

MOLECULAR PROBES®

PRODUCT INSERT

HAMSTER anti-MOUSE CD3

| Product Code | Form | Volume | Antibody* | Excitation (nm) | Peak Emission (nm) | Matching Isotype Controls | |
|--------------|-------------------|--------|-----------|-----------------|--------------------------|------------------------------|-----------|
| HM3400 | Purified | 1.0 ml | 200 μg | N/A | N/A | Hamster IgG Purified | Code HM00 |
| HM3400-5 | Purified | 2.5 ml | 500 μg | | | | |
| HM3415 | Biotin | 1.0 ml | 100 μg | N/A | N/A | Hamster IgG Biotin | Code HM15 |
| HM3415-3 | Biotin | 3.0 ml | 300 μg | | | | |
| HM3428 | Pacific Blue® | 1.0 ml | 100 μg | 405 | 455 | Hamster IgG Pacific Blue® | Code HM28 |
| HM3420 | Alexa Fluor®† 488 | 1.0 ml | 100 μg | 488 | 519 | Hamster IgG Alexa Fluor® 488 | Code HM20 |
| HM3401 | FITC | 1.0 ml | 100 μg | 488 | 525 | Hamster IgG FITC | Code HM01 |
| HM3401-3 | FITC | 3.0 ml | 300 μg | | | | |
| HM3401-5 | FITC | 5.0 ml | 500 μg | | | | |
| HM3421 | Alexa Fluor® 647 | 1.0 ml | 100 μg | 600-650 | 668 | Hamster IgG Alexa Fluor® 647 | Code HM21 |
| HM3411 | Cy [™] 5 | 0.5 ml | 100 μg | 600-650 | 670 | _ | |

PRODUCT DESCRIPTION

Hamster monoclonal antibody to mouse CD3

Clone: 500A2

Isotype: Hamster IgG

Immunogen: C6VL-BS cell lysate¹

Lot No.: See label Expiration: See label

Buffer: Phosphate buffered saline (PBS)

Preservatives: 0.1% *sodium azide*. Sodium azide is an extremely toxic and dangerous compound particularly when combined with acids or metals. Solutions containing sodium azide should be disposed of properly.

Stabilizer: For conjugated products only, a highly purified grade of BSA has been added as a stabilizing protein.

STORAGE & HANDLING

Store reagents at 2-8°C. Light exposure should be avoided with fluorochrome conjugated reagents. Use dim light during handling, incubation with cells and prior to analysis. It is recommended that cells be analyzed within 18 hours of staining. If the reagent is being diluted, it is recommended that only the quantity to be used within one week be diluted.

PRODUCT CHARACTERIZATION

Antigen Specificity: The 500A2 monoclonal antibody (mAb) is specific for the ε chain of mouse CD3¹. In the mouse, CD3 is highly expressed by mature T cells and is expressed at lower levels by CD4⁺CD8⁺ thymocytes². The ε chain of CD3 is a component of the multimeric T cell receptor complex and is involved with signal transduction. Immobilized 500A2 mAb can be used to stimulate mouse T cells in vitro¹. A combination of immobilized 500A2 mAb and anti-mouse CD28 mAb (Cat. # HM3500) is commonly used for the activation of mouse T cells³. Other reported applications for this antibody include immunoprecipitation, IHC with acetone-fixed frozen sections, and immunostaining for flow cytometry¹.

PRODUCT QUALITY CONTROL

Every lot is tested by flow cytometry using freshly harvested mouse splenocytes. From this testing it is recommended that between 0.1 and 0.25 μg of antibody be used per 1 x 10^6 cells in a 100 μl staining volume.

Because conditions may vary, it is recommended that each investigator determine the optimal amount of antibody to be used for each application.

REFERENCES:

- Allison, J. P., W. L. Havran, M. Poenie, J. Kimura, L. DeGraffenreid, S. Ajami, G. Duwe, A. Weiss, and R. Tsien. 1988. Expression and function of CD3 on murine thymocytes. *The T-Cell Receptor*, UCLA Symposia, 73rd edition, Kappler, J., and M. Davis, eds. pp. 33-45.
- Havran, W. L., M. Poenie, J. Kimura, R. Tsien, A. Weiss, and J. P. Allison. 1987. Expression and function of the CD3-antigen receptor on murine CD4⁺8⁺ thymocytes. *Nature* 330: 170-173.
- Harding, F. A., J. G. McArthur, J. A. Gross, D. H. Raulet, and J. P. Allison. 1992. CD28-mediated signaling co-stimulates murine T cells and prevents induction of energy in T cell clones. *Nature* 356: 607-609.
- * Antibody value assigned is based on the Optical Density at 280 nm.

TR, Texas Red®

TC, TRI-COLOR®, PE-Cy5

The efficiency of energy transfer in tandem dyes can be significantly decreased by exposure to visible light. We recommend that longer wavelength fluorochrome conjugates, e.g. PE-Cy7, PE-Alexa Fluor® 700, be protected from light during staining and while awaiting analysis, e.g. cover with aluminum foil.

FIX & PERM® and **COMBI-IC** reagents are produced for Caltag Laboratories by An Der Grub Bio Research GmbH, Austria.

The Texas Red®, Alexa Fluor® and Pacific Blue® dye conjugates in this product are sold under license from Molecular Probes, Inc., for research use only or as analyte specific reagents, except for use in combination with microarrays or high content screening, and are covered by pending and issued patents.

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