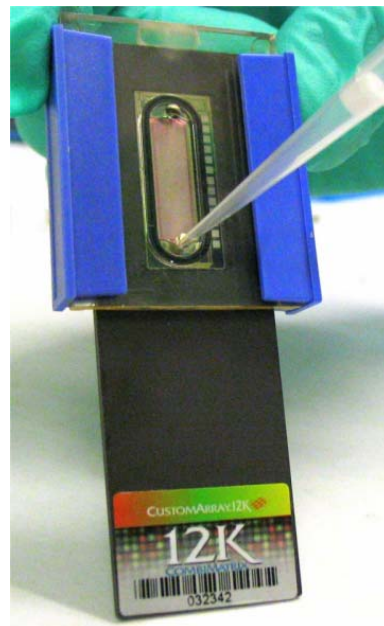
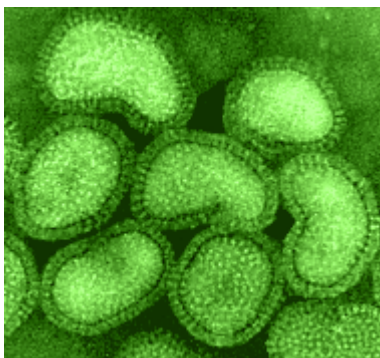


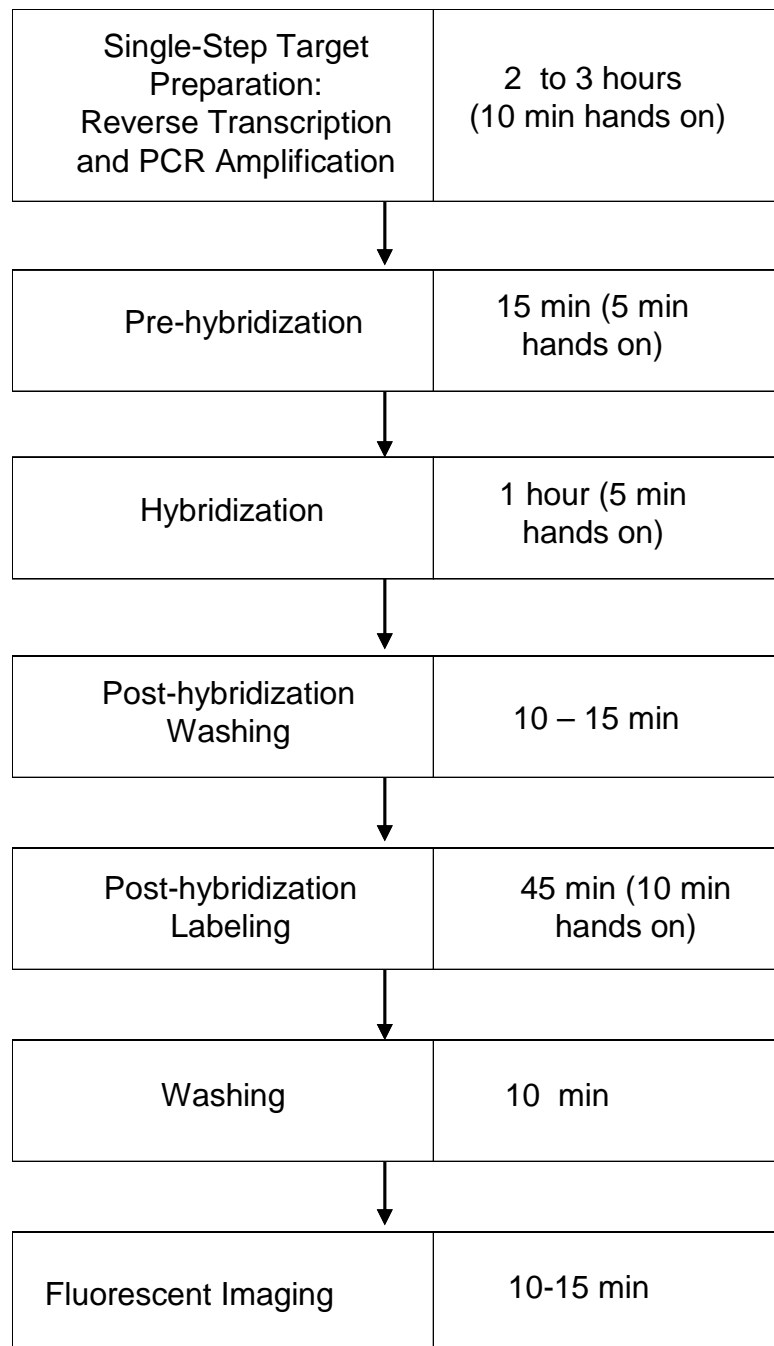
COMBIMATRIX

Influenza A Research Microarray 12K: Single-Step Target Preparation, Hybridization, and Fluorescent Imaging (PTL011)

For laboratory research use only. Not for diagnostic use.



