Proteasome ELISA kit

The only commercially available kit for proteasome quantification.

This kit provides the means to quantify 20S proteasome concentrations in biological samples using a sandwich ELISA technique, utilizing two 20S proteasome specific antibodies for capture and detection purposes together with a highly sensitive substrate. Sample 20S proteasome levels are determined by comparison to a 20S proteasome calibration curve produced in parallel. This kit provides sufficient material for 1x96 well plate set-up to be run.

Proteasomes are non-lysosomal proteolytic complexes localised primarily in the cytoplasm and in the nucleus of eukaryotic cells. The 26S proteasome structure is composed of a 20S proteasome catalytic core complex and one or two 19S regulatory subcomplexes. The 20S core comprises two copies of 14 subunits (7 α-subunits and 7 β-subunits) arranged in a α7β7β7α7 cylindrical array. Varying catalytic subunit composition (β1, β1i; β2, β2i; β5, β5i) results in a variety of possible subtypes. The 19S regulatory subcomplexes, comprised of 6 ATPase and at least 10 non-ATPase subunits, specifically bind ubiquitinylated proteins and provide the 20S core with an ATP-ubiquitin–dependent proteolytic activity. The ubiquitin-proteasome system is the major non-lysosomal system for the degradation of short half-life proteins and peptides that are involved in basic cellular processes, such as cell-cycle regulation and apoptosis, transcriptional regulation, or antigen processing. Thus, protein degradation by the ubiquitin-proteasome pathway has a major regulatory function for proliferation activity and survival of both normal and malignant cells. The 20S proteasome has been detected in normal human blood plasma (known as circulating proteasome), possessing comparatively low specific activity and with a distinct pattern of subtypes. Proteasomes are often overexpressed in cancer cells; abnormally high expression of proteasomes having been found in human leukaemia cells, renal cancer cells and in breast cancer cell lines. In patients suffering from auto-immune diseases, malignant myelo-proliferative syndromes, multiple myeloma, acute and chronic lymphatic leukaemia, solid tumour, sepsis or trauma, the concentration of circulating proteasome has been found to be elevated, to correlate with the disease state, and may have prognostic significance. Proteasome levels have been measured by enzyme-linked immunosorbent assay (ELISA) techniques in cell lysates, serum or plasma samples. This approach has been used to show that proteasome concentrations in peripheral blood are elevated in patients with certain types of malignant

- Determination of 20S proteasome levels in biological samples (cell lysates, tissue extracts, plasma, serum)
- Comparison of 20S proteasome levels in plasma/serum samples associated with a particular disease/illness with samples from healthy controls
- Investigation of variation in 20S proteasome levels in abnormal cell lines/tissues

diseases, including multiple myeloma, suggesting that circulating proteasome levels may be correlated with tumour burden. The link between elevated circulating proteasome levels and disease activity has also been demonstrated in patients with systemic autoimmune diseases.

Citations: 20 View Online »

Ordering Information

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BML-PW0575-0001	96 wells

Manuals, SDS & CofA

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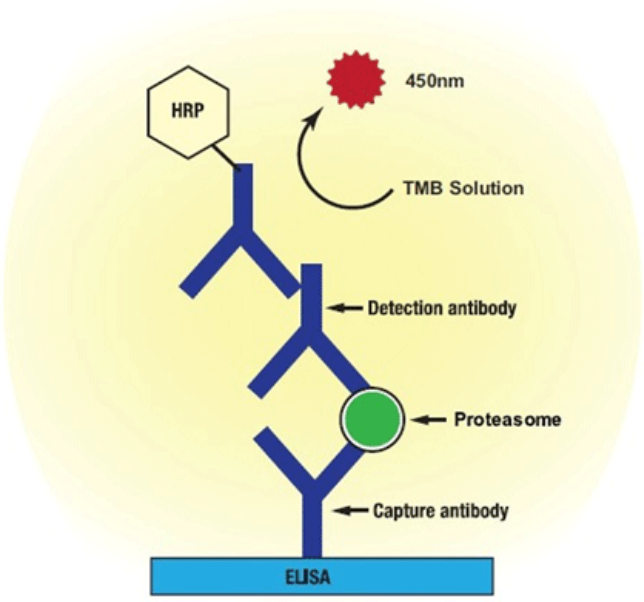

The ELISA plate is coated with the capture antibody and the proteasome sample is added and allowed to complex with the bound antibody. The detection antibody is then added, followed by the secondary antibody. Subsequent reaction between the activated TMB substrate/chromogen complex and horseradish peroxidase (HRP) conjugated secondary antibody produces a blue colored solution. After reaching the desired color intensity, the reaction is terminated by addition of the stop solution, which changes the solution color from blue to yellow. The plate is then analyzed at 450nm using a UV-Vis spectrophotometric plate reader.

Handling & Storage

Use/Stability	Stable for up to six months from receipt when stored at -20°C.
Handling	Avoid freeze/thaw cycles.
Long Term Storage	-20°C
Shipping	Blue Ice

Regulatory Status

RUO - Research Use Only

Product Details

Application	ELISA
Application Notes	For quantification of 20S proteasome levels in biological samples noting that the kit is not suitable for mouse or rat samples.
Assay Time	<3.5 hours
Compatibility	This product is compatible with the **Absorbance 96 Plate Reader**.
Contents	Plate, Capture Antibody, Detection Antibody, Conjugated Antibody, Proteasome Stock Solution, Binding Buffer, Wash Buffer, Blocking Buffer, ELISA Buffer, Lysis Buffer, TMB Substrate Solution, Stop Solution
Quantity	96 wells (24 tests in triplicate)
Species Reactivity	Human
Wavelength	450 nm

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