

Surface Activated Dynabeads® for Cell Separation

- for the innovative scientist

- Use your own specific antibody or ligand
- Isolate specific cells from heterogenous samples such as whole blood
- All in one tube - no columns or centrifugations needed
- Get high yield, purity and viability of your specific cell subset

Introduction

Dynabeads® M-450 Epoxy and Dynabeads® M-450 Tosylactivated offer total flexibility in cell separation and have been used by many scientists working with unconventional cell types.

These Dynabeads® have no antibody attached, but their activated surface can easily be coated with most available antibodies or other proteins (e.g. lectins) and subsequently used for specific cell isolations.

Surface activated Dynabeads® are ideal for making your own cell separation Dynabeads® if no suitable primary or secondary coated product is available, or if you want to add more than one antibody to the same bead.

Advantages of Surface Activated Dynabeads®

- Bind any antibody directly, regardless of species and isotype
- IgM antibodies are particularly suited for use with these Dynabeads®
- Bind two or more antibodies to the same bead
- Bind your own secondary antibody (e.g. anti-monkey or anti-hamster)
- Bind other ligands (e.g. ScFv, lectins, Annexin V) to the Dynabeads®

Cell Separation Techniques

Isolate specific cells directly by coupling the primary antibody onto the surface activated Dynabeads® (Fig. 1). Alternatively, couple your secondary antibody onto the surface activated Dynabeads® and choose between a direct or indirect cell separation technique (Fig. 2). In the indirect technique, add the primary antibody to the sample and allow to react with antigen on the target cells. Add the secondary coated Dynabeads® to the labelled cells. In the direct technique, couple the primary antibody onto the secondary coated Dynabeads® before cell isolation.

Samples

Any cell source may be used, even viscous samples such as whole blood, buffy coat, bone marrow or digested tissue samples. After a short incubation, the Dynabeads® will bind to the target cells and the cell:bead complexes can be easily separated from the crude sample in under 2 minutes with a magnet.

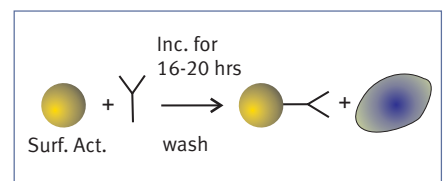


Fig. 1. Coat primary antibody to activated Dynabeads®. The beads are now ready for cell separation.

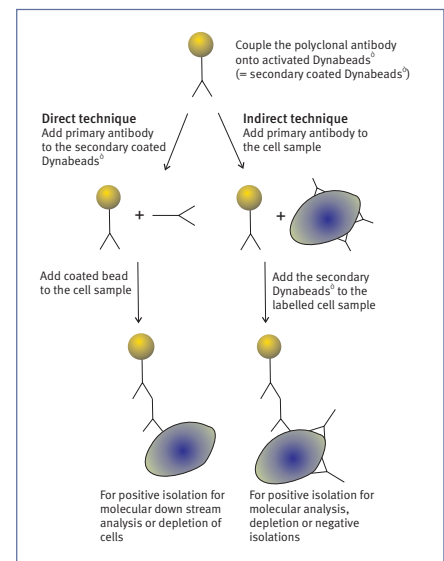


Fig. 2. To make a "universal" secondary bead for cell isolation, coat a polyclonal antibody onto the activated Dynabeads®. The direct technique is suitable for making a stock of beads, while the indirect technique is perfect for making your own negative isolation kits with an antibody cocktail towards the unwanted cell types.

Dynabeads® M-450 Epoxy – properties:

These beads are hydrophobic and covered with **surface epoxy (glycidyl ether) groups**. These groups react covalently with primary amino- and sulphhydryl groups. No further activation is necessary and irreversible binding of antibodies to the epoxy surface is achieved over a wide temperature range and at a neutral to high pH.

Special features: Dynabeads® M-450 Epoxy will efficiently bind proteins at low temperatures, enabling immobilisation of temperature-labile proteins. A range of different antibodies and methodologies have been investigated with these beads and an efficient selection of cells has been shown. The ability to bind two or more antibodies in a fully controlled procedure is demonstrated with the Dynabeads® CD3/CD28 T Cell Expander product.

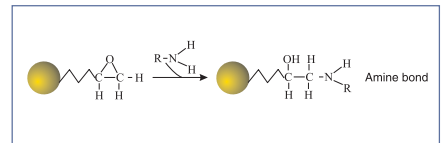
Clinical Research: For *ex vivo* cell separations or modifications as part of clinical trials, a sterile version of the product is available. Dynabeads® *ClinExVivo*™ Epoxy is manufactured under validated aseptic conditions with full quality control and tested for sterility and endotoxins, according to United States Pharmacopeia. It holds a master file from FDA (CBER) and is **CE 0434** marked.

Dynabeads® M-450 Tosylactivated – properties:

Dynabeads® M-450 Tosylactivated are hydrophobic beads covered with **surface tosyl groups**. These groups react covalently with primary amino- and sulphhydryl groups predominantly in the Fc-region of antibodies, ensuring optimal orientation. No further activation is necessary, and covalent binding of antibodies to the surface is achieved at 37°C and a neutral to high pH.

Which product should I choose?

Both Dynabeads® M-450 Epoxy and Dynabeads® M-450 Tosylactivated will covalently bind antibodies or proteins, but the Epoxy product offers the benefit of coating over a wider temperature range. This product is also available in a sterile grade for clinical research.



Dynabeads® M-450 Epoxy

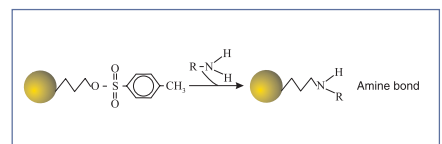
Methodology

1. Wash the Surface Activated Dynabeads®

NOTE: Do not use buffers containing proteins or amino components (Glycine, Tris etc.) for pre-washing or coating.

2. Add PURE antibody solution.
3. Mix and put the tube on a rotator.
4. After 10-30 min, add blocking reagent.
5. Incubate for 16-20 h at 4-37°C, do not let the beads settle.
6. Wash the coated beads 3 times at 2-8°C, using the magnet.
7. Store the coated beads until ready for use.

See package inserts for more details



Dynabeads® M-450 Tosylactivated

References:

1. Takemoto M. *et al.* (2002) A new method for large scale isolation of kidney glomeruli from mice. *Am J Pathol* 161:799-805
2. Wang YS *et al.* (2002) Inhibitory effects of triamcinolone acetonide on bFGF-induced migration and tube formation in choroidal micro-vascular endothelial cells. *Graefes Arch Clin Exp Ophthalmol* 240:42-48
3. Kasimir-Bauer S *et al.* (2002) A summary of two clinical studies on tumor cell dissemination in primary and metastatic breast cancer: Methods, prognostic significance and implication for alternative treatment protocols (Review). *Int J Oncol* 20:1027-34

Ordering information

Product	Volume Range Per Unit	Product No.
Dynabeads® M-450 Epoxy	5 ml	140.11
Dynabeads® <i>ClinExVivo</i> ™ Epoxy	10 ml	402.01
Dynabeads® M-450 Tosylactivated	5 ml	140.13
Dynal MPC®-S	20 µl / 5 ml	120.20
Dynal MPC®-L	1-15 ml	120.21
Dynal MPC®-50	15-50 ml	120.24

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