



Product Information

PROTEASOME FRACTION II RABBIT RETICULOCYTE CELL EXTRACT

Product Number **F 4051**

Product Description

Proteasome Fraction II is the protein fraction of a reticulocyte cell extract that binds to an anion exchange resin. It is essentially free of ubiquitin and ATP and contains E1, most E2s, ubiquitin C-terminal hydrolases (UCHs), and the 20S and 26S proteasomes.

Degradation of short-lived, key regulatory proteins by the ubiquitin-proteasome pathway plays key roles in a number of cellular processes. A number of proteins are degraded by this system including: cyclins, cyclin-dependent kinases^{1,2} and their inhibitors, tumor suppressors, oncoproteins, and transcriptional activators and their inhibitors. Two discrete steps are involved in the ubiquitin-mediated degradation of proteins: signaling by covalent conjugation of multiple ubiquitin moieties and degradation of the tagged substrate. Conjugation occurs by a three-step mechanism involving three different enzymes that act sequentially: E1, E2 and E3. Ubiquitin-activating enzyme (E1) catalyzes the activation of ubiquitin then E2 (ubiquitin-conjugating enzyme) transfers activated ubiquitin to E3, which is bound to substrate. E3 catalyzes the polyubiquitination of the targeted protein. The polyubiquitin tagged protein is then degraded by the 26S proteasome in an ATP-dependent process, and free ubiquitin is released. The 20S proteasome is the ATP-independent catalytic core of the 26S proteasome.³⁻⁵

Reagent

Proteasome Fraction II is supplied as a solution in 25 mM HEPES, pH 7.6.

Precautions and Disclaimer

For laboratory use only. Not for drug, household or other uses. Please consult the Material Safety Data Sheet for handling recommendations before working with this material.

Storage/Stability

Proteasome Fraction II solution should be stored in aliquots at -70 °C. Avoid multiple freeze thaw cycles. Do not store in a frost-free freezer.

Product Profile

Proteasome Fraction II is ideal for demonstrating ubiquitin- and ATP-dependent degradation/conjugation of radiolabeled or immunodetectable substrates. Isopeptidase inhibitor and a proteasome inhibitor are highly recommended for the accumulation of ubiquitin protein conjugates. Typical assay concentration for FII is 0.5 - 4 mg/ml.

References

1. DeSalle, L.M. and Pagano, M., Regulation of the G1 to S transition by the ubiquitin pathway. *FEBS Lett.*, **490**, 179-189 (2001).
2. Yew, P.R., Ubiquitin-mediated proteolysis of vertebrate G1 and S-phase regulators. *J. Cell Physiol.*, **187**, 1-10 (2001).
3. Tanaka, K., et al., The ligation systems for ubiquitin and ubiquitin-like proteins. *Mol. Cells*, **8**, 503-512 (1998).
4. Myung, J., et al., The ubiquitin-proteasome pathway and proteasome inhibitors. *Med. Res. Rev.*, **21**, 245-273 (2001).
5. Benaroudj, N., et al., The unfolding of substrates and ubiquitin-independent protein degradation by proteasomes. *Biochimie*, **83**, 311-318 (2001).

Manufactured for Sigma by Boston Biochem., Inc.

DMG 01/02

Sigma brand products are sold through Sigma-Aldrich, Inc.

Sigma-Aldrich, Inc. warrants that its products conform to the information contained in this and other Sigma-Aldrich publications. Purchaser must determine the suitability of the product(s) for their particular use. Additional terms and conditions may apply. Please see reverse side of the invoice or packing slip.