

## Anti-EGFR aptamer, Magnetic Cell Isolation Kit (Catalog No. EGFR-2369BCI/ EGFR-2369FBCI)

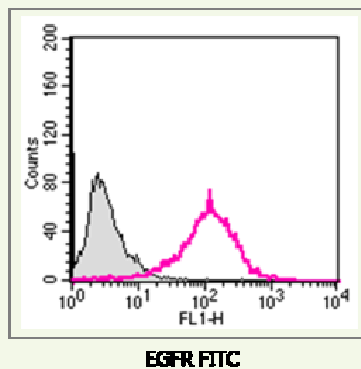
### Description

AptSci provides two types of kits. One is biotinylated aptamer based cell isolation kit (Catalog No. EGFR-2369BCI). The other is bi-labeled (FITC dye conjugated biotin aptamer) aptamer based cell isolation kit (Catalog No. EGFR-2369FBCI). Aptamer based magnetic cell isolation kit products do not adversely affect cells during isolation process, thus can be used to isolate pure, viable and functional cells which advance your biology research.

AptSci EGFR cell isolation kit is ideal for positive isolation of EGFR expressing target cells directly from all types of samples. Cell can also be eluted from bead-cell complexes with releasing buffer included in Kit, and then be used in all downstream experiments, including flow cytometry, cell culture and molecular studies.

### Component description

- AptSci aptamer is a single stranded oligonucleotide that is engineered through advanced SELEX with modified nucleotide.
- Aptamer is generated with recombinant human EGFR protein produced in mammalian cells and binds their cellular target with high affinity and specificity (Fig. 1).
- Magnetic beads are uniform, colloiddally stable and non-porous beads (1µm diameter) covalently coupled with streptavidin.
- Bi-labeled aptamer that has FITC at 5'-end to monitor protein expression by flow cytometry and biotin at 3'-end to separate target cells.



**Fig. 1.** Flow cytometry histograms showing the binding of representative EGFR aptamer in A431 cells.

Approximately  $1 \times 10^6$  cells were incubated with biotinylated EGFR aptamer and stained 2<sup>nd</sup> FITC-streptavidin (Pink histogram). The control cells were stained 2<sup>nd</sup> FITC-streptavidin as background (Gray histogram).

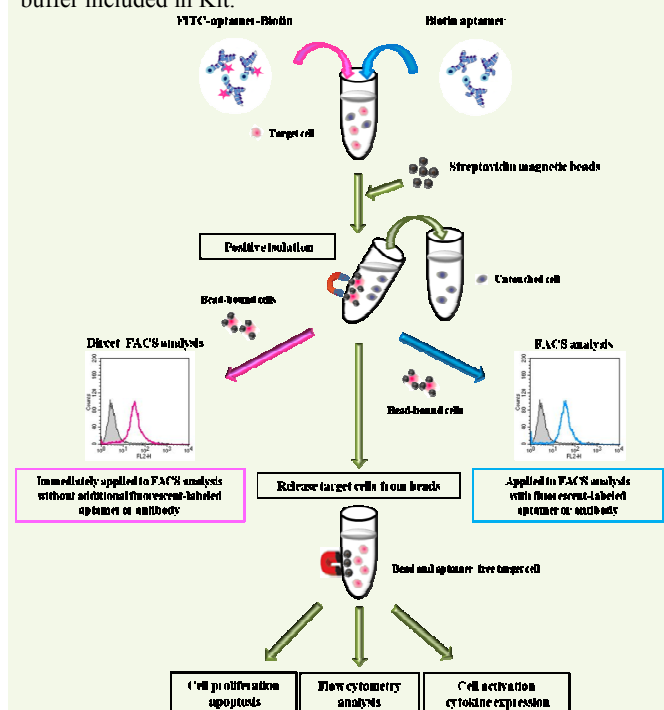
### Principle of the AptoPrep™ Cell Isolation

AptSci cell isolation kit is designed to isolate cells via a indirect method and for positive selection principle using biotinylated aptamers and streptavidin magnetic beads.

Target cells are specifically labeled with biotinylated aptamer against cell surface target of desired cells. Streptavidin magnetic beads allow for efficient binding to the aptamer labeled cell. Magnetically labeled target cells are then separated from unlabeled cells using magnet. FACS analysis can immediately be performed with bi-labeled aptamer during cell isolation process (Catalog No. EGFR-2369FBCI). In final step, bead-free and aptamer-free target cells were released from bead-bound cells (positive fraction) using releasing component (Fig. 2).

*Positive isolation:* Discard the supernatants and use the bead-bound cells for downstream application.

*Release target cells from beads:* Bead-bound cells are washed and target cells are released from the beads with releasing buffer included in Kit.