Influenza A Microarray Research Microarray 12K: Assay Workflow



Single-Step Target Preparation, Hybridization and Imaging of Influenza A Research Microarray 12K

Table of Contents

Table of Contents	ii
Introduction	1
Materials and Equipment Provided.....	2
Materials and Equipment Required (not provided).....	2
Preparation of Single-Strand DNA targets using the CombiMatrix Influenza A Sub-typing Primer Kit	3
Influenza A Research Microarray 12K Assembly	4
Handling of Assembled Microarrays	5
Preparation for Hybridization.....	6
Pre-Hybridization	7
Hybridization	8
Hybridization Washing.....	9
Post-hybridization Blocking, Labeling, and Washing.....	10
Imaging of the Influenza A Research Microarray 12K	10
Appendix A. Related Products Available from CombiMatrix.....	12
CombiMatrix Rotisserie Holders for CustomArray™ 12K Microarrays.....	12
CombiMatrix CustomArray™ Stripping Kit and Clamp	12

Single-Step Target Preparation, Hybridization and Imaging of Influenza A Research Microarray 12K

Introduction

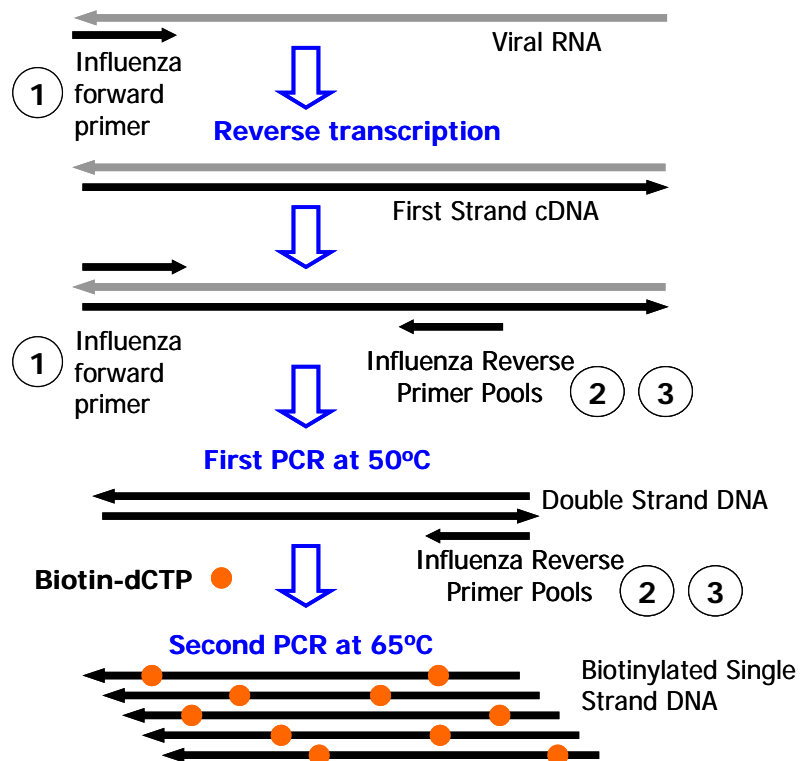
The CombiMatrix Influenza A 12K Research Microarray is a member of the CatalogArray™ product line designed for fluorescent detection of hybridization signals. The microarray is designed with oligonucleotide probes that can detect and accurately type Influenza A virus strains with hemagglutinin (HA) subtypes 1 through 16 and neuraminidase (NA) subtypes 1 through 9. This manual describes how to prepare labeled target samples of the HA and NA genes, and how to hybridize them to the Influenza A 12K Research Microarrays.

For target preparation, total RNA isolated from virus-containing material is reverse-transcribed into cDNA, which is then PCR-amplified to produce biotinylated single-strand DNA (Figure 1). Since the influenza-specific reverse primers are grouped into two different pools (to increase sensitivity of the multiplex reactions), each sample is amplified in two tubes, one for HA subtypes 1 to 7, and another one for HA subtypes 8 to 16, and NA subtypes 1 to 9. Amplification is performed as a single-step procedure including reverse transcription and PCR. CombiMatrix provides the Influenza A Sub-typing Primer Kit containing:

- Influenza A Forward primer (Tube 1);
- Two Influenza A Reverse primer pools: one for HA subtypes 1 to 7 (H1-7, Tube 2), and another one for HA subtypes 8 to 16, and NA subtypes 1 to 9 (H8-16+N1-9, Tube 3).
- Positive Hybridization Control, a biotinylated 15-mer target that hybridizes to control probes.

