

Product Information

ExtrAvidin®-Magnetic beads, high binding

For purification of biotinylated macromolecules and complexes

Product Number **E2642**

Storage Temperature: 2-8 °C

Product Description

ExtrAvidin® is a tetrameric protein containing four biotin binding sites. The avidin-biotin high affinity interaction ($K_d = 10^{-15}$ M) is considered one of the strongest non-covalent interactions known in nature.¹⁻³ The use of the avidin-biotin complex for affinity purification was described in 1970. It has since been used in numerous studies and biotechnological applications.¹⁻³ This was achieved because of the ability to couple chemically biotin with different binders, without disturbing its function or structure. This allows unique interaction with a variety of avidin carriers, including protein or DNA molecules, avidin protein bound to a solid surface matrix, reporter molecules, probes or carriers.

Avidin-biotin interaction-based applications include purification, enrichment, detection, amplification and other research medical and industrial processes.¹⁻⁷ In addition, the ExtrAvidin® high specificity binding to biotin, together with the low background staining, grants it a significant advantage compared to non-modified avidin or streptavidin produced by *Streptomyces avidinii*.

ExtrAvidin®-Magnetic beads are a unique, de-glycosylated, avidin-modified form of an affinity-purified egg white avidin. The ExtrAvidin® is conjugated to cyanogen bromide-activated magnetic beads at a ratio of ~4 mg/mL protein-to-beads.

ExtrAvidin®-Magnetic beads provide high affinity with high specificity binding to biotin (also known as vitamin B7), including biotinylated proteins and biotin-tagged fusion proteins. The product may be used in various immunological techniques, including immunoprecipitation and immunoaffinity purification.

Reagent

ExtrAvidin®-Magnetic beads are provided as a suspension containing 50% beads, in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

For continuous use and extended storage, store at 2 °C to 8 °C. **Do not freeze.**

Product Profile

Binding capacity: at least 650 nmoles of biotin per 1 mL of magnetic beads-settled resin.

Recommended Procedure for Purification of Biotinylated Proteins or Biotinylated Antibodies

Pre-washing ExtrAvidin®-Magnetic beads: To ensure homogeneity, mix the beads thoroughly before use by repeated inversion, gentle vortexing or using a rotating platform.

1. Add 200 µL of ExtrAvidin®-Magnetic beads into a 1.5 mL microcentrifuge tube.
2. Place the tube into a magnetic stand to collect the beads against the side of the tube.
3. Remove and discard the supernatant. Wash with 200 µL PBS. Repeat the washing 3-5 times.
4. Apply the sample containing the biotinylated protein/antibody at ~750 µg/test. Incubate for 30 min at RT using a rotating platform.
5. Wash with PBS until the absorbance at 280 nm is minimal. Save flow-through for calculating the binding capacity.
6. Elution of biotinylated proteins:
 - Boiling for 5 min with 1× SDS sample buffer, followed by SDS-PAGE analysis, or:
 - Incubation with 2 M acetic acid or 2 M Glycine HCL pH 2.2. If required, immediately neutralize eluted samples.

Note: In order to obtain the best results in different techniques and preparations, we recommend determining the optimal working concentration by a titration test.

Binding capacity and elution capacity may vary, depending on the characteristics of the Biotin-tagged fusion proteins. For optimal results, it is recommended to try different elution buffers.

References

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4. O'Connor, E. *et al.*, *J Immunol Methods.*, **229(1-2)**, 155-160 (1999).
5. Gao, H. *et al.*, *Proc. Natl. Acad. Sci. USA*, **105(51)**, 20146-20151 (2008).
6. Iikura, M. *et al.*, *J. Leukoc. Biol.*, **70(1)**, 113-120 (2001).
7. Kellenberger, L.D., and Petrik, J., *Gynecol. Oncol.*, **149(2)**, 361-370 (2018).

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SS,BW,NB,GCY-01/21-1