Antibody Microarray

User's Guide

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INTRODUCTION

Antibody microarrays are a high-throughput, ELISA-based platform for protein expression profiling and comparative analysis of normal, diseased, and treated samples. Full Moon BioSystems' antibody arrays enable the simultaneous detection of hundreds of native proteins on a single glass slide, allowing efficient use of samples while reducing experimental variability. Available array formats include phospho-specific arrays for phosphorylation analysis, broad exploratory arrays for large-scale protein profiling, and pathway-focused arrays targeting specific biological processes. Compatible sample types include cell lysates, fresh, frozen, or FFPE tissue lysates, serum, culture supernatants, and other bodily fluids.

ANTIBODY ARRAYS

The antibodies on the array are covalently immobilized on a high-quality glass surface coated with proprietary three-dimensional polymer materials to ensure high binding efficiency and specificity. Arrays are printed on standard-size microscope slides, with one complete array per slide, and use fluorescent detection compatible with microarray scanners that support 76 × 25 × 1 mm (3 in × 1 in × 1 mm) slides.

Each array set includes two identical slides, allowing parallel analysis of control and treated samples. To enhance data reliability, antibodies are printed in replicate. Pathway arrays with fewer than 400 antibodies include six replicates per antibody, while larger formats, such as the Explorer, Phospho Explorer, and Signaling Explorer arrays, include two replicates per antibody.

Each array also incorporates internal controls, including Cy3-labeled positive markers to define array boundaries, beta-actin and GAPDH as positive controls, and BSA as a negative control. Empty spots contain no material and may be used to estimate background signal during data analysis.

Detailed antibody lists, reactivity information, and array layouts are available on the Full Moon BioSystems website under the Documentation section for each specific array.

ANTIBODY ARRAY ASSAY KIT

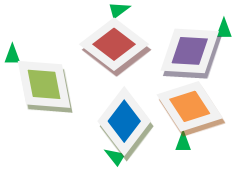
The Antibody Array Assay Kit is designed for reliable processing of Full Moon BioSystems' antibody arrays and provides the core reagents required for protein extraction, biotinylation, conjugation, and detection. All reagents are optimized for use with Full Moon BioSystems' antibody array platform to ensure consistent performance.

The 2-reaction kit contains sufficient reagents to process two array slides, while the 20-reaction kit provides reagents for processing up to 20 slides.

HOW IT WORKS

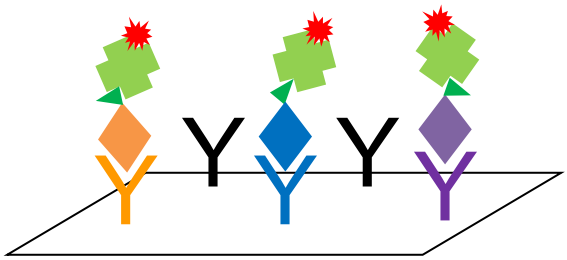
Protein Extraction from cells, tissues, or bodily fluids

Biotinylation of Proteins



Protein Conjugation to Antibody Array

Detection by Dye-Streptavidin



The ELISA-based antibody array platform consists of four main steps: (1) protein extraction using a non-denaturing extraction buffer; (2) biotinylation of protein samples; (3) incubation of labeled samples with the antibody array; and (4) detection using dye-conjugated streptavidin.

Proteins are not denatured during the assay, preserving native tertiary structures and multiprotein complexes. As a result, some target epitopes may be inaccessible, which can lead to incomplete detection of certain proteins and an increased likelihood of false-negative results.