The PCR primers are designed based on conserved sequences to enable amplification of a wide variety of known HA and NA genes. The reverse primer pools are periodically updated to include novel virus sequences.

Figure 1. Target preparation for hybridization with the Influenza A Research Microarray. The procedure is done using CombiMatrix Influenza A Subtyping Primers Kit.



For each sample, the two amplified biotin-labeled targets are combined and hybridized to the same Influenza A Research Microarray 12K. The Influenza A Subtyping Primers Kit provides a special positive hybridization control that should be added to the hybridization buffer. The biotinylated targets hybridized to the microarray are labeled

using Cy5®-streptavidin. The hybridized Influenza A Research microarrays must be imaged using a high-resolution fluorescent scanner that is compatible with a 1"x3" slide format, with a minimum resolution of 5 µm and an adjustable focus.

The hybridized Influenza A Research Microarray 12K can be stripped of targets using the CombiMatrix CustomArray™ Stripping Kit (see Appendix A), and then re-used according to the hybridization protocol described in this manual.

Materials and Equipment Provided

IMPORTANT! Do not touch the semiconductor microarray surface. Wear gloves when handling. Microarrays can be stored in a cool dry place for up to 4 months.

- Influenza A Research Microarray 12K
- Hybridization Cap (100 µl Volume)
- Clips for Hybridization Cap
- Gasket (O-ring)
- LifterSlip™ coverslip
- Imaging Solution

Materials and Equipment Required (not provided)

- Total RNA sample (10 to 100 ng)
- CombiMatrix Influenza A Sub-typing Primer Kit for Single-Step Target Preparation, Product Number 610028
- Standard, high-resolution fluorescent microarray scanner (CombiMatrix recommends the Axon Instruments Genepix® 4000B and 4200A, and the Perkin Elmer ScanArray® 4000, 5000, Lite and Express microarray scanners).
- Thermal cycler
- SuperScript™ One-Step RT-PCR System containing the RT/ Platinum® Taq Mix (a mixture of SuperScript™ II Reverse Transcriptase and Platinum® Taq DNA Polymerase) and the 2X Reaction Mix (a buffer system optimized for reverse transcription and PCR amplification with Mg²⁺, and deoxyribonucleotide triphosphates), Invitrogen Cat. #10928-034
- 0.4 mM Biotin-14-dCTP, Invitrogen, Cat. #19518-018
- Rotisserie oven for array hybridization
- CombiMatrix CustomArray™ holders for rotisserie oven
- 95°C Heating Block
- Pipettors and nuclease-free tips
- Sterile plastic ware
- Gloves (powder-free)
- Adhesive tape: Scotch® Brand Magic® Transparent Tape is suitable for hybridization temperatures of 50°C or less; for extended incubations at higher temperatures use a PCR sealing tape such as Nunc Brand PN 232702 (clear polyolefin liner) or PN 276014 (aluminum liner)
- Nuclease-free water
- 50x Denhardt's solution
- 0.5 M EDTA (pH 8.0)
- 10% Tween-20
- 20x SSPE Buffer
- 1% SDS
- 10x Phosphate Buffered Saline (PBS: 1.37M Sodium Chloride, 0.027M Potassium Chloride, 0.08M Sodium Phosphate dibasic, 0.02M Sodium Phosphate monobasic, pH 7.4; Ambion, Cat.# 9625)
- 5X PBS-Casein Blocking Buffer, BioFX Laboratories, Cat.# PBSC-0100-01
- Biotin detection reagent: Fluorolink™ Cy5®-labeled streptavidin, GE Healthcare/Amersham Biosciences Cat. # PA45001

Preparation of Single-Strand DNA targets using the CombiMatrix Influenza A Sub-typing Primer Kit

We recommend starting with high-quality, intact total RNA that has been purified using a Qiagen RNeasy kit or equivalent. Please note that the presence of impurities (such as traces of phenol) may result in inhibition of the reverse transcription reaction. We recommend using 10 -100 ng of purified total RNA suspended in 5 µl of nuclease-free water.

1. For each sample, prepare two reaction mixtures in PCR tubes on ice. Keep all components and samples on ice.

Reagent	H1-7 (50 µl)	H8-16+N1-9 (50 µl)
Nuclease-free water	13.5 µl	13.5 µl
Template (total RNA)	2.5 µl	2.5 µl
2X Reaction Mix	25 µl	25 µl
0.4 mM Biotin-14-dCTP	5.0 µl	5.0 µl
Influenza Forward primer (Tube 1)	1 µl	1 µl
Influenza Reverse Primer Pool (Tubes 2 and 3)	H1-7: 2 µl	H8-16+N1-9: 2 µl
RT/ Platinum® Taq Enzyme Mix	1 µl	1 µl
Total Volume	50 µl	50 µl

2. Mix well by pipetting, and spin the tube 5 seconds in a microcentrifuge.
3. Place into the thermal cycler preheated to 40°C and immediately start the RT-PCR amplification program as follows:

Reverse Transcription:

- 10 minutes at 40°C
- 20 minutes at 50°C
- 3 minutes at 94°C

1st PCR of 30 cycles, each cycle as follows:

- Denature at 94°C for 30 seconds,
- Anneal at 50°C for 45 seconds,
- Extend at 72°C for 30 seconds.

