



# Hydrazide Slide for Glycan Array Applications

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### **Introduction:**

Z Biotech provides a full spectrum of high quality microarray slides for printing glycan microarray. The hydrazide-functionalized substrate slides provide the only microarray slides on the market that are capable for fabricating glycan microarrays with native glycans containing free reducing-end. Additionally, the hydrazide-functionalized slide also can be used for site-specific immobilization of proteins through the carbohydrate portion after  $\text{NaIO}_4$  oxidation. The Hydrazide slides are highly recommended for glycomics, high-throughput glycan microarrays, and all other applications requiring premium, high quality microarray surface. For customer to obtain best microarray results, we also provide validated printing buffers, blocking buffer, and assay buffers for each type of microarray slides.

### **Storage and Handling:**

Store at  $-20\text{ }^\circ\text{C}$  prior to use! Allow package to equilibrate at room temperature (about 20 minutes) before opening. After opening, seal any unused slides in the reusable pouch with desiccant inside and refrigerate.

Avoid contact with the surface of the slides to minimize contamination and abrasion of the surface. Wear gloves and hold slide along with the edges.

### **Buffers Required:**

- Blocking Buffer: HGBB (10109), Hydrazide Glycan Blocking Buffer, 1X concentration (or 1% BSA in PBST (PBS with 0.05% (v/v) Tween-20, pH 7.4))
- Assay Buffer: GAAB (10107), Glycan Array Assay Buffer, 1X concentration
- Wash Buffer: 20 mM Tris-HCl, pH 7.6, 150 mM NaCl, 0.05% Tween 20 (TBST)

### **Blocking:**

1. Affix an assay adaptor onto slide surface;
2. Block the slide for at least 0.5 hour in Blocking Buffer at room temperature with gentle shaking at 80 rpm;

### **Binding Assay or Immunoassay:**

1. Prepare samples (biotinylated or fluorescence-labeled) in GAAB Assay Buffer;
2. Remove the Blocking Buffer by vacuum suction or pipetting the liquid out of each assay chamber;
3. Immediately apply  $\sim 80\ \mu\text{l}$  samples (prepared in step 1) to the appropriate chamber;
4. Cover the chamber windows with an adhesive seal to prevent evaporation during incubation;
5. Incubate the slide on the rotating shaker at 80 rpm for at least 1 hour;
6. After 1 hour incubation, remove the slide from shaker and remove cover;
7. Remove sample from each chamber by vacuum suction or pipetting out;



8. Wash each chamber by adding 100  $\mu$ l Wash Buffer twice (5-minutes incubation for each wash on shaker);

For Biotinylated samples, following procedures below to stain the slides:

9. Add  $\sim$ 80  $\mu$ l of Cy3 or Cy5 labeled streptavidin at 1  $\mu$ g/ml concentration to each assay chamber;
10. Cover the chambers with an adhesive seal;
11. Incubate the slide on the rotating shaker at 80 rpm for at least 30 minutes;
12. Remove the sample from each chamber by vacuum suction or pipetting out;
13. Wash each chamber by adding 100  $\mu$ l Wash Buffer;
14. Remove the 16-chamber adaptor and wash the slides in a Coplin jar of 100 ml Wash Buffer (5-minutes incubation on shaker);
15. Rinse the slide with Millipore water for 2 minutes on shaker;
16. Spin the slide in a slide centrifuge for 15 seconds;
17. Scan the slide with a fluorescent microarray scanner at the appropriate wavelengths and setting;
18. Save the scanning image and process the data with appropriate quantitation software.

For unlabeled antibody samples, following procedures below to stain the slides:

9. Add  $\sim$ 80  $\mu$ l of Cy3 or Cy5 labeled secondary antibody at 1  $\mu$ g/ml concentration to each assay chamber;
10. Cover the chambers with an adhesive seal;
11. Incubate the slide on the rotating shaker at 80 rpm for at least 30 minutes;
12. Remove the sample from each chamber by vacuum suction or pipette;
13. Wash each chamber by adding 100  $\mu$ l Wash Buffer;
14. Remove the 16-chamber adaptor and wash the slides once in a Coplin jar of 100 ml Wash Buffer (5-minutes incubation on shaker);
15. Rinse the slide with Millipore water for 2 minutes;
16. Spin the slide in slide centrifuge for 15 seconds;
17. Scan the slide with a fluorescent microarray scanner at the appropriate wavelengths and setting;
18. Save the scanning image and process the data with appropriate quantitation software.