EXPERIMENTAL CONSIDERATIONS

- All reagents and materials are intended for research use only.
- Handle antibody array slides by the barcode-labeled area only. Do not touch the array surface.
- Exercise care throughout the assay, as variations in buffers, operators, pipetting technique, washing procedures, incubation time, or temperature may affect assay performance.
- Use only the reagents and materials specified in this user's guide. Substitution with reagents from other sources is not recommended.
- Do not allow arrays to dry during blocking, coupling, or washing steps, as this may result in elevated background signal.
- Wash arrays thoroughly with Wash Solution followed by water to ensure complete removal of residual reagents from the slide surface.
- The Antibody Array Assay Kit does not contain protease inhibitors. To minimize protein degradation, proceed promptly through the extraction and assay steps once sample preparation has begun. Protease inhibitors may be added if desired or if protein samples will be stored for one week or longer.
- Always wear gloves when handling reagents and array slides.

COMPONENTS

Antibody Arrays

Material/Reagent	Quantity	Purpose	Storage Condition
Antibody Microarray	2 slides	Microarray	4°C (6 months)

Antibody Array Assay Kit (sold separately from the arrays)

Catalog No.	Description	Quantity
KAS02	Antibody Array Assay Kit, 2 Reactions	2 Reactions
KAS20	Antibody Array Assay Kit, 20 Reactions	20 Reactions

Material/Reagent	Quantity		Purpose	Storage Condition
	2-Rxn Kit	20-Rxn Kit		
Biotin Reagent	1 mg	5 x 1mg	Labeling	-20 °C
Blocking Reagent	60 mL	600 mL	Blocking	4 °C
Coupling Chamber	1	5	Coupling	RT
Coupling Reagent	12 mL	120 mL	Coupling	-20 °C
Detection Buffer	60 mL	600 mL	Detection	4 °C
DMF	200 uL	1 mL	Labeling	4 °C
Dry Milk	1.8g & 0.36g	18g & 3.6g	Coupling	4 °C
Extraction Buffer	1.5 mL	15 mL	Cell and tissue lysis	4 °C
Labeling Buffer	2 mL	20 mL	Labeling	4 °C
Lysis Beads	2 tubes	20 tubes	Cell and tissue lysis	RT or 4 °C
Spin Columns	2 sets	20 sets	Buffer Exchange	RT
Stop Reagent	100 uL	1 mL	Stop labeling reaction	4 °C
10X Wash Buffer	100 mL	500 mL x 2	Washing	4 °C

ADDITIONAL MATERIALS REQUIRED

- 1X PBS, pH=7.4
- Dye conjugated streptavidin
  - Cy3-Streptavidin is commonly used. It can be purchased from any vendors including GE Healthcare: PA43001; Invitrogen: S-32355 (Alexa Fluor 555 streptavidin); Sigma-Aldrich: S6402, and others.
  - Other dye labeled streptavidin can be used in place of Cy3-streptavidin provided that the dye is compatible with your array scanner.
- 50 mL conical tube with cap
- Centrifuge with 1.5 mL microtube rotor
- Milli-Q Grade Water or dd H_2O
- Orbital shaker
- Petri dishes, 100 x 15mm (9 cm in diameter)
- Slide drying (only one option is required)
  - By compressed air: compressed nitrogen/clean air supplied by a central line or from a cylinder. Do not use canned air duster or heat gun/blower.
  - By centrifugation: centrifuge with 50 mL swinging bucket rotor
- UV Spectrophotometer
- Vortexer
- Microarray scanner compatible with 3 x 1 inch (76 x 25 mm) slides

  If a compatible scanner is not available, finished array slides may be sent to Full Moon BioSystems for scanning. For details, visit www.fullmoonbio.com/services/array-scanning-image-analysis/.

REAGENT PREPARATION

REAGENTS TO WARM BEFORE USE

Blocking Reagent **Coupling Reagent** **Wash Buffer**	**Warm to 25-30°C in a water bath.**
Biotin **Detection Buffer** **DMF** **Dry Milk** **Labeling Buffer** **Stop Reagent**	**Warm to room temperature.**

2-RXN Kit (KAS02)