2nd PCR of 40 cycles, each cycle as follows:

- Denature at 94°C for 30 seconds,
- Anneal and extend at 65°C for 60 seconds.

NOTE: The time required for target preparation can be reduced by decreasing the number of PCR cycles, (25 instead of 30 for the 1st PCR, and 30 instead of 40 for the 2nd PCR).

4. Perform a final incubation at 72°C for 5 minutes.
5. Maintain the reactions at 4°C after cycling.
6. Spin the tubes briefly in a microcentrifuge to collect condensate.

IMPORTANT! We recommend that you analyze the amplification products by gel electrophoresis. Run a 2 μ l aliquot of each reaction on an acrylamide gel, or a 5 μ l aliquot on an agarose gel. The reaction should show one or more bands at approximately 500-600 bp. Since the reverse primer pools consist of multiple primers, and the PCR conditions are not stringent, you should expect to see multiple bands. The presence of additional bands of various sizes will not adversely affect the results of microarray hybridization.

Influenza A Research Microarray 12K Assembly

IMPORTANT! The Hybridization Cap, Clips and O-ring are intended for single use only.

1. Insert the provided Gasket (O-ring) in the groove of the Hybridization Cap.
2. Align the Hybridization Cap over the slide so that the top edge of the slide is flush against the stop on the Hybridization Cap, and the Cap is centered over the semiconductor area.
3. Secure the Hybridization Cap in place with the Clips provided (see Fig. 1 and 2). The Clips can be attached only if the Hybridization Cap is positioned with the top edge of the slide at the stop of the Cap.

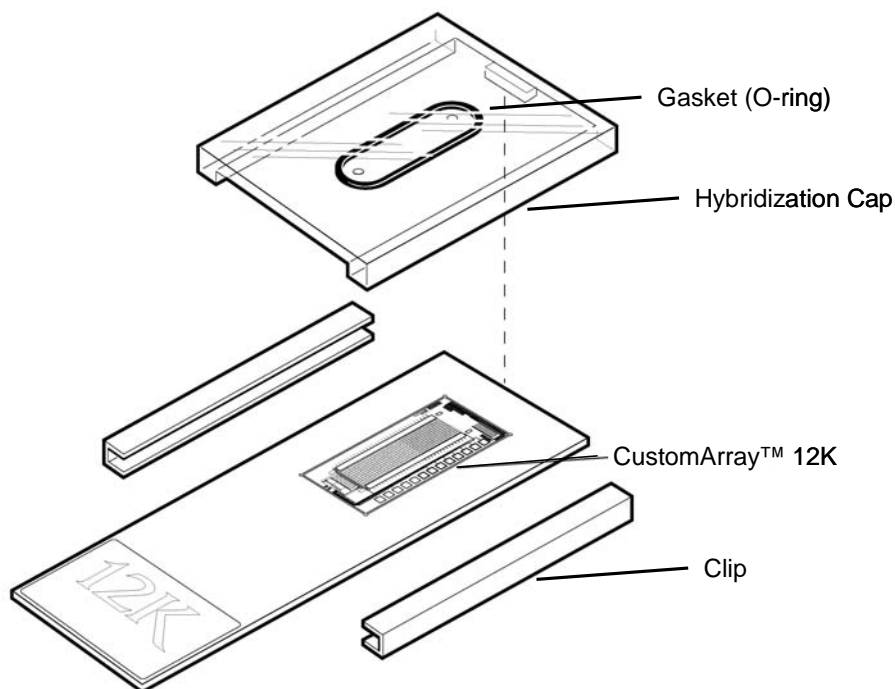
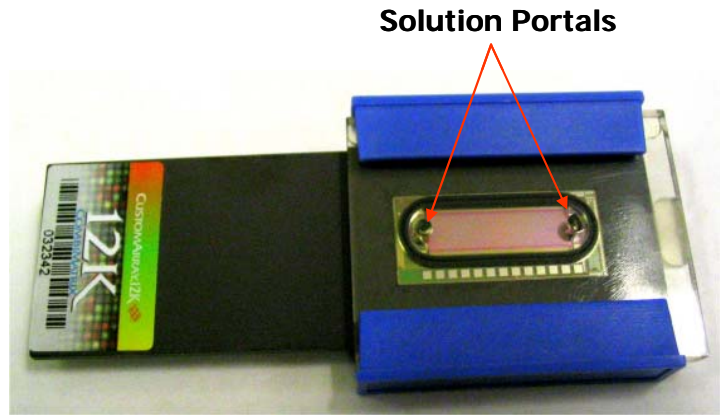


Figure 2. CustomArray™ 12K microarray, Hybridization Cap and Clips.

