

## Application Note

### Performing Protein Affinity Purification with SOMAmer® Reagents

#### Introduction

SOMAmer reagents (Slow Off-rate Modified Aptamers) are DNA-based high affinity (average  $K_d < 1$  nM) protein binding reagents with proprietary chemical modifications that provide hydrophobic characteristics not present in natural DNA. These modifications enhance protein binding through direct hydrophobic contacts with the target protein, resulting in increased binding affinities and slower complex dissociation rates compared to unmodified aptamers. This Application Note provides an example of a repeated purification of a target using a SOMAmer reagent column. **Please note that not all SOMAmer reagents have been tested in all applications.** For help or technical support, please submit your questions by either calling the U.S. Technical Support at 1-800-324-0783, emailing [techsupport@somallogic.com](mailto:techsupport@somallogic.com) or by [clicking here](#) to visit the technical support website.

The synthetic nature of SOMAmer reagents enables their immobilization onto a solid support through a reactive group at the 5' end of the sequence (e.g. biotin, amine, or thiol). Additionally, the stability of single-stranded DNA enables easy regeneration and reuse of SOMAmer reagent columns. Specific elution conditions should be devised to release the purified protein under conditions that are compatible with the downstream use of the target protein.

Inclusion of a competitor during or after SOMAmer incubation can be valuable in distinguishing between specific and non-specific binding in all SOMAmer reagent applications. While many applications do not involve high levels of proteins with high random DNA affinity, they often include very high concentrations of proteins with modest affinity (e.g., some highly abundant plasma proteins that can bind DNA). Addition of a competitor can help reduce artifacts that may arise from random DNA binding layered on specific SOMAmer reagent binding. The SomaLogic® Polyanionic Competitor [Product Number 910-00001] or dextran sulfate at concentrations ranging from 0.1  $\mu$ M to 10 mM should be evaluated for use in specific applications.

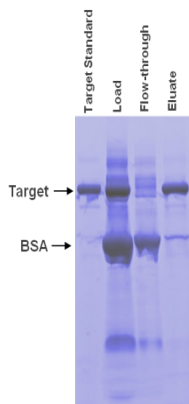
The example below describes a SOMAmer reagent with a 5' amine coupled to an epoxy resin. A similar procedure can also be performed using a SOMAmer reagent with a 5' biotin coupled to streptavidin resin.

#### Recommended Materials

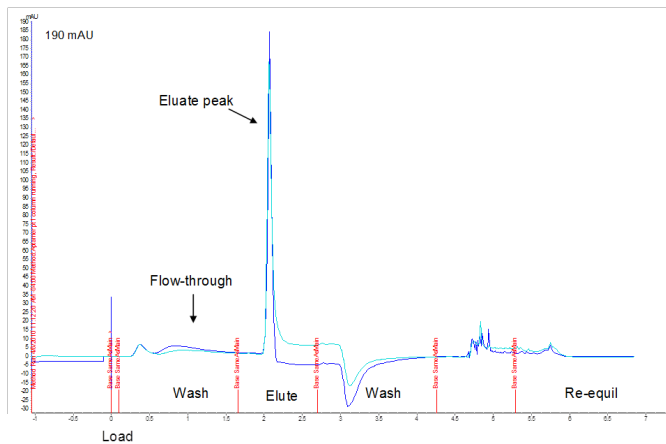
- 5' amine- labeled SOMAmer reagent
- Protein Sample
- Epoxy resin
- SBT: 120 mM NaCl, 5 mM KCl, 5 mM MgCl<sub>2</sub>, 40 mM HEPES pH 7.5, 0.05% Tween® 20 detergent
- 80 mM methanolamine
- 50 mM NaOH

#### Purification Protocol

1. Couple a SOMAmer reagent to a porous epoxy resin via the 5' amine according to manufacturer's protocols
2. Load sample onto the column and wash with SBT
3. Elute the target protein with 80 mM methanolamine
4. Wash the column with 50 mM NaOH to remove any residual protein material
5. Wash the column with SBT to regenerate



**Figure 1: SDS-PAGE demonstrating target recovery.** In this example a cell culture harvest over-expressing the target was spiked with BSA. The eluate following SOMAmer reagent affinity chromatography shows significant enrichment with minimal carryover of BSA or other cell culture impurities.



**Figure 2: Chromatogram demonstrating column reusability.** In this example the column was reused five times with no significant change in performance. The first chromatography run is shown in blue and the 5th run is shown in cyan. There is no significant loss in capacity over repeated uses.

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