1	1X Wash Solution	Make 1:10 dilution. In a one-liter reagent bottle, add 100 mL of 10X Wash Buffer to 900 mL of dd H_2O. Shake to mix.
2	Blocking Solution	Add 1.8 g of Dry Milk to 60 mL of Blocking Reagent. Shake to mix. Be sure the milk powder is completely dissolved. **Use within one week.**
3	Coupling Solution	Add 0.36 g of Dry Milk to 12 mL of Coupling Reagent. Shake to mix. Be sure the milk powder is completely dissolved. **Use within one week.**

20-RXN Kit (KAS20)

1	1X Wash Solution	Make 1:10 dilution. For example, add 100 mL of 10X Wash Buffer to 900 mL of ddH_2O to make 1L of 1X Wash Solution. Shake to mix.
2	Blocking Solution	If you plan to perform 20 assays within one week, add 18 g of Dry Milk to 600 mL of Blocking Reagent. Be sure the milk powder is completely dissolved. For two assays, aliquot 60 mL of Blocking Reagent and add 1.8 g of Dry Milk. Shake to mix. **Use within one week.**
3	Coupling Solution	If you plan to perform 20 assays within one week, add 3.6 g of Dry Milk to 120 mL of Coupling Reagent. Be sure the milk powder is completely dissolved. For two assays, aliquot 12 mL of Coupling Reagent and add 0.36 g of Dry Milk. Shake to mix. **Use within one week.**

ASSAY PROTOCOL – DETECTION BY CY3-STREPTAVIDIN

IMPORTANT – PREPARE AND WARM REAGENTS BEFORE USE

A. Sample Preparation

Note: Cell and tissue lysates should be prepared using the mild, non-denaturing Extraction Buffer provided in the Antibody Array Assay Kit (KAS02) to ensure optimal performance.

Alternative protein extraction and lysis buffers may be used; however, high concentrations of certain components can interfere with protein biotinylation. Extraction buffers containing Tris and detergents should be limited to no more than 50 mM Tris, 0.1% SDS, 1% Triton X-100, or 1% NP-40. Please contact technical support if you are uncertain about the compatibility of your lysis buffer.

Note: The reagents supplied in the Antibody Array Assay Kit do not contain protease or phosphatase inhibitors. Commonly used protease and/or phosphatase inhibitors (e.g., Roche inhibitor cocktails) may be added to the Extraction Buffer to prevent protein degradation or dephosphorylation.

I. Cells

1. Cell Preparation
   - Adherent Cells: Remove culture medium and wash the cells with ice cold 1X PBS 3 – 5 times. Remove remaining PBS and add 100 – 200 µL of Extraction Buffer to 1 to 5 million cells. Detach cells using a scraper and transfer the cells and remaining supernatant to a clean microcentrifuge tube. Add one tube of Lysis Beads.
   - Suspension Cells: Transfer the culture medium containing cells to a clean tube. Pellet cells by centrifugation at 500 x g for 2 minutes at 4°C. Remove media completely without disrupting the cells. Wash the pellet with ice cold 1X PBS and centrifuge again. Repeat the wash step three times to ensure complete removal of media. Discard any remaining liquid. Add Extraction Buffer and one tube of Lysis Beads to the cells. For 1 – 5 million cells, use 100 - 200 µL of Extraction Buffer.

   Important: Use only PBS for washing the cells. Do not use trypsin or other reagents.

   Note: For primary cells or samples with low protein yield, reduce the volume of Extraction Buffer or increase cell number as needed.
2. Vortex sample rigorously for 30 seconds to 1 minute, then incubate on ice for 10 minutes.
3. Repeat this cycle five times.

4. Centrifuge the mixture at 10,000 x g for 5 minutes at 4°C.
5. Transfer all liquid to a clean tube. Discard the beads.
6. Centrifuge again at 18,000 x g for 15 – 20 minutes at 4°C.
7. Carefully transfer the clear, colorless supernatant to a clean tube.

   Important: The lysate supernatant should appear clear and transparent. If the supernatant is cloudy, centrifuge again at 18,000 x g for 15 to 20 minutes at 4°C. If cloudiness persists, freeze the lysate at −70°C for 10–20 minutes, then centrifuge immediately at 18,000 × g for 15–20 minutes at 4°C. Retain only the top clear layer and discard the remainder.
8. Proceed immediately to Step B (Buffer Exchange/Lysate Purification).

