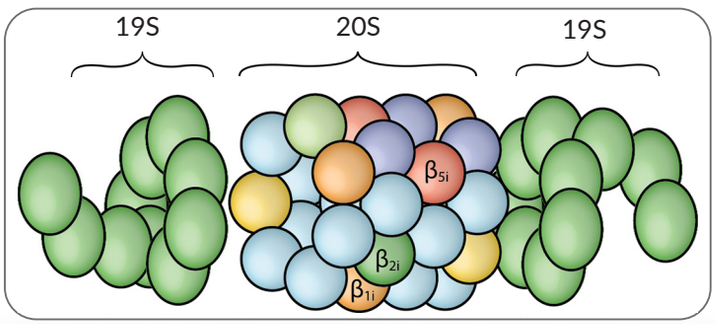
26S Proteasome (HEK 293)

Cat. No. SBB-PP0101
Lot. No. 232510101

South Bay Bío

26 Proteasome

The 26S Proteasome is the major nonlysosomal protease in eukaryotic cells and is responsible for the degradation of ubiquitinated substrates and misfolded proteins. It is composed of two subcomplexes: the 20S Proteasome core particle and the 19S Proteasome regulatory particle. The 20S Proteasome facilitates proteolytic cleavage of protein substrates and is composed of 28 subunits arranged into four stacked rings[1,2]. The outer rings of the 20S Proteasome are composed of seven related but nonidentical, noncatalytic subunits, alpha17, that form a gate and restrict substrate access. The inner rings of the 20S Proteasome are composed of seven related but nonidentical subunits, beta 1-7. Beta 1, 2, and 5 have proteyolytic activity. The 19S Proteasome caps one or both ends of the core particle and regulates substrate access to the catalytic core in an ATPdependent manner by modulating 20S Proteasome conformation[3,5]. The 19S Proteasome consists of a base subcomplex and a lid subcomplex. The base subcomplex is composed of six AAA+family members, scaffolding proteins, and regulatory proteins involved in Ubiquitin recognition[2,6]. The 19S Proteasome lid subcomplex contains eight subunits plus one deubiquitinating enzyme, Rpn11. Small molecules that inhibit the activity of the 26S Proteasome are used to study the function of shortlived intracellular proteins and for the clinical treatment of certain forms of cancer (7). This highly purified 26S Proteasome preparation can be used in vitro for the degradation of peptide substrates and poly-ubiquitinated proteins.



Product Information

Quantity: 25 µg **Molecular Weight:** >2100 kDa

Concentration: 0.4 µM, 0.85 mg/mL

Purity: >95% by SDS-PAGE

Storage Buffer: 50 mM HEPES pH 7.5, 20 mM NaCl, 2 mM Mg-ATP, 10% Glycerol

Storage: Store at -80˚C. Avoid multiple freeze thaw cycles.

Usage: Typical working concentrations range is from 2.5 to 5 nM.

Quality Control and Performance Data

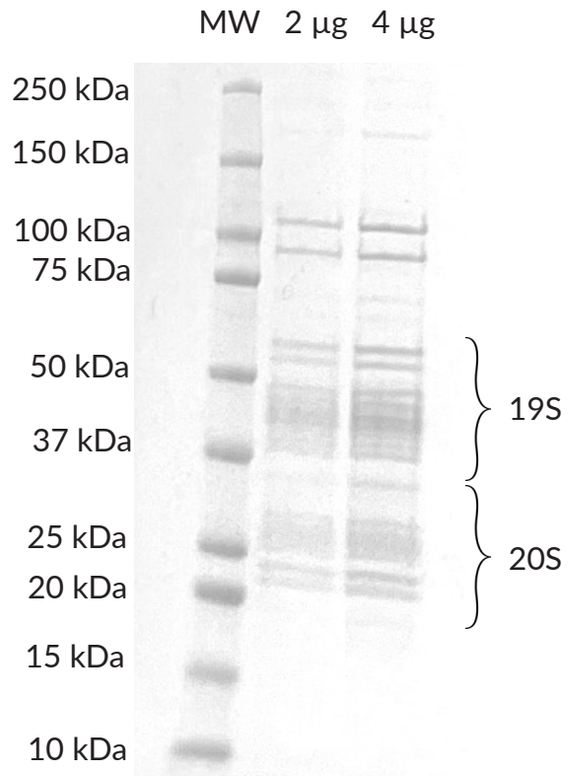


Figure 1. 26S Proteasome, SDS-PAGE. From left to right, increasing amounts of 26S Proteasome loaded onto a 4-20% SDS-PAGE gel, stained with coomassie brilliant blue.

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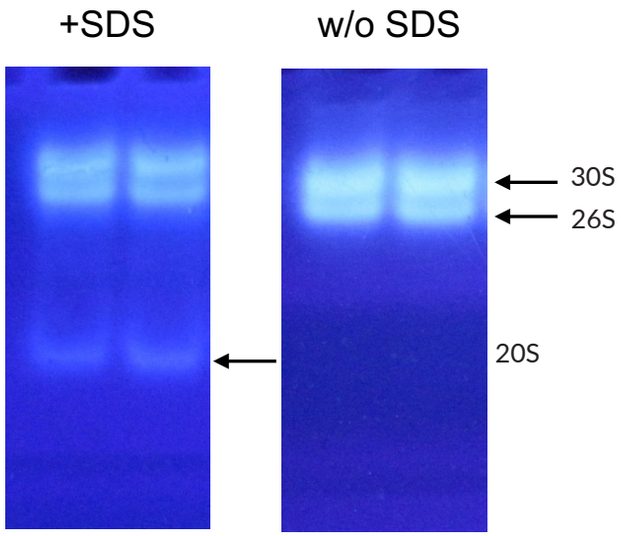

Figure 2. Proteasome was separated using a 5% native PAGE gels then analyzed for activity by soaking in LL-VY-AMC substrate. Left panel: Assay done with 0.035% SDS. Right panel: no SDS. Fluorescence shows double capped 30S and single capped 26S Proteasome.

References

1) Kim, H.M. et al. (2011), "Structure characterization of the 26S proteasome". Biochim. Biophys. Acta 1809:67.

2) Xie, Y. (2010), "Structure, assembly and homeostatic regulation of the 26S proteasome". J. Mol. Cell Biol. 2:308.

3) Adams, G.M. et al. (1998) "Formation of proteasome-PA700 complexes directly correlates with activation of peptidase activity". Biochemistry 37:12927.

4) ChuPing, M. et al. (1994) "ATP Binding by Proteasomal ATPases Regulates Cellular Assembly and Substrate-induced Functions of the 26 S Proteasome". J. Biol. Chem. 269:3539.

5) Kohler, A. et al. (2001) "The axial channel of the proteasome core particle is gated by the Rpt2 ATPase and controls both substrate entry and product release". Mol. Cell 7:1143.

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