single slide. The dimensions of the microarray slide are fully compatible with all microarray printers and scanners, allowing users to run multiple samples on a single slide without any additional hardware.

16-well silicone superstructure and sealing strip

For NEXTERION® Slide MPX-16 a superstructure is available. It is self-adhesive, and adheres to the 16-well patterning of the glass slide, increasing the well volume to between 30 and 130 μ L. Placing the adhesive on the superstructure rather than on the glass slide avoids the risk of adhesive contamination in the wells. The clear self-adhesive sealing strip seals all the individual wells during hybridization, preventing evaporation and cross contamination. If necessary, the superstructure and sealing strip may be removed prior to scanning.

Packaging and storage

NEXTERION® MPX slides are available as 5-slide packs sealed in stable plastic boxes under an inert atmosphere. The slides are ready-to-use from the box. The exact shelf life of the slides is determined by the coating and is indicated on the external packaging. The 16-well superstructures are not included with the slides and must be ordered separately.

Key product features

- Standard slide format compatible with all standard commercial microarray printing, liquid handling and scanning equipment
- Simultaneous analysis of 16 biological samples on one slide
- Excellent reproducibility compared to conventional single-array applications

Multi-well microplate format

NEXTERION® MPX plates



Key product features

- SBS compliant microplate format
- MPX-96 glass plate easily detaches from the plate holder for spotting and scanning flexibility
- Maximum printable area of 6 mm x 6 mm allows over 1600 features per well
- High optical transmission and low auto fluorescence glass for optimal performance
- Available as a complete microtiter plate, or as individual components with or without well patterning and/or functional chemistry
- Compatible with all contact or non-contact microplate arrayers

Introduction

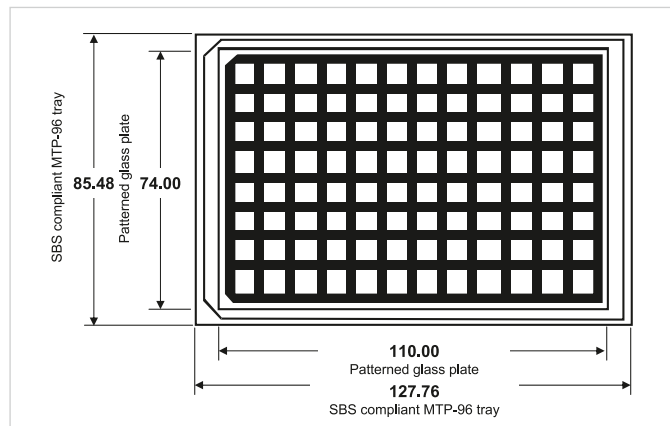
The microtiter plate format is a standard research tool for clinical diagnostics and drug discovery. The advantages associated with this format are obvious and include high sample throughput, cost savings and compatibility with automated handling systems. In recent years, there have been efforts to make the microplate format available for microarraying with the emergence of high-resolution plate scanners and compatible arrayers. However, there are clearly a number of limitations with the conventional 96-well polymer and glass bottom microtiter plates currently utilized in microarraying, including:

- Microarray print heads cannot access the entire well area because the wells are recessed. This limits the number of features per well and arrayer compatibility.
- The depth of the wells, and the subsequent additional z-axis travel, makes printing time-consuming.
- The intra-well print area may be restricted due to the contamination of the well edges with bonding adhesive. In addition, adhesive out-gassing can affect functional coating performance.
- The round wells have a smaller printable area than the square MTP wells.
- Difficulty associated with applying functional coatings to plastic plates.
- Conventional plastic microplates suffer from poor optical transparency and flatness.

The SCHOTT NEXTERION® MPX microarraying products were specifically developed to overcome these limitations. The system is designed to offer users a high degree of flexibility. The MPX-96 system consists of three main components: a microarray quality 96-well patterned glass substrate, a 96-well silicone superstructure and a microtiter plate holder and lid.

Product components

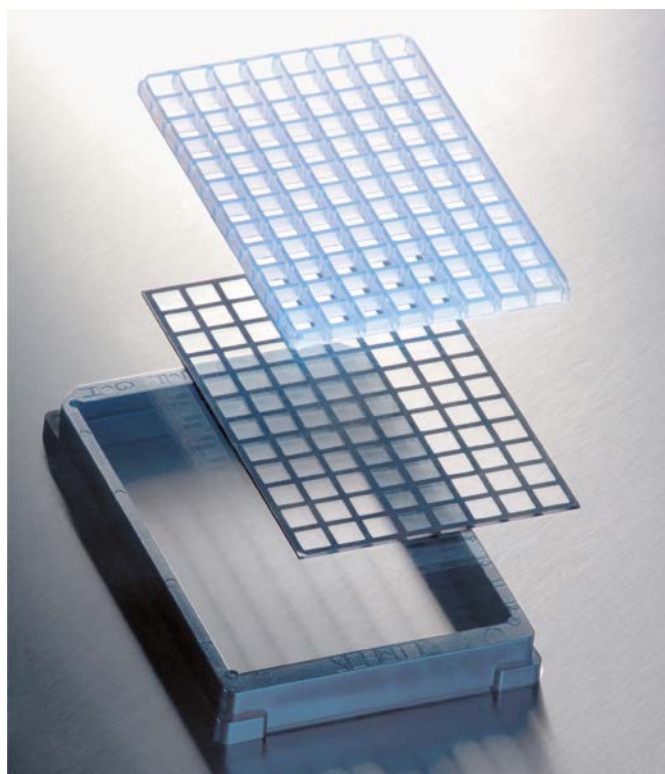
The three main components comprising the MPX-96 system are available as separate items.



