Anti-Akt2 aptamer, Indirect 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 Akt2 aptamer molecule is a specific affinity ligand and has been proven well suited for pull down experiments of Akt2 proteins. 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 biotinylated aptamer has low nonspecific binding characteristic and streptavidin magnetic beads enable convenient magnetic isolation of protein targets. Mild elution condition enables isolation of non-denatured proteins which can be used for further study.

Result of Aptoprecipitation (AP)

Figure 1 shows that the Akt2 proteins were precipitated from MCF7 cell extract using the biotinylated Akt2 aptamer. An intense Akt2 band was clearly obtained by using the biotinylated Akt2 aptamer, while there was no Akt2 detected when precipitating with blank beads.

In summary, the biotinylated Akt2 aptamer molecule is highly specific to Akt2 protein and Akt2 aptamer efficiently precipitates Akt2 from a protein complex.

Fig. 1. Aptoprecipitation of Akt2 protein from MCF7 cells using the AptSci Indirect Akt2 AP Kit. Cell extract was prepared from MCF7 cells. Cell lysates (1mg/lane) were incubated with biotinylated Akt2 aptamers (20 pmol). The mixed solution was further incubated with streptavidin magnetic beads. After washing the beads, the bound protein was eluted with 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 Akt2 Ab.

Product Information

- **Product name**: Anti-Akt2 aptamer, Dual Magnetic AP Kit
- **Catalog number**: Akt2-2154IM
- **Content**: Magnetic agarose conjugated Akt2 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-Akt2 aptamer binds to human Akt2. Cross reactivity with other species has not been tested.
- **MW**: ~18 kDa
- **Conjugation yield**: > 90% as determined by spectrometer analysis.
- **Tested applications**: 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.

www.aptsci.com. Aptamer Sciences Inc.
Postech Biotech Center, San31, Hyoja-Dong, Pohang, Gyeongbuk 790-784, South Korea. TEL +82-54-279-8691
FAX +82-54-279-8245. E-mail: aptamer@aptsci.com.