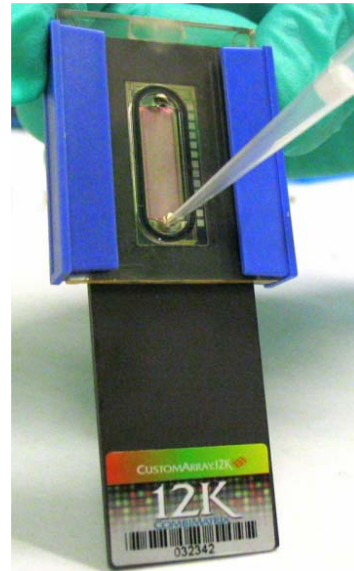
Figure 3. Assembled Influenza A Research Microarray with Hybridization Cap and Clips.



Handling of Assembled Microarrays

1. Wear gloves at all times when handling microarrays and reagents.
2. After an Influenza A Research Microarray is assembled and re-hydrated, keep the Hybridization Cap in place during all hybridization and wash steps. Do NOT allow the semiconductor microarray surface to become dry at any step in the protocol.
3. To add or remove solutions, hold the microarray with the assembled Hybridization Cap at a 45 degree angle. Use a 200 μ l micropipettor with nuclease-free pipet tips to add or remove solutions through the lower solution portal of the Hybridization Cap (see Fig. 4).

Figure 4. Pipetting of solutions into the hybridization chamber of the assembled Influenza A Research Microarray.



4. Proceed rapidly when changing solutions during the pre-hybridization, hybridization, and washing steps. Do not leave the hybridization chamber empty for any significant length of time.
5. For all incubations longer than 10 minutes, seal the solution portals of Hybridization Cap with non-permeable adhesive tape to prevent evaporation. Use Scotch® Brand Magic® Transparent Tape for hybridization temperatures of 50°C or less; for higher temperatures use a PCR sealing tape such as Nunc Brand PN 232702 (clear polyolefin liner) or PN 276014 (aluminum liner). Wipe the surface clean with a lint-free tissue before sealing.
6. If you plan to strip and re-use a hybridized Influenza A Research Microarray (see Appendix A), do not allow the semiconductor surface to dry, as this will prevent any further use. Avoid prolonged storage of hybridized microarrays prior to stripping; instead, strip the microarrays, then store them wet in Imaging Solution or 1X PBS at 4°C for a maximum of 2 weeks.

Preparation for Hybridization

The following solutions can be prepared beforehand. All reagents used should be RNase and DNase-free.

1. Prepare the **2X Hyb Solution Stock** (see Table 1). This stock will be used for preparation of the Pre-hybridization and Hybridization Solutions. The 2X Hyb Solution Stock should be filter-sterilized and can be stored at room temperature for up to six months.

Table 1. 2X Hyb Solution Stock		
<u>Reagent</u>	Volume for 10 ml	Final Concentration
20X SSPE	6 ml	12X
10% Tween-20	100 µl	0.1%
0.5M EDTA	560 µl	40mM^a
Nuclease-free water	3.34 ml	
Total Volume	10 ml	

^a Final concentration includes the EDTA from the SSPE.

2. Prepare the **Wash Solutions** (see Table 2). The prepared solutions should be filter-sterilized and can be stored at room temperature for up to 6 months.

Table 2. Wash Solutions		
<u>Step</u>	Solution	For 10 ml
6X SSPET Wash	6X SSPE, 0.05% Tween-20	3 ml 20X SSPE 50 µl 10% Tween-20 6.95 ml Nuclease-free water
3X SSPET Wash	3X SSPE, 0.05% Tween-20	1.5 ml 20X SSPE 50 µl 10% Tween-20, 8.45 ml Nuclease-free water
0.5X SSPET Wash	0.5X SSPE, 0.05% Tween-20	250 µl 20X SSPE 50 µl 10% Tween-20 9.7 ml Nuclease-free water
PBST Wash	2X PBS, 0.1% Tween-20	2 ml 10X PBS, 100 µl 10% Tween-20, 7.9 ml Nuclease-free water
PBS Wash	2X PBS	2 ml 10X PBS, 8 ml Nuclease-free water

3. Prepare the 1 mg/ml stock solution of the Fluorolink™ Cy5®-labeled streptavidin. This reagent is supplied as a lyophilized powder. Dissolve in 1.0 ml of nuclease-free water, dispense into 10-20 µl aliquots, freeze and store at -20°C. Avoid repeated freeze-thaw cycles.

