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Product Information

Protease Inhibitor Cocktail

For use with mammalian cell and tissue extracts, DMSO solution

P8340

Product Description

Crude cell extracts contain various endogenous enzymes, such as proteases and phosphatases, which can degrade proteins in the extracts. The best way to increase the yield of intact proteins is to add inhibitors of those enzymes known to be present.

The P8340 protease inhibitor cocktail has been optimized and tested for mammalian cell and tissue extracts. P8340 contains inhibitors with a broad specificity for serine, cysteine, and acid proteases, and aminopeptidases.

P8340 is supplied as a ready-to-use solution in DMSO. The inhibitors in P8340 are as follows, with respective specific inhibitor targets and target classes of each inhibitor listed:

- AEBSF [4-(2-Aminoethyl)benzenesulfonyl fluoride hydrochloride]: serine proteases, such as trypsin, chymotrypsin, plasmin, kallikrein and thrombin
- Aprotinin: serine proteases, such as trypsin, chymotrypsin, plasmin, and kallikrein; human leukocyte elastase, but not pancreatic elastase
- Bestatin hydrochloride: aminopeptidases, such as leucine aminopeptidase and alanyl aminopeptidase¹⁻⁴
- E-64 [*N*-(trans-Epoxysuccinyl)-L-leucine 4-guanidinobutylamide]: cysteine proteases, such as calpain, papain, cathepsin B, and cathepsin L
- Leupeptin hemisulfate salt: serine proteases and cysteine proteases, such as plasmin, trypsin, papain, and cathepsin B
- Pepstatin A: acid proteases, such as pepsin, renin and cathepsin D, and many microbial aspartic proteases

Several theses 5 and dissertations $^{6\mathchar`-22}$ have cited use of product P8340 in their protocols.

Storage/Stability

Store the product at -20 °C.

Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Usage

One mL of P8340 is recommended for the inhibition of endogenous enzymes found in 100 mL of lysate from 20 g (wet weight) of bovine liver, or in 10 mL of cell lysate from CHO cells at a cell density of 10⁸ cells per mL. CHO cells were grown on DMEM with 10% FCS (heat-inactivated).

Note: Not all lysates contain the same levels of endogenous enzymes. It may be necessary to adjust the volume of cocktail required.

References

- Umezawa, H., Ann. Rev. Microbiol., 36, 75-99 (1982).
- Aoyagi, T. *et al.*, *Biochem. Int.*, **9(4)**, 405-411 (1984).
- Aoyagi, T., and Umezawa, H., Acta Biol. Med. Ger., 40(10-11), 1523-1529 (1981).
- 4. Mumford, R.A. *et al.*, *Biochem. Biophys. Res. Comm.*, **103(2)**, 565-572 (1981).
- 5. Wang, Yifei, "MYC-binding lncRNA EPIC1 promotes AKT-mTORC1 signaling and Rapamycin resistance in breast and ovarian cancer". University of Pittsburgh, M.S. thesis, p. 7 (2020).
- Crosland, Rachel, "Studies of The PTEN Tumour Suppressor in Endometrial Cancer". Sheffield Hallam University, Ph.D. dissertation, p. 85 (2004).



- Wiczer, Brian Michael, "Biochemical and functional characterization of fatty acid trasnport proteins". University of Minnesota, Ph.D. dissertation, p. 35 (2009).
- Malkus, Kristen Ashley, "Regional Lysosomal Malfunction Underlies the Selectivity of a-Synuclein Neurodegeneration". University of Pennsylvania, Ph.D. dissertation, pp. 42, 43, 48, 49, 77, 96, 97, 99 (2011).
- Yu, Daozhan, "Omentin Activates AMP-activated Protein Kinase and Plays a Role in Energy Metabolism and Immune Response". University of Maryland Baltimore, Ph.D. dissertation, p. 41 (2011).
- Gray, Amy Jetaun, "Novel Phosducin-Like Protein Binding Partners: Exploring Chaperone and Tumor Suppressor Protein Interactions". Brigham Young University, Ph.D. dissertation, pp. 18, 36, 67 (2012).
- Rajgor, Dipen, "Characterisation of the multi-isomeric protein nesprin-1 in p-body and mRNA dynamics". King's College London, Ph.D. dissertation, p. 94 (2012).
- Hoque, Rukshana, "The effects of quercetin on iron metabolism". King's College London, Ph.D. dissertation, p. 72 (2013).
- Seabrook, Jill L., "The role of LIN28 in the molecular regulation of placenta development and function". Colorado State University, Ph.D. dissertation, pp. 59, 87 (2013).
- 14. Zano, Stephen P., "Therapeutic Approaches for the Treatment of Canavan Disease and Regulation of Bacterial Quorum Sensing Pathway". University of Toledo, Ph.D. dissertation, p. 14 (2013).
- Bartkowiak, Bartlomiej, "Characterization of dCDK12, hCDK12, and hCDK13 in the Context of RNA Polymerase II CTD Phosphorylation and Transcription-Associated Events". Duke University, Ph.D. dissertation, pp. 63, 65, 85 (2014).
- Nagati, Jason Sharif, "Dual mechanisms regulating alpha subunit-specific activity in hypoxia-inducible factor signaling". University of Texas Southwestern Medical Center, Ph.D. dissertation, p. 88, 95 (2015).
- 17. Pua, Khian Hong, "Investigating the Mechanism of Action of Sanglifehrin A". Harvard University, Ph.D. dissertation, p. 74 (2015).

- Tang, Zhen (Sophie), "Does the IGF axis influence EMT to play a role in bladder cancer progression?" University of Bristol, Ph.D. dissertation, p. 63 (2017).
- Chan, Jessica Sze Ki, "The Oncogenic Role of TRIP13 in Hepatocellular Carcinoma". Ruperto Carola University Heidelberg, Dr. rer. nat. dissertation, p. 88 (2018).
- 20. Kulas, Joshua Adam, "Amyloid Precursor Protein And Insulin Homeostasis". University of North Dakota, Ph.D. dissertation, p. 24 (2018).
- 21. Ravandi, Elnaz Ghotbi, "Recruitment of Polycomb-Group Proteins at *giant* in *Drosophila* Embryos". Southern Methodist University, Ph.D. dissertation, pp. 41, 42, 50 (2019).
- Sharma, Aman, "Mechanism of DNA damage associated with Estrogen Receptor Alpha – interplay of non-canonical DNA secondary structures". University of Massachusetts Amherst, Ph.D. dissertation, p. 30 (2021).

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