



Recombinant Version of Classic Clone

**Cat nr AE00333**

**Product Datasheet**

Mouse Recombinant Antibody, rM2 to:

**DYKDDDDK epitope tag**

Epitope tag, also known as FLAG tag, or DDDDK tag, for the detection of recombinant proteins tagged with the DYKDDDDK sequence, which can be C-terminal, N-terminal, or inside the target protein.

Cellular localization

Official Symbol (Gene)

GenelD

SwissProt

Confirmed Applications	ELISA, ICC, IHC, IAP, IP, WB
Positive controls	multitag protein
Aeonian Rating©	100

Purification By Protein A from bioreactor concentrate

Formulation  1 mg IgG/ml in PBS with 0.02% Proclin 300

Amount  200ug  1000ug

Isotype  Mouse IgG1, kappa, recombinant version of mouse IgG1 clone M2.1

Immunogen DYKDDDDK peptide

Epitope DYKDDDDK

Storage instructions	Avoid repeated freeze/thaw cycles. For long term storage, keep small aliquots at -20C or -80C and keep one aliquot at 4C for daily experimentations. Azide will preserve antibody at 4C for 6-12 months, when kept away from direct sun light.
Expiration	Integrity warranted for 24 months after purchase when handled and stored according to instructions, see below.
Warranty	This product is only warranted for the specifications as described in this product sheet and only when the product is handled and stored according to instructions. User should validate this antibody in the application and tissue/cell type as required, after confirmation of integrity upon receipt is obtained by reproducing the performance as described below. Should such confirmation not be attempted, any warranty is void. In case of non-conformance, user needs to contact us immediately for replacement or refund.
Liability	This product is for in vitro research use only. Any other applications, such as diagnostics or therapeutics, or in vivo experiments, and the validation of this product therein, are solely at the responsibility of the buyer/user.
Product performance	see next pages

## Product data:

### Immunoassays

#### ImmunoAffinity Purification (IAP):

The original clone M2 was successfully used to affinity purify DYKDDDDK-tagged bacterial alkaline phosphatase.

Brizzard BL, Chubet RG, Vizard DL. Immunoaffinity purification of FLAG epitope-tagged bacterial alkaline phosphatase using a novel monoclonal antibody and peptide elution. *Biotechniques*. 1994 Apr;16(4):730-5. PMID: 8024796.

#### ImmunoPrecipitation (IP):

The original clone M2 was successfully used to pull down epitope-peptides from a DNA display library and 3X DYKDDDDK-tagged protein from yeast lysates using M2-agarose beads. It was also successfully used to immunoprecipitate DYKDDDDK-tagged human MUC1 from CHO cells.

Yonezawa M, Doi N, Kawahashi Y, Higashinakagawa T, Yanagawa H. DNA display for in vitro selection of diverse peptide libraries. *Nucleic Acids Res*. 2003 Oct 1;31(19):e118. doi: 10.1093/nar/gng119. PMID: 14500846.

Gerace E, Moazed D. Affinity Pull-Down of Proteins Using Anti-FLAG M2 Agarose Beads. *Methods Enzymol*. 2015;559:99-110. doi: 10.1016/bs.mie.2014.11.010. PMID: 26096505.

Hoshino H, Ohta M, Ito M, Uchimura K, Sakai Y, Uehara T, Low S, Fukushima M, Kobayashi M. Apical membrane expression of distinct sulfated glycans represents a novel marker of cholangiolocellular carcinoma. *Lab Invest*. 2016 Dec;96(12):1246-1255. doi: 10.1038/labinvest.2016.104. PMID: 27748735.

#### Enzyme-Linked immunosorbent assay (ELISA):

The original clone M2 was successfully used as a reporting antibody to detect DYKDDDDK-tagged peptides selected from a phage display library.

Srila W, Yamabhai M. Identification of amino acid residues responsible for the binding to anti-FLAG™ M2 antibody using a phage display combinatorial peptide library. *Appl Biochem Biotechnol*. 2013 Oct;171(3):583-9. doi: 10.1007/s12010-013-0326-8. PMID: 23807489.

#### ImmunoCytoChemistry (ICC):

The original clone M2 was successfully used to detect DYKDDDDK-tagged D1 and D2 in HEK293 cells, and DYKDDDDK-tagged mLOX in MCF7 cells.

Baqui MM, Gereben B, Harney JW, Larsen PR, Bianco AC. Distinct subcellular localization of transiently expressed types 1 and 2 iodothyronine deiodinases as determined by immunofluorescence confocal microscopy. *Endocrinology*. 2000 Nov;141(11):4309-12. doi: 10.1210/endo.141.11.7872. PMID: 11089566.

Kirschmann DA, Seftor EA, Fong SF, Nieva DR, Sullivan CM, Edwards EM, Sommer P, Csiszar K, Hendrix MJ. A molecular role for lysyl oxidase in breast cancer invasion. *Cancer Res*. 2002 Aug 1;62(15):4478-83. PMID: 12154058.

#### ImmunoHistoChemistry (IHC):

The original clone M2 was successfully used to detect DYKDDDDK-tagged mCREB in transgenic mouse tissue

Newton SS, Dow A, Terwilliger R, Duman R. A simplified method for combined immunohistochemistry and in-situ hybridization in fresh-frozen, cryocut mouse brain sections. *Brain Res Brain Res Protoc*. 2002 Jun;9(3):214-9. doi: 10.1016/s1385-299x(02)00148-4. PMID: 12113781.

**Western Blot (WB):**

The original clone M2 was successfully used to detect several 3X DYKDDDDK-tagged proteins from *Vibrio cholerae* lysates.

Shin JH, Lanz M, Smolka MB, Dörr T. Characterization of an anti-FLAG antibody binding protein in *V. cholerae*. *Biochem Biophys Res Commun*. 2020 Jul 30;528(3):493-498. doi: 10.1016/j.bbrc.2020.05.169. PMID: 32505345.