II. Tissues

1. Wash tissues with ice cold 1X PBS with vortexing. Remove and discard PBS. Repeat 3 – 5 times until blood is completely removed.

   Important: Blood contamination will lead to high background on the arrays. Tissues should appear white, and the PBS wash should be clear and colorless once blood is fully removed. Increase the number of washes if needed.
2. Add one tube of Lysis Beads to 10 – 40 mg of tissues.
3. Add Extraction Buffer according to tissue mass:
   - 100 µL for 10–20 mg tissue
   - 200 µL for 20–40 mg tissue
4. Vortex rigorously for 30 seconds to 1 minute. Incubate on ice for 10 minutes. Repeat this step five times.
5. Centrifuge the mixture at 10,000 x g for 5 minutes at 4°C.
6. Transfer all liquid to a clean tube. Discard the beads.
7. Centrifuge again at 18,000 x g for 15-20 minutes at 4°C.
8. Carefully transfer the clear, colorless supernatant to a clean tube.

   Important: The lysate supernatant should appear clear and transparent. If the supernatant is cloudy, centrifuge again at 18,000 x g for 15 to 20 minutes at 4°C. If cloudiness persists, freeze the lysate at −70°C for 10–20 minutes, then centrifuge immediately at 18,000 × g for 15–20 minutes at 4°C. Retain only the top clear layer and discard the remainder.
9. Proceed immediately to Step B (Buffer Exchange/Lysate Purification).

III. Serum/Plasma

1. Centrifuge the serum or plasma sample at 18,000 x g for 10 – 15 minutes at 4°C.
2. Transfer 3–4 µL of the clear, pale-yellow supernatant to a new tube and proceed directly to

Step D (Protein Labeling).

Important: After centrifugation, a thin, milky film may form at the top of serum/plasma sample. This layer must not be transferred to the new tube. Helpful tips: to avoid disturbing the milky film, press and hold the pipette plunger down, insert the pipette tip through the milky film directly into the liquid below, then release the plunger to aspirate only the clear portion. Upon removing the pipette tip from liquid, wipe off the outside of the tip with a Kimwipe before dispensing the sample in a new tube.

B. Buffer Exchange/Lysate Purification

1. Quickly spin the column on a mini-centrifuge to ensure the dry gel settles to the bottom.
2. Remove the screw cap and reconstitute the column by adding 500 µL of Labeling Buffer.
3. Replace the cap and vortex vigorously for approximately 5 seconds. Remove air bubbles by sharply tapping the bottom of the column. Allow the column to hydrate at room temperature for at least 2 – 3 hours prior to use.

   Note: If the column was stored at 4°C, allow the column to warm to room temperature before use.
4. After hydration, loosen the screw cap and then break off column's bottom stopper.
5. Place the column into a wash tube. Spin the column in its wash tube at 1500 x g for 1 minute.
6. Discard excess fluid in the wash tube. Return the column to its wash tube.
7. Add 300 µL of Labeling Buffer to the column. Spin at 1500 x g for 1 minute.
8. Discard the excess fluid and wash tube. Place the column into a collection tube.

   Important: Do not allow the gel bed to dry excessively. Process the sample within a few minutes.
9. Note: the sample volume capacity of each spin column is 100 uL. Carefully dispense up to 100 µL of protein extract directly onto the center of the gel bed without disturbing the gel surface. Avoid touching the sides of the column with the pipette tip, as this may reduce purification efficiency.
10. Spin the column in its collection tube at 1500 x g for 2 minutes.
11. Collect the purified protein from the bottom of the collection tube. Discard the used spin column.
12. Proceed immediately to the next step.

C. Lysate quantification and QC

1. Protein concentration can be measured using UV absorbance at 280 nm (A280), BCA assay, Bradford assay, or other suitable quantification methods.