Pre-Hybridization

NOTE: The Influenza A Research Microarray must be re-hydrated prior to hybridization. A pre-hybridization step is recommended to block non-specific binding of targets.

1. Pre-heat an incubator to 65°C.
2. Set a hybridization rotisserie oven to 45°C.
3. Prepare the fresh **Pre-hybridization Solution** (see Table 3).

Table 3. Pre-hybridization Solution		
Reagent	Volume for 100 µl	Final Concentration
2X Hyb Solution Stock	50 µl	6X SSPE, 0.05% Tween-20, 20mM EDTA
Nuclease-free water	35 µl	
50X Denhardt's solution	10 µl	5X
1% SDS	5 µl	0.05%
Total Volume	100 µl	

4. Fill the hybridization chamber with nuclease-free water. Avoid introducing air bubbles into the chamber. Cover the solution portals with adhesive tape to prevent evaporation. Incubate at 65°C for 10 min. Remove the microarray from the incubator and bring to room temperature. Remove the adhesive tape and aspirate the water out of the hybridization chamber.
5. Fill the hybridization chamber with the Pre-hybridization Solution. Mix gently by pipetting. A small air bubble can be introduced to improve the mixing process if the arrays are rotated during incubation. Cover the solution portals with adhesive tape.
6. Load the microarray onto the rotisserie in the hybridization oven and incubate at 45°C for 15 minutes with gentle rotation.

NOTE 1: To improve microarray performance, use of a rotisserie oven or a rotating incubator is recommended to ensure mixing during hybridization. The Influenza A Research Microarray can be attached to standard rotisseries using holders available from CombiMatrix (see Appendix A).

NOTE 2: The hybridization temperature is recommended specifically for the Influenza A 12K Microarray. Other CombiMatrix microarray designs may require different temperatures.

Hybridization

1. For each sample, mix together 15 μl aliquots of the purified target samples (H1-7 and H8-16+N1-9).
2. Denature the HA-NA target mixture by incubating at 95°C for 5 min
3. Place on ice.
4. When you are ready to hybridize microarrays, prepare the **Hybridization Solution** as shown in Table 4.

NOTE: The Hybridization Solution should be prepared fresh each time you hybridize microarrays.

Table 4. Hybridization Solution		
Reagent	Volume for 100 μl	Final Concentration
Denatured HA and NA target mixture	30 μl	
2X Hyb Solution Stock	50 μl	6X SSPE, 0.05% Tween-20, 20mM EDTA
1% SDS	5 μl	0.05%
Positive Hybridization Control from the CombiMatrix Influenza A Sub-typing Primer Kit	2 μl	
Nuclease-free water	13 μl	
Total Volume	100 μl	

5. Spin down the solution in a microcentrifuge for 5 seconds at maximum speed.
6. Remove the adhesive tape from the microarray and pipet the Pre-hybridization Solution out of the hybridization chamber.
7. Fill the hybridization chamber with the Hybridization Solution and mix gently with repeated pipetting. A small air bubble can be introduced to improve the mixing process if the arrays are rotated during hybridization.

NOTE: Usually the volume of Hybridization Cap is slightly less than 100 μl . You can adjust the final volume of Hybridization Solution to 95 instead of 100 μl , if you want the whole amount to fit into the chamber.

8. Carefully wipe excess solution from the surface of the Hybridization Cap with a lint-free tissue, and cover the solution portals with adhesive tape.
9. Load the microarray onto the rotisserie in the hybridization oven and incubate at 45°C for 1 hour with gentle rotation.

Hybridization Washing

IMPORTANT! Do not allow the microarray to become dry at any time. Proceed rapidly when changing solutions. Do not leave the hybridization chamber empty for any significant length of time.

- NOTE 1: Keep the Hybridization Cap in place during all wash steps. Hold the microarray with the assembled Hybridization Cap at a 45° angle (as shown in Figure 4), and add/remove solutions through the lower solution portal by using a 200 µl micropipette with nuclease-free pipet tip.
- NOTE 2: For every wash step, CombiMatrix recommends rinsing the hybridization chamber with the corresponding Wash Solution prior to the wash incubation. Add the Wash Solution to the chamber, gently mix by pipetting, remove it, and fill the chamber again with the same solution.
- NOTE 3: With the exception of the first wash step, microarrays should be incubated in the Wash solutions for a minimum of 30 seconds. However, if processing multiple arrays, extend the wash incubation time until all hybridization chambers are rinsed and filled.
-

