

Ac-YVAD-cmk

Caspase-1 inhibitor

Catalog code: inh-yvad

<https://www.invivogen.com/ac-yvad-cmk>

For research use only

Version 20110-MM

PRODUCT INFORMATION

Contents

- 5 mg Ac-YVAD-cmk

Storage and stability

- Ac-YVAD-cmk is shipped at room temperature. Upon receipt, store at -20°C.

- Upon resuspension, prepare aliquots of Ac-YVAD-cmk and store at -20°C. Resuspended Ac-YVAD-cmk is stable for 6 months when properly stored.

Quality control

- Purity ≥97% (UHPLC)

- The inhibitory activity has been validated using in-house cellular assays.

- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK Blue™ TLR4 cells.

DESCRIPTION

Ac-YVAD-cmk is a potent and irreversible inhibitor of the inflammatory caspase-1¹. Caspase-1, also known as IL-1 converting enzyme (ICE), is a cysteine protease that cleaves the precursors of the IL-1β and IL-18 pro-inflammatory cytokines, as well as the gasdermin D (GSDMD) pore-forming protein². Ac-YVAD-cmk is a tetrapeptide sequence based on the target sequence of caspase-1 in pro-IL-1β (YVHD)^{1,3}. This drug was described as blocking inflammatory cell death in experimental models⁴. Additional reports showed that Ac-YVAD-cmk effectively blocks inflammasome activation, and that it displays anti-inflammatory, anti-apoptotic, and anti-pyroptotic effects^{5,6}.

1. Garcia-Calvo M. *et al.*, 1998. Inhibition of human caspases by peptide-based and macromolecular inhibitors. *J Biol Chem.* 273(49):32608-13. 2. Talanian, R.V., *et al.*, 1997. Substrate specificities of caspase family proteases. *J Biol Chem.* 272(15):9677-82. 3. Schierle G.S. *et al.*, 1999. Caspase inhibition reduces apoptosis and increases survival of nigral transplants. *Nat Med.* 5(1):97-100. 4. Van Opdenbosch N. *et al.*, 2014. Activation of the NLRP1b inflammasome independently of ASC-mediated caspase-1 autoproteolysis and speck formation. *Nat Commun.* 5:3209. 5. Zhang F. *et al.*, 2016. The caspase-1 inhibitor AC-YVAD-CMK attenuates acute gastric injury in mice: involvement of silencing NLRP3 inflammasome activities. *Sci Rep.* 6:24166.

CHEMICAL PROPERTIES

Solubility: 50 mg/ml (92.4 mM) in DMSO

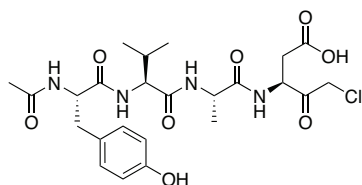
Synonym: Ac-Tyr-Val-Ala-Asp-chloromethyl ketone

CAS number: 178603-78-6

Formula: C₂₄H₃₀ClN₄O₈

Molecular weight: 541 g/mol

Structure:



METHODS

Preparation of 10 mg/ml (18.5 mM) stock solution

- Add 500 µl of DMSO to 5 mg Ac-YVAD-cmk. Mix by vortexing.
- Prepare further dilutions with endotoxin-free water.

Working concentration: 0.1-30 µg/ml for cell culture assays

In vitro inhibition of caspase-1:

The following protocol describes the monitoring of caspase-1 inhibition in human THP1-Null2 cells by assessing the inhibition of IL-1β production.

1. Pre-incubate THP1-Null2 cells (3x10⁵ cells/well) with Ac-YVAD-cmk (0.1-30 µg/ml) in a 96-well plate for 1 hour at 37°C in 5% CO₂.
2. Prime cells by adding 1 µg/ml LPS-EK for 3 hours at 37°C in 5% CO₂.
3. Gently remove medium and add 180 µl of fresh test medium.
4. Stimulate cells by adding IL-1β inducers, such as MSU crystals (100-200 mg/ml) in the presence or absence of Ac-YVAD-cmk.
5. Incubate from 6 hours to overnight at 37°C in 5% CO₂.
6. Determine caspase-1 inhibition by detecting mature IL-1β with InvivoGen's HEK-Blue™ IL-1β cells, which are specifically engineered to detect bioactive IL-1β.

PROTOCOLS

For reference only; as described in the indicated publications.

Cell Culture Assay³

Cells: Bone marrow cells

Working concentration: 50 µM (27 µg/ml)

Incubation time: 30 minutes

Method: Detection of pyroptosis by measuring released lactate dehydrogenase activity

Animal Study⁴

Animal model: BALB/c mice

Dose: 8 mg/kg

Administration: Intraperitoneal injection

RELATED PRODUCTS

Product	Description	Cat. Code
LPS-EK	LPS from <i>E. coli</i> K12	tlrl-eklps
Nigericin	Inflammasome inducer	tlrl-nig
MSU Crystals	Inflammasome inducer	tlrl-msu
Poly(dA:dT)	Inflammasome inducer	tlrl-patn
THP1-Null2 Cells	Human monocytes	thp-null2
HEK-Blue™ IL-1β cells	IL-1β reporter cells	hkb-il1b
VX-765	Caspase -1 and -4 inhibitor	inh-vx765i-1
Z-VAD-FMK	Pan-caspase inhibitor	tlrl-vad

TECHNICAL SUPPORT

InvivoGen USA (Toll-Free): 888-457-5873

InvivoGen USA (International): +1 (858) 457-5873

InvivoGen Europe: +33 (0) 5-62-71-69-39

InvivoGen Hong Kong: +852 3622-34-80

E-mail: info@invivogen.com