STREPTAVIDIN COATED SLIDES

Instructions for use

GENERAL INFORMATION

SHC slides are coated with a 1–2 µm thick strepavidin modified nanocomposite hydrogel. They allow convenient immobilization of biotinylated proteins, nucleic acids, carbohydrates and other biomolecules as well as low MW compounds. The strepatvidin-hydrogel conjugate is covalently coupled to an adhesion promoter layer and tolerates non-oxidative aqueous buffers from pH 2–11.

MATERIALS

- SHC Slide.
- Washing Buffer I: Any physiological buffer works. A good choice is Tris, which can also be used as Incubation Buffer.
- Washing Buffer II: Washing buffer I + 0.2 % Tween.
- Washing Buffer III: Washing buffer I + 1 M NaCl pH 8.0.
- Protein Solution: 0,01–0,5 mg/mL in spotting buffer. Sensitive proteins might require addition of small amounts (less then 0.1 %) protective agents such as saccharides to preserve their activity. 0.1–0.5 % Alginate can be added to increase the viscosity of the spotting solutions.
- Spotting Buffer: The choice of the Spotting Buffer depends on the proteins to be spotted. In many cases Tris works well. Generally, the concentration of buffer salts and/or additives should be as low as possible, i.e. 2–10 mM.
- Blocking Buffer: 0.1 mg Biotin/mL Washing Buffer I. Addition of BSA, casein or other blockers is not required but can further increase the S/N ratio.
- Dd water

HANDLING

At least 15 min before opening take the bag out of the freezer and let it reach room temperature. The slides should be handled with clean tweezers; do not touch the surface. Both sides are coated, the spotting area is 60 x 20 mm, so 2.5 mm to the right and left and 7.5 mm to the upper and lower end can be used for handling.

SPOTTING PROTOCOL

SHC slides are ready to spot, i.e. can be used without any pre-treatment. However – equilibration over 10–30 min in Washing Buffer I prior to spotting usually leads to higher immobilization capacities. After this step rinse the slides carefully with dd. water and dry with a quick spin or a sharp jet of clean compressed air/nitrogen. It is essential that no droplets dry on the surface as even dd water leaves trace contaminations behind which might interfere with later processing steps.

1. If not already done, adjust the spotter to the standard slide thickness of 1.00 mm.
2. Place the slides in a spotter and spot the protein solutions. Recommended humidity: > 80 %. The spotting area is 60x 20 mm, so at least 2.5 mm to the right and left and 7.5 mm to the upper and lower end should be left blank.
3. Incubate at > 80 % (the higher the better) humidity over 2–12 hrs.
4. Wash with Blocking Buffer. Agitation or other means to increase the convection during the first 10 min of the blocking process leads to sharper spots and a lower background.
5. Use directly or store dry. If dry storage is required, rinse with dd. water and dry the slides as described above. Some proteins can lose their activity upon drying without protective agents. To avoid such deactivation, XanTec's Stabilization Buffer should be used. Store the spotted and dried slides at ≤ -20 °C under nitrogen in the dark.

LIGAND INCUBATION

Buffer composition and incubation time depend on the nature of the immobilisate and the corresponding ligands. Generally, physiological buffers, such as PBS or Tris and an incubation time of at least 2 h (optimal: 10 h) gave good results. Increasing the convection during ligand incubation can significantly enhance the signal intensity. Optionally, some mg BSA or Casein/mL can be added to the incubation buffer in order to increase the specifity of the array.

To minimize potential ionic interactions between the analyte matrix and slightly negatively charged hydrogel, we recommend washing after the ligand incubation 2x 15 min with Washing Buffer II and another 2x 15 min with Washing Buffer III.

Finally, rinse carefully with dd water and dry as described above.

STORAGE

The slides are packed under nitrogen and can be stored at ≤ -20 °C for at least six months up to one year. Once opened, they should be stored in an inert atmosphere (N_2 or Ar) and used within three months.

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