Ac-ANW-AMC

Cat. No.: HY-D1705

CAS No.: 2357123-49-8

Molecular Formula: $C_{30}H_{32}N_6O_7$

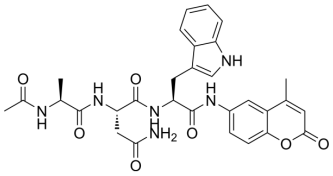
Molecular Weight: 588.61

Target: Proteasome

Pathway: Metabolic Enzyme/Protease

Storage: -20°C, protect from light

* In solvent : -80°C, 6 months; -20°C, 1 month (protect from light)



SOLVENT & SOLUBILITY

In Vitro

DMSO : ≥ 100 mg/mL (169.89 mM)

* "≥" means soluble, but saturation unknown.

Preparing Stock Solutions

Solvent / Mass / Concentration	1 mg	5 mg	10 mg
1 mM	1.6989 mL	8.4946 mL	16.9892 mL
5 mM	0.3398 mL	1.6989 mL	3.3978 mL
10 mM	0.1699 mL	0.8495 mL	1.6989 mL

Please refer to the solubility information to select the appropriate solvent.

BIOLOGICAL ACTIVITY

Description

Ac-ANW-AMC is a fluorogenic substrate for immunoproteasome. Ac-ANW-AMC can be used to measure β5i activity (Ex=345 nm, Em=445 nm)[1][2].

IC_{50} & Target

β5i[1]

In Vitro

Guidelines (Following is our recommended protocol. This protocol only provides a guideline, and should be modified according to your specific needs)[2].

1. For the proteasome activity assay, cells are lysed in the proteasome activity lysis buffer.
2. Cells are homogenized by passing the lysates 15 times through a 26G×1/2″ needle attached to a 1-mL syringe.
3. The lysates are then centrifuged at 12,000 rpm for 15 min. Protein concentrations of the samples are determined using the Bradford assay.
4. Proteasome activity is determined using model peptide substrates by measuring free Ac-ANW-AMC fluorescence on a TECAN infinite m200 fluorometer.
5. The fluorescence unquenched after hydrolysis by proteasomes is monitored every three minutes at 345 nm excitation and 445nm emission wavelengths at 30°C.

Note: Each sample is assayed in triplicate.

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

REFERENCES

[1]. Winter, M.B., La Greca, F., Arastu-Kapur, S., et al. Immunoproteasome functions explained by divergence in cleavage specificity and regulation. eLife 6:e27364, (2017).

[2]. Sumin Kim, et al. Evaluation of Immunoproteasome-Specific Proteolytic Activity Using Fluorogenic Peptide Substrates. Immune Netw. 2022 Apr 15;22(3):e28.

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