

P4HB Inhibitor Screening Assay Kit ab139480

2 References 画像数 3

医薬用外劇物

製品の概要

製品名	P4HB Inhibitor Screening Assay Kit
検出方法	Fluorescent
サンプルの種類	Purified protein, Inhibitor compounds
アッセイタイプ	Enzyme activity
検出感度	37 µM
製品の概要	<p>Abcam's P4HB (PDIA1) Inhibitor Screening Assay Kit (ab139480) provides a simple, homogenous assay for screening modulators of protein disulfide isomerase (PDIA1) enzymatic activity in a 96-well microplate. The basis for the assay is the P4HB (PDIA1)-catalyzed reduction of insulin in the presence of DTT, resulting in the formation of insulin aggregates which in turn bind to the red-emitting fluorogenic P4HB (PDIA1) Detection Reagent.</p> <p>Relative to analogous turbidimetric assays of P4HB (PDIA1) activity, the fluorescence-based assay provides a vastly improved assay signal window, improved lower detection limit and superior Z'-score (&gt;0.8).</p> <p>The kit includes human recombinant P4HB (PDIA1) enzyme as positive control, as well as the P4HB (PDIA1) inhibitor bacitracin as a negative control.</p> <p>This kit will provide a quantitative readout of P4HB (PDIA1) enzymatic activity in a robust and HTP fashion and can be applied to identification of P4HB (PDIA1) inhibitors from chemical libraries.</p>
試験プラットフォーム	Microplate reader

製品の特性

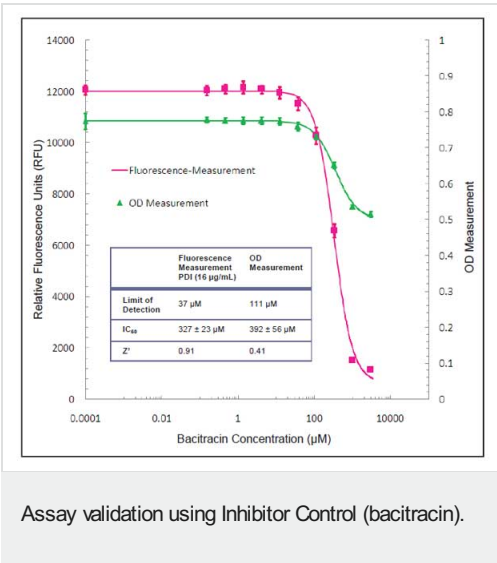
保存方法 Please refer to protocols.

内容	2 x 96 tests
Deionized Water	1 x 5ml
DTT	2 x 1.3ml

内容	2 x 96 tests
Inhibitor Control (Bacitracin)	1 vial
Insulin (from bovine pancreas)	2 vials
PBE Buffer	1 x 25ml
P4HB (PDIA1) (Human, Recombinant)	2 x 165µl
P4HB (PDIA1) Detection Reagent	1 x 20µl
Stop Reagent	1 x 1ml

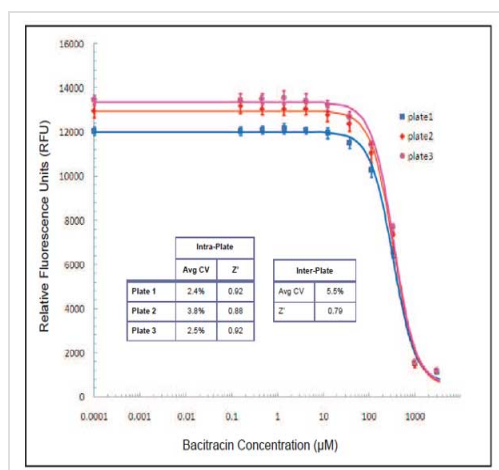
機能	This multifunctional protein catalyzes the formation, breakage and rearrangement of disulfide bonds. At the cell surface, seems to act as a reductase that cleaves disulfide bonds of proteins attached to the cell. May therefore cause structural modifications of exofacial proteins. Inside the cell, seems to form/rearrange disulfide bonds of nascent proteins. At high concentrations, functions as a chaperone that inhibits aggregation of misfolded proteins. At low concentrations, facilitates aggregation (anti-chaperone activity). May be involved with other chaperones in the structural modification of the TG precursor in hormone biogenesis. Also acts a structural subunit of various enzymes such as prolyl 4-hydroxylase and microsomal triacylglycerol transfer protein MTTP.
配列類似性	Belongs to the protein disulfide isomerase family. Contains 2 thioredoxin domains.
細胞内局在	Endoplasmic reticulum lumen. Melanosome. Cell membrane. Highly abundant. In some cell types, seems to be also secreted or associated with the plasma membrane, where it undergoes constant shedding and replacement from intracellular sources (Probable). Localizes near CD4-enriched regions on lymphoid cell surfaces. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

画像



Assay validation using Inhibitor Control (bacitracin).

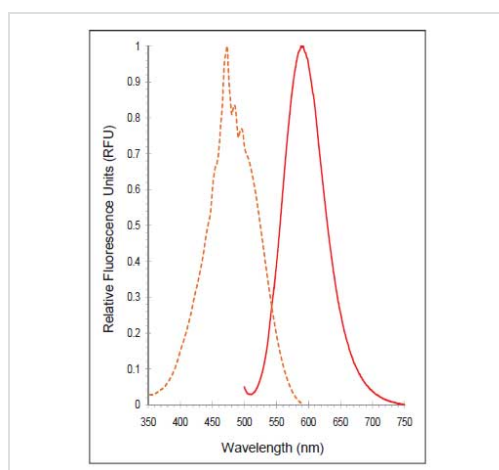
Dose response assay was performed with 0 to 3000 µM bacitracin added 15 min prior to the initiation of enzymatic reaction. Reactions were performed as described in Assay Protocol section. The fluorescence-based assay provides a vastly improved assay signal window and improved lower detection limit. In addition, the Z'-factor score obtained using the assay (0.91 for assay with and without P4HB) demonstrates excellent signal-to-noise and signal-to-background ratio



Intra-plate and inter-plate reproducibility using Inhibitor Control (bacitracin)

Dose response assay was performed with 0 to 3000  $\mu\text{M}$  bacitracin added 15 minutes prior to the initiation of enzymatic reaction.

Reactions were performed as described in Methods and Procedures section. Intra-plate and inter-plate CVs using the assay are typically 3-6%.



Absorption and fluorescence emission spectra for P4HB (PDIA1) Detection Reagent.

All spectra were determined in PBE Buffer. Example data only.

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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