

Anti-puromycin [3RH11] Standard Size, 50  $\mu g,$  Ab02366-21.0 View online

## Anti-puromycin [3RH11] Standard Size Ab02366-21.0

This antibody does not have a J-chain and therefore presents as a hexamer, rather than a pentamer.

Isotype and Format: Mouse IgM, Kappa

Clone Number: 3RH11

Alternative Name(s) of Target: aminonucleoside antibiotic puromycin

**UniProt Accession Number of Target Protein:** 

Published Application(s): functional assays, WB, ELISA, IF

Published Species Reactivity: Species independent

**Immunogen:** The original mouse IgG1 antibody was raised against puromycin hydrochloride. Specificity: This antibody recognizes puromycin, an aminonucleoside antibiotic, derived from the Streptomyces alboniger bacterium, that causes premature chain termination during translation. Part of the molecule resembles the 3' end of the aminoacylated tRNA, making it useful for protein translation analysis. **Application Notes:** This monoclonal antibody to puromycin provides a non-radioactive method to measure rates of global protein synthesis (mRNA translation) in cells or tissue slices incubated with puromycin, or animals treated with puromycin in vivo (Frolinger et al., 2018; pmid: 29702026). 3RH11 offers advantageous alternative to traditional pulse-chase methods, which rely on radioactive amino acid labeling as it allows for the simple evaluation and quantification of translation directly using standard immunochemical methods (Balukoff et al., 2020; pmid: 33188200). Classical pulse-chase or flooding dose methods used to monitor protein synthesis rely on the measurement of radioactive methionine and cysteine labels. Analysis using puromycin immunodetection is an advantageous alternative to radioactive amino acid labeling, and allows for the evaluation/quantification of translation directly using standard immunochemical methods (Frolinger et al., 2018; pmid: 29702026). Practical comments: Puromycin inhibits protein synthesis, which some cells may be more or less sensitive to. It is therefore highly recommended that the concentration of puromycin for protein translation assays be optimized for cell type. The suggested concentration of puromycin is 1uM, but for cells that are more sensitive, lower concentrations of puromycin may give better results. Because cell growth (and protein synthesis) slows dramatically as cell near confluence, it is recommended that experiments are performed during growth phase (40-50% confluence). Label with an optimized concentration of puromycin for at least 20-30 min. 3RH11 is a very popular antibody used in countless experiments analysing global protein synthesis (mRNA translation) (Balukoff et al., 2020; pmid: 33188200; Crowell et al., 2016; pmid: 27464336; Khoutorsky et al., 2015; pmid: 26678009; Frolinger et al., 2018; pmid: 29702026; Wang et al., 2016; pmid: 27454289).

Antibody First Published in: Kelleher et al. The mTORC1 signaling repressors REDD1/2 are rapidly

induced and activation of p70S6K1 by leucine is defective in skeletal muscle of an immobilized rat hindlimb Am J Physiol Endocrinol Metab. 2013 Jan 15;304(2):E229-36. doi: 10.1152/ajpendo.00409.2012. Epub 2012 Nov 27. PMID:23193052

**Note on publication:** Describes the method of measuring protein synthesis using an anti-puromycin antibody.

## **Product Form**

**Size:** 50 µg Purified antibody.

Purification: Affinity Purified using a recombinant lectin column

**Supplied In:** PBS with 0.02% Proclin 300.

**Storage Recommendation:** Store at 4°C for up to 3 months. For longer storage, aliquot and store at - 20°C.

Concentration: 1 mg/ml.

Important note – This product is for research use only. It is not intended for use in therapeutic or diagnostic procedures for humans or animals.