UV Absorbance A280: Measure lysate's absorbance (OD) using Labeling Buffer or water as the blank.

Note:

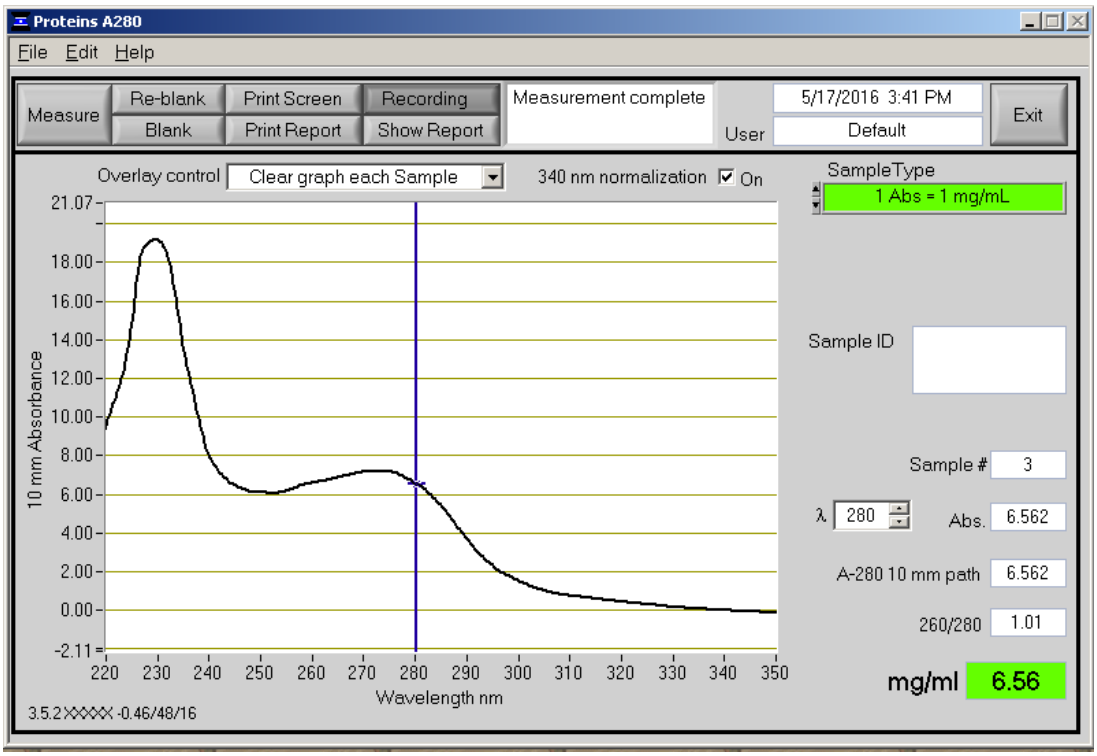
- A target absorbance of 3 OD or higher is recommended. If the OD is below this level, the lysate should be concentrated at 4°C using a vacuum centrifuge (e.g., SpeedVac) or YM-10 filters.
- Only the OD value is needed for subsequent calculations. Conversion to mg/mL is not required, as the common assumption that 1 OD = 1 mg/mL does not apply reliably to lysates; non-protein components such as nucleic acids and insoluble material can interfere with the measurement.

BCA and Bradford assays: both are acceptable alternatives for determining protein concentration. For these assays, the lysate should have a minimum protein concentration of 2 mg/mL.

2. Lysate Quality Control by A280 Assay – Optional, but highly recommended. This step can be performed at the same time when UV absorbance A280 is measured in Step 1 above.

   The quality of lysate sample directly affects assay results. The lysate sample should be as clear and transparent as water. Cloudy or unclear lysate sample will result in low labeling efficiency and high background. A280 spectrum from a UV spectrometer (e.g., Nanodrop) is a good way to determine whether the lysate sample has sufficient clarity.

