



Miltenyi Biotec

Antigen targeting and analysis

Novel tools for antigen targeting and effective cross-priming

Antigen targeting and analysis

Introduction

Antigen targeting to antigen-presenting cells (APCs) via specific receptors, such as CD205 (DEC-205), DCIR2 (33D1), or Dectin-2 has been used to induce effective antigen-specific T cell responses. A reliable orchestration of T_H1 and T_H2 cell responses promises the establishment of efficient tumor vaccination protocols since both cell types are required for tumor defense. Ovalbumin is widely used as a model antigen for the characterization of antigen uptake, processing, and presentation. Especially the induction of $CD8^+$ T cell responses after targeting antigen via antigen uptake receptors to dendritic cells (DCs), commonly termed cross-priming, has raised major interest.¹⁻⁵

Miltenyi Biotec now offers the Ova Antigen Delivery Reagent and the Ova Antigen Delivery Module Set to enable antigen targeting to any desired antigen uptake receptor on APCs or a fast and flexible functional investigation of new receptors on APCs.

Features at a glance:

- Efficient targeting of ovalbumin to APCs via antigen uptake receptors
- Flexible: any desired antigen uptake receptor
- Reproducible results
- Easy-to-standardize procedure
- Includes comprehensive experimental protocol

Ova Antigen Delivery Reagent—the principle

APCs, such as DCs, can be targeted via, for example, CD205 (DEC-205). For effective antigen targeting, mouse $CD11c^+$ DCs are first labeled using a CD205 (DEC-205)–Biotin antibody. Subsequently, the cells are loaded with the Ova Antigen Delivery Reagent—a standardized, lot-to-lot-consistent Anti-Biotin antibody conjugated to ovalbumin and FITC (fig. 1). After *in vitro* maturation DCs effectively present ovalbumin peptides on MHC class I and MHC class II molecules. The ovalbumin antigen presentation can be detected by proliferation of ovalbumin-specific transgenic T cells derived from OT-I, OT-II, or DO11.10 mouse strains.

Product	Order no.
Ova Antigen Delivery Reagent	130-094-663

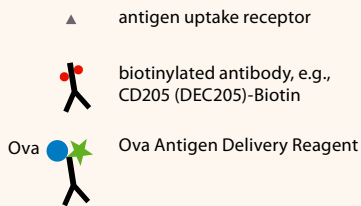
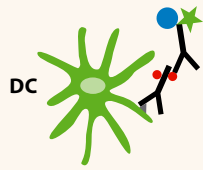


Figure 1: Principle of antigen targeting using the Ova Antigen Delivery Reagent

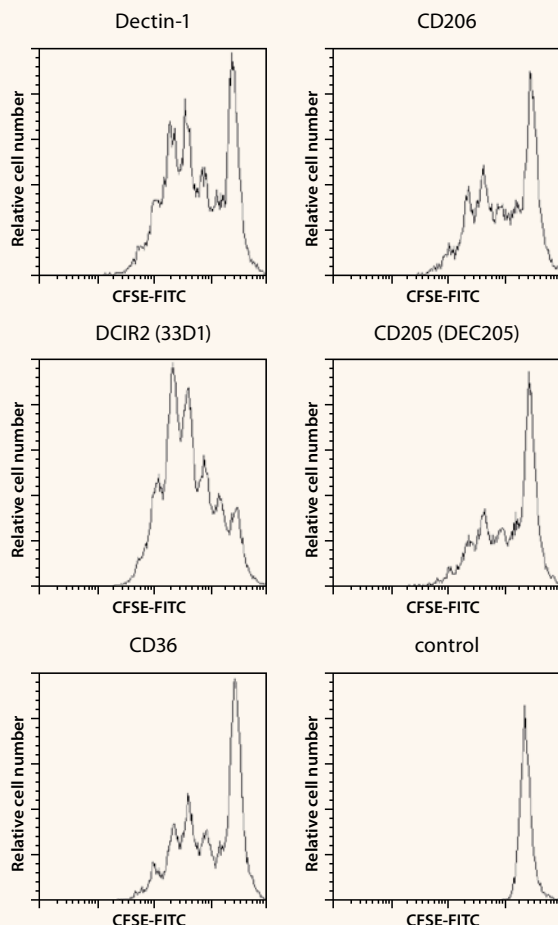


Figure 2: Proliferation of CD4⁺ T cells detected by CFSE labeling. CD4⁺ T cells from DO11.10 mice were cultured with DCs that were previously isolated using CD11c MicroBeads. Ovalbumin was targeted to DCs via Dectin-1, CD206, DCIR2 (33D1), CD205 (DEC205), or CD36, using biotinylated antibodies against the respective antigen uptake receptors in combination with the Ova Antigen Delivery Reagent. Cells were cultured in the presence of Pam3CysSK4 that induces DC maturation via toll-like receptors (TLR). In the absence of Ova Antigen Delivery Reagent (bottom right) no T cell proliferation was detected.

Ova Antigen Delivery Module Set: the complete solution for the effective delivery of ovalbumin to APCs

The Ova Antigen Delivery Module Set comprises all the reagents that are required for the isolation of DCs, antigen delivery, and subsequent analysis of antigen presentation. The CD205 (DEC205)–Biotin antibody that is included in this set allows antigen delivery via CD205 (DEC205). However, the Ova Antigen Delivery Module Set is also designed for antigen delivery via the uptake receptor of your choice, provided an appropriate antibody is available; the antibody can easily be biotinylated by using the One-Step Antibody Biotinylation Kit. The biotinylated antibody can then be used in combination with the Ova Antigen Delivery Reagent for effective antigen delivery. An Anti-MHC Class II antibody for the analysis of maturation is included in the set. It is designed for the comparison of antigen uptake by antigen-presenting cells via various receptors^{1–6}.

References:

1. Bonifaz *et al.* (2002) *J. Exp. Med.* 196: 1627–1638.
2. Dudziak *et al.* (2007) *Science* 315: 107–111.
3. Mouriès *et al.* (2008) *Blood* 112: 3713–3722.
4. Caminschi *et al.* (2008) *Blood* 112: 3264–3273.
5. Sancho *et al.* (2008) *J. Clin. Invest.* 118: 2098–2110.
6. Caminschi *et al.* (2009) *Eur. J. Immunol.* 39: 931–938.

Product	Order no.
Ova Antigen Delivery Module Set	130-094-831

Ova Antigen Delivery Module Set

Component	Benefit
CD11c MicroBeads	Isolation of APCs, i.e., DCs
CD205 (DEC205)–Biotin, antibody	Biotin-labeling of antigen uptake receptor
Ova Antigen Delivery Reagent	Delivery of antigen via labeled antigen uptake receptor
Anti-MHC Class II-PE, antibody	Labeling of APCs for analysis

Related products

Product	Order no.
Anti-DCIR2 (33D1), mouse	coming soon
Anti-Ly-6C-APC, mouse	130-093-136
Anti-mPDCA-1-APC, mouse	130-091-963
CD205 (DEC205)-APC, mouse	130-092-285
CD4 (L3T4) MicroBeads, mouse	130-049-201
CD4 ⁺ T Cell Isolation Kit, mouse	130-090-860
CD8a (Ly-2) MicroBeads, mouse	130-049-401
Pan T Cell Isolation Kit, mouse	130-090-861
One-Step Antibody Biotinylation Kit	130-093-385



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