

Anti-HGFR aptamer, Direct Magnetic AP Kit

Introduction

When a protein is expressed at low levels and is difficult to detect with western blot analysis, aptoprecipitation (AP, Aptamer based protein pull down method) may be the method of choice. An aptoprecipitating reagent has to be specific in order to avoid precipitation of unwanted protein. Furthermore, sufficient affinity is required to pull down the protein and it has to withstand stringent washing steps. AptSci HGFR aptamer molecule is a specific affinity ligand and has been proven well suited for pull down experiments of HGFR proteins(c-Met). Most commonly encountered problems with IP approach is interference from antibody heavy and light chains that may co-migrate with relevant bands, masking important results. However aptamer as an oligonucleotide will not contribute to protein/peptide background that can interfere with subsequent analysis.

AptSci has developed proprietary protein pull down method using target protein-specific aptamers. The aptamer-coupled magnetic bead included in the kit has low nonspecific binding characteristic and enables convenient magnetic isolation of protein targets and reusable magnetic beads. Mild elution condition enables isolation of non-denatured proteins which can be used for further study.

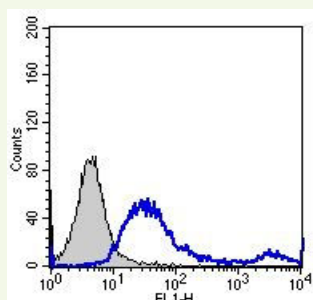


Fig. 1. Flow cytometry histograms showing the binding of representative HGFR aptamer against the target A549 cells. Approximately 1×10^6 cells were washed and incubated with FITC-conjugated HGFR aptamer (Blue histogram). The untreated cell was used as background fluorescence signal (Gray histogram).

Result of Aptoprecipitation (AP)

As shown in Figure 2, Western blot analysis revealed that HGFR aptamer precipitated HGFR with high specificity, while no HGFR was observed when precipitating with blank beads. This result indicates that HGFR aptamer is highly specific to HGFR protein and HGFR aptamer-coupled magnetic bead efficiently precipitates HGFR from a protein complex.

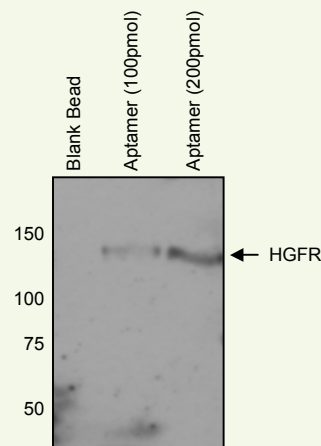


Fig. 2. Aptoprecipitation of C-Met protein from A549 cells using the AptSci direct HGFR AP Kit. A549 cell lysates (1mg/lane) were incubated with HGFR aptamer-coupled magnetic bead. After washing the beads, the bound protein was eluted with boiling SDS loading buffer. The eluate was loaded on a SDS-PAGE (4-15% gradient gel) and blotted onto a PVDF membrane. Western blot was probed with anti HGFR Ab.

Product Information

- **Product name:** Anti-HGFR aptamer, Direct Magnetic AP Kit
- **Catalog number:** HGFR-2308DM
- **Content:** Magnetic agarose conjugated HGFR aptamer molecule and all buffers required to perform small scale AP
- **Form:** As 25% slurry in 20% ethanol containing 0.04% (w/v) sodium azide.
- **Protein source for generation of aptamer:** Recombinant protein produced in mammalian cells
- **Specificity:** Anti-HGFR aptamer binds to human HGFR. Cross reactivity with other species has not been tested.
- **MW:** ~17 kDa
- **Conjugation yield:** > 90% as determined by spectrometer analysis.
- **Tested applications:** FACS and Aptoprecipitation.
- **Storage:** At +4°C.
- **Shipping:** At ambient temperature.
- **Stability:** There is no decrease in performance of the kit after storage for 6 months at ambient temperature.



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LIMITATIONS

Warranty: AptSci AptoPrep™ products are warranted to meet stated product specifications and to conform to label descriptions when used and stored properly. Unless otherwise stated, this warranty is limited to one year from date of sales for products used, handled and stored according to AptSci's instructions. AptSci's sole liability is limited to replacement of the product or refund of the purchase price. AptoPrep™ products are supplied for research use only. They are not intended for medicinal, diagnostic or therapeutic use. AptoPrep™ products may not be resold, modified for resale or used to manufacture commercial products without prior written approval from AptSci.