   When UV A280 is measured, a clear lysate sample produces two well separated peaks at 200-230nm and 240-280nm. If the peaks are not well separated, it indicates the lysate is not sufficiently clear. To improve lysate quality, store the lysate at -70°C for 10 to 20 minutes. Remove from the freezer and immediately centrifuge at 18,000 x g for 15 to 20 minutes at 4°C. Save the top clear layer and discard the rest.



The UV absorbance spectrum shown represents a clear cell lysate. The spectrum displays one peak at approximately 230nm and a second peak near 270nm, with the two peaks well separated. The pattern indicates that the lysate is sufficiently clear and acceptable for use in subsequent steps.

3. Proceed immediately to the next step (Protein Labeling) or store the lysate at -80°C.

D. Protein Labeling – Biotinylation of Protein Samples

1. Biotin Preparation
   a. Briefly centrifuge Biotin Reagent before use.
   b. Add 100 µL of DMF (*N,N*-Dimethylformamide) to 1 mg of Biotin Reagent to prepare a 10 µg/µL biotin stock. Mix thoroughly by vortexing, then quickly spin down. Label this solution as Biotin/DMF.
2. Labeling
   - **Lysates** – Determine the amount of sample needed for the protein/antibody coupling reaction (Step F). For most lysate samples, use 50 – 150 OD of protein per reaction. If BCA or Bradford assay was used to measure protein concentration, use 30 – 100 ug of protein.
     1) Use the lysate concentration obtained from Step C to calculate the required. Example: if 80 OD of lysate sample is needed, and the lysate concentration is 4 OD, the required

volume of lysate is 20 µL (80 ÷ 4 = 20).

2) Aliquot and transfer the amount of lysate sample needed to a new tube. (This aliquot should be no more than 50 uL.)

- **Serum/Plasma** – Continue with the sample prepared in Step A.III.

a. Add Labeling Buffer to the sample to bring the total volume to 75 ul.

b. Add 3 µL of the Biotin/DMF solution to the sample. Incubate the mixture at room temperature for 1–2 hours, vortexing every 10 minutes.

Note: Remaining Biotin/DMF solution may be stored at -20°C for future use.

c. Add 35 µL of Stop Reagent, mix by vortexing, and quickly spin down. Incubate for 30 minutes at room temperature with mixing.

d. Proceed immediately to the next step, or store the biotinylated sample at -80°C for future use.

E. Blocking

Pre-blocking preparation: Remove the antibody arrays from refrigeration but do not open the pouch. Allow the packaged arrays to warm to room temperature for 60 minutes, then remove the pouch and let the slides dry for 30 minutes or longer. Adjust warm-up and drying time based on ambient temperature and humidity.

High humidity locations (relative humidity >50%): warm the packaged arrays in a 30°C oven for 60 minutes, remove the pouch, and continue drying in the oven for an additional 10 minutes.

Important: Before starting Step 1, ensure that the Blocking Solution (See "Reagent Preparation") is at room temperature and that dry milk is fully dissolved.

1. Add 30 mL of Blocking Solution to a 100 x 15 mm Petri dish.
2. Submerge one slide in the solution with the barcode label facing up.
3. Incubate on an orbital shaker rotating at 55 rpm for 30 to 45 minutes at room temperature.
4. Rinse the slide **extensively** with Milli-Q grade or DI water:
   a. Place the slide in a 50-ml conical tube. Fill the tube with 45 mL of water, close the cap, shake by hand for 10 seconds, and discard the water.
   b. Repeat 10 times.

   Important: Complete removal of Blocking Solution is essential for achieving low background. After the final rinse, the thin layer of water on the slide surface should appear uniformly smooth. If the surface looks spotty, repeat the rinse steps.
5. Shake off excessive water on the slide surface. Proceed immediately to the next step.

   Note: Do not allow the slide to dry. If you are not ready to begin the coupling step, store the slide submerged in clean water in a conical tube.

F. Coupling

Note: The Protein Coupling Mix (Step F.1) may be prepared in advance so that coupling can begin immediately after blocking.