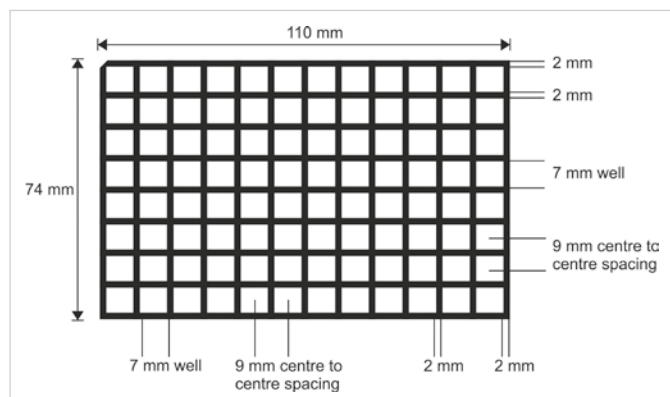
Dimensions of NEXTERION® MTP-96 system

High quality glass substrate

Microarray plate scanners typically read fluorescence signals from the spots through the bottom of the microplate. The NEXTERION® MPX glass plates have a high transmittance of over 92% and exceptional optical clarity for wavelengths used in fluorescence-based microarray assays. The plates are manufactured by melting extremely pure raw materials to ensure an excellent internal glass quality with absence of inclusions, bubbles, streaks or other defects. The ultra-flat glass plate is manufactured from low auto-fluorescent borosilicate glass measuring 74 mm x 110 mm x 1 mm. The uniform flatness of the microplate glass ensures highly reproducible contact printing and scanning across the wells. The microarray quality glass plate is available with or without a 96- or 384-well hydrophobic patterning, and with a number of functional coatings, making it suitable for all microarray applications using DNA, peptide or protein probes. The glass plate may be printed separately or in the microtiter plate tray holder.



Three components of the MTP-96 system



Dimensions of NEXTERION® plate MPX-96

96-well silicone superstructure

The optional superstructure is self-adhesive and adheres to the 96-well patterning on the glass plate, increasing the well volume to between 30 and 130 μL . Placing the adhesive on the superstructure rather than on the glass plate avoids the problem of adhesive contamination in the wells.

Microtiter plate holder with lid

The SBS (Society for Biomolecular Screening) compliant rigid polycarbonate holder supports the glass substrate and superstructure. It also acts as an alignment jig when attaching the superstructure. The holder has molded alpha/numeric well labels for easy identification. The four fixing pins firmly hold the glass plate at the four corners of the microtiter plate holder, ensuring the glass is kept flat. The lid and sealing film limit sample contamination and evaporation during processing.

Instrument compatibility

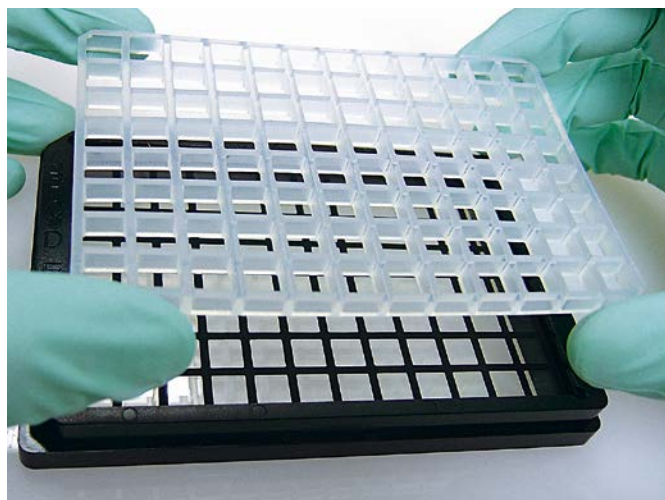
Information on compatible microplate scanners and detailed instructions for printing into the NEXTERION[®] plate MPX-96 are available on request.

Technical instructions

Further information about printing and processing the MPX plates are available in the FAQ section of the NEXTERION[®] web site.

Packaging and storage

NEXTERION[®] MPX-96 glass plates are available in 5-plate packs sealed in stable plastic boxes under an inert atmosphere. The plates are packaged separately from the plastic MPX-96 components to avoid any changes to the coatings caused by outgassing. The exact shelf life of the plates is determined by the coating and is indicated on the external packaging.



Placing superstructure on MPX 96-well plate



MPX-96 plate holder with glass plate

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 nos. 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 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 Jenaer Glas 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. To inquire about licensing under the OGT patents, please contact OGT at licensing@ogt.co.uk.

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