1. Prior to starting the wash procedure, preheat the **6X SSPET Wash** solution to 45°C.
2. Remove the microarray from the hybridization oven. Remove the adhesive tape, and pipet the Hybridization Solution out of the chamber.
3. Using the pre-heated **6X SSPET Wash** solution, rinse the hybridization chamber, fill the chamber, cover the portals with adhesive tape, and return the microarray to the hybridization oven for 5 minutes (with gentle rotation). Remove the 6X SSPET Wash solution from the hybridization chamber.
4. Using the **3X SSPET Wash** solution, rinse the hybridization chamber, fill the chamber, and incubate the microarray at room temperature for 30 seconds (see NOTE 3). Remove the 3X SSPET Wash solution from the hybridization chamber.
5. Using the **0.5X SSPET Wash** solution, rinse the hybridization chamber, fill the chamber, and incubate the microarray at room temperature for 30 seconds. Remove the 0.5X SSPET Wash solution from the hybridization chamber.
6. Using the **PBST Wash** solution, rinse the hybridization chamber, fill the chamber, and incubate the microarray at room temperature for 30 seconds. Remove the PBST Wash Solution from the hybridization chamber.
7. Using the **PBS Wash** solution, rinse and fill the hybridization chamber. Retain the PBS Wash Solution in the hybridization chamber until you are ready to proceed to the Post-hybridization Blocking step.

Post-hybridization Blocking, Labeling, and Washing

1. Bring the 5X PBS-Casein Blocking Buffer to room temperature.
2. Remove the PBS Wash Solution from the hybridization chamber.
3. Add the 5X PBS-Casein Blocking Buffer to the hybridization chamber. Cover the solution portals with adhesive tape, and incubate the microarray at room temperature for 15 minutes.
4. Prepare the **Dye Labeling Solution** using the 1 mg/ml stock solution of the Fluorolink™ Cy5®-labeled streptavidin. Thaw an aliquot, and make a 1:1000 dilution (v/v, 1 µl per 1 ml) in the 5X PBS-Casein Blocking Buffer. Discard the used aliquot.

NOTE: Prepare the Dye Labeling Solution fresh each time you hybridize arrays.

5. Remove the 5X PBS-Casein Blocking Buffer, and fill the hybridization chamber with the **Dye Labeling Solution**. Cover the solution portals with adhesive tape, and incubate the microarray at room temperature for 30 minutes. Protect the microarray from light by covering with aluminum foil. Remove the Dye Labeling Solution from the hybridization chamber.
6. Using the PBST Wash solution, rinse the hybridization chamber, fill it, and incubate the microarray at room temperature for 1 minute. Repeat the PBST Wash one more time.
7. Using the PBS Wash solution, rinse the hybridization chamber, fill it, and incubate the microarray at room temperature for 1 minute. Repeat the PBS Wash one more time.
8. Retain the PBS Wash Solution in the hybridization chamber until you are ready to proceed to the Imaging step.

Imaging of the Influenza A Research Microarray 12K

IMPORTANT! The Influenza A Research Microarray 12K must be scanned wet using the Imaging Solution supplied. The LifterSlip™ coverslip provided with the microarray has been specifically designed to retain the Imaging Solution without contacting the microarray surface. Do not use a standard coverslip.

NOTE: The Imaging Solution contains phosphate buffer, which may precipitate during shipping. If a precipitate is visible, heat the Imaging Solution at 60-70°C for about 5 minutes until it dissolves. Allow the solution to cool to room temperature before applying it to the microarray.

1. Remove the PBS Wash Solution from the hybridization chamber.
2. Carefully remove the hybridization chamber from the microarray by removing the clips and lifting the Hybridization Cap off the slide surface.
3. Immediately cover the semiconductor microarray surface with the Imaging Solution.
4. Using thin-tipped forceps, pick up a fresh LifterSlip™ and hold it so that the raised edges face the microarray. The raised edges can be detected by gently rubbing an edge with the tip of the forceps – the raised edge will feel rougher than the glass surface.

5. Lay the LifterSlip™ at an angle onto the microarray so that it is centered over the semiconductor area (see Fig. 5). First touch the Imaging Solution with one side of the LifterSlip™, then slowly lower the slip down, taking care not to introduce air bubbles. If bubbles still form, lift one side of the LifterSlip™ with forceps (or a razor blade) to let the bubbles out, and lower it down again.