Important: Before starting, ensure that the Coupling Solution (See "Reagent Preparation") is at room temperature and that dry milk is fully dissolved.

1. In a new tube, combine 6 mL of Coupling Solution with biotinylated sample from Step D. Vortex briefly to mix. Label the tube as "Protein Coupling Mix."
2. Place the slide in Well 1 (or any clean well) of the Coupling Chamber.
3. Slowly pour all 6 mL of Protein Coupling Mix over the slide, ensuring the slide is fully submerged. Cover the Coupling Chamber with its lid.
4. Incubate on an orbital shaker rotating at 35 rpm for 1 – 2 hours at room temperature.
5. Transfer the slide to a 100x15 mm Petri dish containing 30 mL of 1X Wash Solution (See "Reagent Preparation"). Shake on an orbital shaker at 55 rpm for 10 minutes at room temperature. Discard the wash solution once shaking is completed.
6. Add 30 mL of fresh 1X Wash Solution to the same dish. Wash again by shaking at 55 rpm for 10 minutes at room temperature. Discard the wash solution. Repeat this step once more.
7. Rinse the slide **extensively** with Milli-Q grade or DI water:
   a. Place the slide in a 50-mL conical tube. Fill the tube with 45 mL of water. Close the cap. Shake by hand for 10 seconds. Discard the water.
   b. Repeat 10 times.

   Important: Complete removal of Coupling Solution is essential. After the final rinse, the thin layer of water on the slide surface should appear uniformly smooth. If the surface looks spotty, repeat the rinse steps.
8. Shake off excessive water off the slide surface and proceed to the next step immediately.

   Note: Do not allow the slide to dry. If you are not ready to begin the coupling step, store the slide submerged in clean water in a conical tube.

G. Detection

1. Prepare Cy3-streptavidin detection solution: add 60 µL of Cy3-streptavidin (0.5 mg/mL) to 60 mL of Detection Buffer.

   Note: If the Cy3-streptavidin is in dry form, add biological grade ultrapure water to make 0.5 mg/mL.
2. Pour 30 mL of detection solution into a 100 x 15 mm Petri dish.
3. Submerge the slide in the detection solution. Incubate on an orbital shaker rotating at 35 rpm for 20 minutes at room temperature, protected from light (cover with aluminum foil).

4. Transfer the slide to a new 100 x 15 mm Petri dish containing 30 mL of 1X Wash Solution. Shake at 55 rpm for 10 minutes at room temperature. Discard the wash solution once shaking is completed.
5. Repeat the wash step twice more with fresh 30 mL 1× Wash Solution each time, shaking at 55 rpm for 10 minutes per wash.
6. Rinse the slide extensively with Milli-Q grade or DI water:
   a. Place the slide in a 50-ml conical tube. Fill the tube with 45 mL of water, close the cap, shake by hand for 10 seconds, and discard the water.
   b. Repeat 10 times.

   Important: Complete removal of Detection Solution ensures uniform, low-background signal. After the last rinse, the water layer should appear smooth and uniform; repeat rinsing if spotty.
7. Shake off excess water from the slide.
8. Dry the slide immediately:
   a. Compressed nitrogen or air: use cylinder/nitrogen line (≤40 psi), hold nozzle ~1 inch from slide at 30° angle, and push water off front and back surfaces. Do not use canned air.
   b. Centrifugation: place slide uncapped in a 50-mL conical tube, centrifuge at 1,000 × g for 5 minutes.
9. The slide is now ready for scanning.

If you do not have access to a microarray scanner, you may send the finished slides to our lab for scanning at no charge. For details, visit:
www.fullmoonbio.com/services/array-scanning-image-analysis/

To prepare slides for shipping, place them back into the slide holder and cover the holder with aluminum foil to protect the slides from light. Ship the package at room temperature, and include the array information along with your contact details (name, organization, phone number, and email address).