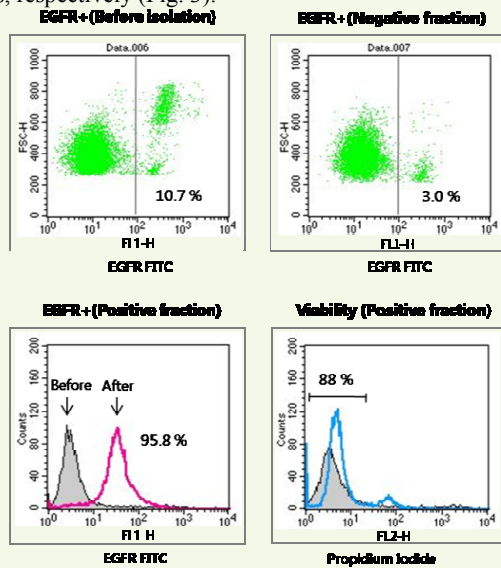


**Fig. 2.** Overview of AptoPrep cell isolation procedure.

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### Typical results of the AptoPrep™ EGFR Cell Isolation

Isolation of EGFR<sup>+</sup> cells from lymphocytes was performed with AptSci EGFR cell isolation kit. Human epidermoid carcinoma A431 cells (EGFR over-expressed cell,  $\sim 1 \times 10^6$  cells) were spiked with lymphocytes ( $1 \times 10^7$  cells). Yield of EGFR<sup>+</sup> cells isolation was measured at 71.9 %. Purity and viability of recovered EGFR positive cells were measured at 95.8% and 88%, respectively (Fig. 3).



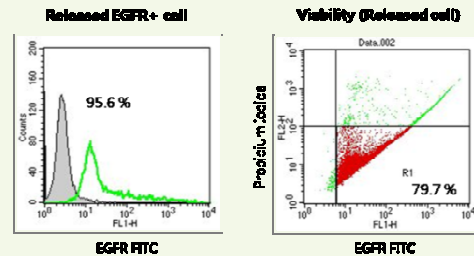
**Fig. 3. Isolation of EGFR<sup>+</sup> cells from lymphocytes with AptSci EGFR cell isolation kit.**

Both start sample before isolation and negative fraction after isolation were stained with FITC-human EGFR aptamer (Green dot plot). Positive fraction is stained with FITC-human EGFR aptamer (Pink histogram) and propidium iodide for cell viability (Blue histogram). The control cells were stained with FITC conjugated control aptamer.

### Downstream application

EGFR<sup>+</sup> cells can be efficiently isolated from a sample with AptSci EGFR cell isolation kit. Lyse the cells directly after isolation, and isolate proteins, DNA, or mRNA to be used in PCR, microarrays, proteomics, and other applications where the removal of beads is not required. For functional studies such as cytokine expression, proliferation/apoptosis induction or for flow cytometry analysis, the cells need to be released from beads after positive isolation of cell. Releasing buffer included in Kit will allow you to collect the bead-free and aptamer-free EGFR<sup>+</sup> cell.

After elution of EGFR<sup>+</sup> cells with releasing buffer, elution yield was calculated by counting cells with hemocytometer and measured at 51% (data not shown). Purity and viability of released EGFR<sup>+</sup> cells were measured at 95.6% and 79.7%, respectively (Fig. 4).



**Fig. 4. Flow cytometry showing bead-free and aptamer-free EGFR<sup>+</sup> cells are released with releasing buffer.**

Released cell fraction (Green histogram and Green dot plot) was stained with human FITC-EGFR aptamer and propidium iodide for cell viability (Red dot plot). The control cells were stained with FITC conjugated control aptamer (Gray histogram).

### Product Information

- **Product name:** Anti-EGFR aptamer, Magnetic Cell Isolation Kit
- **Catalog number:** EGFR-2369BCI (biotinylated aptamer based cell isolation kit), EGFR-2369FBCI (bi-labeled aptamer based cell isolation kit)
- **Content:** EGFR-2369BCI (Biotinylated aptamer, streptavidin-coated magnetic bead and buffer), EGFR-2369FBCI (FITC dye conjugated biotin aptamer, streptavidin-coated magnetic bead and buffer)
- **Form:** Dried aptamer and bead in 50 mM Tris pH8.0, 150 mM NaCl, 0.05% NaN<sub>3</sub>.
- **Protein source for generation of aptamer:** Recombinant protein produced in mammalian cells.
- **Specificity:** Anti-EGFR aptamer binds to human EGFR. Cross reactivity with other species has not been tested.
- **MW:**  $\sim 18$  kDa
- **Tested applications:** FACS and cell isolation
- **Shipping & Storage :** At 2°C to 8°C. There is no decrease in performance of the kit after storage for 1 year at 2°C to 8°C.



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### LIMITATIONS

Warranty: AptSci AptoPrep™ products are warranted to meet stated product specifications and to confirm 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™ may not be resold, modified for resale or used to manufacture commercial products without prior written approval from AptSci.