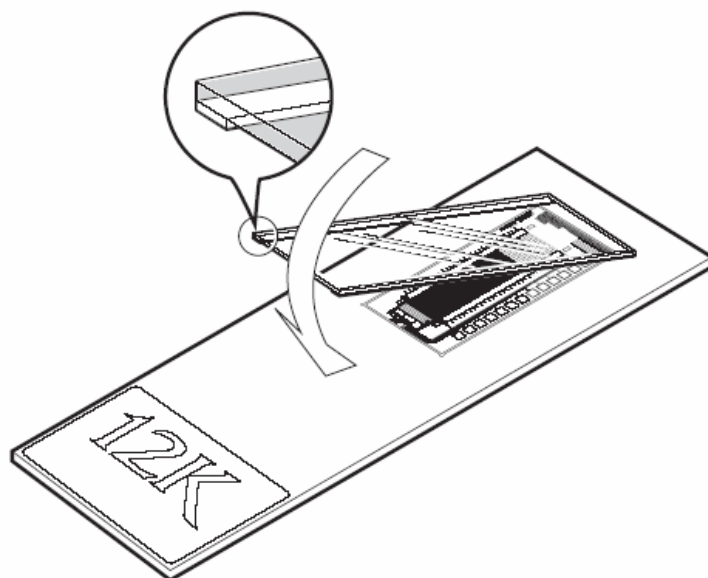


Figure 5. Microarray with LifterSlip™ coverslip

6. Carefully remove any excess Imaging Solution from the edge of the LifterSlip™ using a lint-free tissue, until it is resting evenly over the microarray.
7. Load the microarray into the scanner, taking care not to disturb the LifterSlip™ coverslip. Follow the manufacturer's recommendations for loading the slide into the scanner.
8. After you complete the scan, save the image as a .tiff image file. The data can be extracted from the image using the CombiMatrix Microarray Imager Software. Please refer to the Quick Start Guide or the Microarray Imager User's Manual on our web site (<https://webapps.combimatrix.com>).
9. After imaging is completed, you can proceed to stripping of the microarray for subsequent re-hybridization using the CombiMatrix CustomArray™ Stripping Kit (see Appendix A). Do not allow the semiconductor microarray surface to dry; keep the microarray wet, either in a tube or slide-holder containing 1X PBS, or in the Imaging Solution with the LifterSlip™ attached. Avoid prolonged storage of hybridized microarrays prior to stripping; instead, first strip the microarray, then store wet in Imaging Solution or 1X PBS at 4°C for a maximum of 2 weeks.
10. CombiMatrix has created an Excel worksheet for Influenza A sub-typing. This tool can be obtained by contacting support@combimatrix.com or calling CombiMatrix Customer Support at 800-985-2269.

Appendix A. Related Products Available from CombiMatrix

CombiMatrix Rotisserie Holders for CustomArray™ 12K Microarrays

To improve microarray performance, use of a rotisserie oven or a rotating incubator is recommended to ensure mixing during hybridization. 12K microarrays can be attached to standard rotisseries using holders available from CombiMatrix.

- **CombiMatrix 32 CustomArray™ Rotisserie for 12K**, Product Number 610004.

The 32 CustomArray™ Rotisserie for 12K allows the hybridization of 32 microarrays at a time. The unit has four plates, each with eight positions, using ball detent clamps to secure the microarray. The 32 CustomArray™ Rotisserie is supplied with a shaft that fits most Fisher Scientific Isotemp Hybridization Incubators. The Rotisserie can also be adapted to fit other incubation ovens.

- **CombiMatrix 8 CustomArray™ Rotisserie for 12K:**
 - Product Number for 3/8" diameter shaft is 610012,
 - Product Number for 3/4" diameter shaft is 610013.

The 8 CustomArray™ Rotisserie for 12K allows the hybridization of 8 microarrays at a time. The Rotisserie will mount on a 3/8" or 3/4" diameter shaft of an incubation oven with simple spring clasps. The model can also be adapted to fit other incubation ovens. Please indicate the oven shaft diameter when ordering.

- **CombiMatrix 3-Tray CustomArray™ for 12K**, Product Number 610008.

CustomArray™ 3-Tray for 12K is a three-position tray that fits in place of the standard Affymetrix tray/bucket in an Affymetrix oven. As many as eight trays can be loaded in one oven, allowing for 24 of 12K microarrays to be hybridized at a time.

CombiMatrix CustomArray™ Stripping Kit and Clamp

- **CombiMatrix CustomArray™ Stripping Kit**, Product Number 610011.

The Stripping Kit enables re-hybridization of a CombiMatrix microarray three times, thus allowing the use of a single microarray up to four times. Each kit contains reagents for 25 stripping reactions, plus accessories for subsequent re-hybridizations.

- **CombiMatrix CustomArray™ Stripping Clamp**, Product Number 610010.

The CombiMatrix Stripping Clamp is designed to be used with the **CombiMatrix CustomArray™ Stripping Kit**. The CustomArray™ Stripping Clamp is recommended for up to 100 stripping procedures.