Shipping address:

Attn: Array Scanning Service
Full Moon BioSystems, Inc.
754 North Pastoria Avenue
Sunnyvale, CA 94085
United States

Phone: 408-737-1702
Email: support@fullmoonbio.com

ARRAY SCANNING

Microarray scanners that are compatible with 76mm x 25mm (3in x 1in) microscope slides may be used to scan Full Moon BioSystems' antibody arrays.

Recommended Scanning Resolution: 10um or higher (10um, 5um, etc.)

Commonly Used Compatible Array Imaging Systems

Manufacturer	Product Name	Required Accessory
Agilent Technologies	DNA Microarray Scanner SureScan Microarray Scanner	
Alpha Innotech	AlphaScan™ Microarray Scanner NovaRay™ Detection Platform	
Applied Precision	arrayWoRx®e Biochip Reader	
Aurora Photonics	PortArray 5000™	
Azure Biosystems	Sapphire and Sapphire Biomolecular Imager	
Biomedical Photometrics	The DNAscope™ HR	
Bio-Rad	VersArray ChipReader™	
GE Healthcare, formerly Amersham Biosciences	Typhoon™ 8610 and newer	Microarray Slide Holder Kit
Genewave	AmpliReader™ 4600 Microarray Reader	
InDevr	Vidia™ Microarray Imaging System	
INNOPSYS	InnoScan® Microarray Scanner	
Miltenyi Imaging GmbH	SensoSpot® Fluorescence Microarray Analyzer	4-slides Holder Adapter
Molecular Devices, formerly Axon Instruments	GenePix® Microarray Scanner All models	
PerkinsElmer, formerly Packard Bioscience	ProScanArray® HT Microarray Scanner ProScanArray® Microarray Scanner ScanArray® GX Microarray Scanner ScanArray® GX PLUS Microarray Scanner	
Tecan	LS Reloaded™ Versatile Scanner PowerScanner™ Microarray scanner	
Vidar Systems	Revolution™ 4200 Microarray Scanner	

Array Scanning Service

Our Antibody Array Scanning Service is available free of charge for customers who purchase our antibody arrays but have no access to a microarray scanner. With this service, you will receive the original raw image files in TIFF format by email. The images may be analyzed with compatible image quantification software.

To use this service, the antibody array assay must be performed using Cy3 or Cy5-streptavidin or equivalent. Please refer to the wavelength table below when choosing alternative dyes.

	Cy3	Cy5
Excitation wavelength	~550 nm	~650 nm
Emission wavelength	~570 nm	~670 nm

This service may be ordered independently or with Image Analysis Service.

IMAGE ANALYSIS

Antibody array images may be analyzed using a variety of image quantification software, including tools provided by scanner manufacturers (e.g., GenePix Pro, Agilent Feature Extraction), third-party software, or open-source programs such as ImageJ.

Consumer-grade picture viewing software (e.g., Microsoft Picture Manager) is not suitable for viewing or analyzing microarray images.

GenePix Array List (GAL) File

A GAL file defines the size and position of blocks, the layout of printed features, and the identifiers associated with each feature. When loaded into compatible analysis software, the GAL file automatically generates grids for spot quantification.

GAL files for each array are available on the Full Moon BioSystems website under the Support section. Parameters in these files are optimized for images scanned at 10 µm resolution in portrait orientation. Images scanned at different resolutions require parameter adjustments, and images acquired in other orientations must be rotated before using the GAL file. A rotation guide for Agilent scanners is available on our website.

If your analysis software does not support GAL files, grid files must be created manually using the array parameters and the array map. Array parameters are available in the User's Guide section of the

website, and array maps can be found under the Documentation tab of each array's webpage. Refer to your software's user guide for instructions on generating grid files.

Image Analysis Service

An optional Array Image Analysis Service is available for users who have obtained array images through the Array Scanning Service but are unable to analyze the data themselves. Results are delivered by email within 2–7 business days. For more information, visit:
https://www.fullmoonbio.com/services/array-image-analysis/

CONTACT SUPPORT

For questions related to antibody arrays, please contact our technical support team:

Phone: 408.737.2875 Email: support@fullmoonbio.com