



INSTRUCTIONS FOR USE

Bead Blocking Solution (Catalog # S-4023)

Note: This protocol and all documents linked below can be downloaded from the appropriate category in the Solulink Library at <http://www.solulink.com/library>.

Bead Blocking Solution contains 5 mg/mL Hammarsten-grade casein dissolved in 25 mM Tris, 150 mM NaCl, pH 7.4 containing Kathon® CG/IP anti-microbial agent and 0.05% anti-foam A.

Storage: Upon receipt store at 4 °C. Product is shipped at ambient temperature. Kathon® is a registered trademark of Rohm & Haas.

Introduction

Bead Blocking Solution is formulated as a NanoLink™ or MagnaLink™ Streptavidin Magnetic bead compatible casein blocking solution containing 5 mg/mL casein in Tris-buffered saline and a microbial preservative. This bead blocking solution is specially filtered to maintain its colloidal stability (no precipitates) when stored at 4°C. The block solution contains no detergents and rapidly blocks non-specific protein binding sites on NanoLink™ or MagnaLink™ Streptavidin Magnetic beads without compromising biotin binding capacity. Bead Blocking Solution in conjunction with ultra-sonication is also used to reduce or eliminate mild bead-to-bead aggregation; leading to a microscopically mono-disperse bead population (Figure 1).

Note: No blocking reagent is optimal for all systems. Bead blocking solution contains phosphorylated casein proteins and is therefore not recommended for use as a blocking agent in phosphorylated protein assays.

General Procedure for Blocking Nonspecific Protein Binding Sites on NanoLink™ and MagnaLink™ Streptavidin Magnetic Beads

1. Add 9 mL of Bead Blocking Solution to 1 mL NanoLink™ or MagnaLink™ Streptavidin Magnetic Beads (10 mg/mL) (9:1 ratio).
2. Vortex vigorously for 1 minute to mix.
3. Incubate the beads for 30-60 minutes at room temperature on a platform or rotary shaker.
4. Place the beads on a strong magnet for 2 minutes and remove the block solution.
5. Wash the beads 3X with a suitable volume of desired wash buffer (with or without Tween-20) before use.

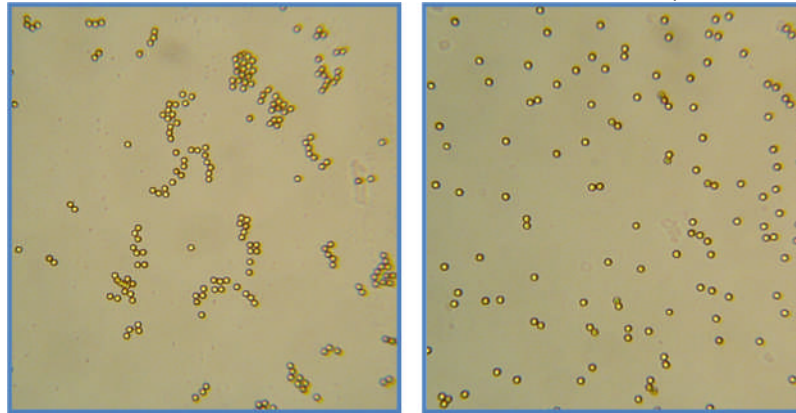
Note: always use a minimum of 9:1 ratio of block solution to bead volume. For example, when blocking 100 µL beads at 10 mg/mL use 900 µL block solution. SDS detergent is not compatible with the use of NanoLink™ or MagnaLink™ Streptavidin Magnetic Beads.

General Procedure for Ultrasonication of NanoLink™ and MagnaLink™ Streptavidin Magnetic Beads using Bead Blocking Solution

Eliminates mild bead-to-bead aggregation and helps maintain mono-dispersity of the bead population

1. Add 9 mL of Bead Blocking Solution to 1 mL of MagnaLink™ or NanoLink™ Streptavidin Magnetic beads (10 mg/mL) (9:1 ratio).
2. Vortex vigorously for 1 minute.

3. Incubate the beads in an ultrasonicator for 5 minutes at room temperature (e.g. Branson 1210 or 1510).
4. Incubate the beads in Bead Blocking Solution for an additional 30-60 minutes at room temperature on a platform or rotary shaker to complete the block step.
5. Place the beads on a strong magnet for 2 minutes and remove the block solution.
6. Wash the beads 3X with a suitable volume of desired wash buffer (with or without Tween-20) before use.



MagnaLink™ (Before)

MagnaLink™ (After)

Figure 1. MagnaLink™ Streptavidin Beads (2.8 μ) before and after sonication (5 minutes) in Bead Blocking Solution (400X magnification). Note the dramatic increase in bead population mono-dispersity.

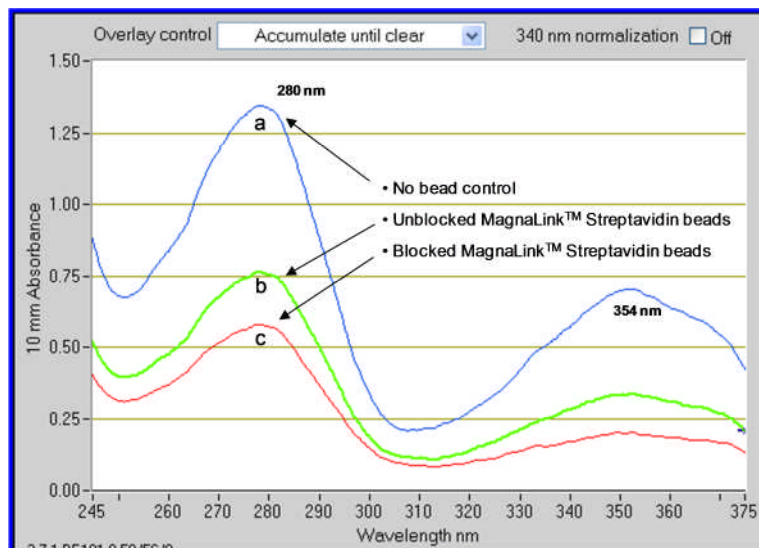


Figure 2. Binding of ChromaLink™ biotin-modified bovine IgG @ 0.9 mg/mL (3.9 biotins/IgG) to either unblocked or blocked MagnaLink™ Streptavidin Magnetic Beads (500 μg bead mass). Superimposed spectra of bead supernatants after a 60 minute incubation **a**) no bead control containing 140 μL ChromaLink™ biotin-modified bovine IgG @ 0.9 mg/mL (125 μg) **b**) unblocked MagnaLink™ Streptavidin bead control supernatant containing 140 μL ChromaLink™ biotin modified bovine IgG @ 0.9 mg/mL (125 μg) **c**) blocked MagnaLink™ Streptavidin Magnetic beads (Bead Blocking Solution) containing 140 μL ChromaLink™ biotin modified bovine IgG @ 0.9 mg/mL (125 μg). The reduction in supernatant A280 and A354 are a direct measure of the mass of biotinylated bovine IgG left unbound in the supernatant. Note that 0.5 mg of blocked MagnaLink™ Streptavidin beads bind more biotinylated IgG than unblocked beads (71 μg vs. 54 μg). No loss of biotin binding capacity on blocked beads indicates that Bead Blocking Solution is free of all endogenous biotin.



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