UBE2D2, His-Tag Recombinant

Catalog: 79370 240111

Product Information

Description: Recombinant Xenopus UBE2D2 (ubiquitin-conjugating enzyme E2 D2), full length. This

construct contains an N-terminal His-tag. This protein was affinity purified.

Species:

Construct: UBE2D2 (His-Full Length) (Xenopus)

Concentration: 0.10 mg/ml **Expression System:** E. coli ≥90% **Purity:**

Format: Aqueous buffer solution.

Formulated In: 50 mM Sodium Phosphate, pH 7.0, 300 mM NaCl, 150 mM Imidazole, 0.1 mM PMSF,

0.25 mM DTT, and 25% glycerol

MW: 17 kDa

Genbank Accession: NM 001093036

Stability: At least 6 months at -80°C.

Storage:

Instructions for Use: Thaw on ice and gently mix prior to use. DO NOT VORTEX. Perform a quick spin before

opening. Aliquot into small volumes and flash freeze for long term storage. Avoid

multiple freeze/thaw cycles.

Specific Activity: 25 pmol/min/ug

Assay Conditions: UBE2D2 activity was measured by using wild-type ubiquitin diluted in Ubiquitination

Buffer to a working concentration of 170 ng/µl, in an AMP-Glo™ based assay (Promega #V5011). The reaction was initiated by mixing increasing amounts of UBE2D2 with UBA1 (ubiquitin-like modifier activating enzyme 1), BIRC7 (baculoviral IAP-repeat containing protein 7) and ATP in Ubiquitination Buffer (40 mM Tris (pH

7.5), 20 mM MgCl₂, 0.1 mg/ml BSA and 0.5 mM DTT).

First a 2x Master Mix containing 170 ng/ µl of ubiquitin, 15 ng/µl UBA1, 48 ng/µl of BIRC7 and 50 μM ATP was prepared. 5 μl of 2x Master Mix were added to 5 μl of

UBE2D2 serial dilutions prepared at 2x the final desired concentrations in Ubiquitination Buffer, and incubated for 60 minutes at 37°C. The plate was

equilibrated to room temperature (RT). 10 µl of AMP-Glo™ Reagent I were added, and the plate was incubated at RT for another 60 minutes. 20 µl of Detection Solution were added to each reaction and the plate was incubated at RT for 30 minutes. The plate was read on a GloMax plate reader (Promega #E7031) using the KinaseGlo Luminescence Protocol. The Enzyme Specific Activity (SA) was calculated as follows: [[(Concentration of AMP generated in μΜ) x (Reaction volume in μΙ)]/ [(Reaction time in min) *(Enzyme amount in mg)]]* 10-3. The blank was determined from a "no enzyme" sample by replacing UBE2D2 with an equal volume of Ubiquitination Buffer. Useful for the study of enzyme kinetics, screening inhibitors, and selectivity profiling.

Applications:



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Quality Control Data

4-20% SDS-PAGE Coomassie Staining

1 2 - 170 kBa - 95 kDa - 95 kDa - 95 kDa - 55 kDa - 55 kDa - 43 kDa - 34 kDa - 26 kDa - 17 kDa - 10 kDa

UBE2D2 Activity

