Tribioscience

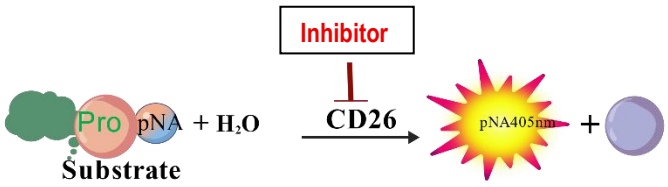
CD26 (DPP4) Inhibitor Screening Colorimetric Assay Catalog: TBS2104
(100 Assays, Store at -20°C)

DESCRIPTION

CD 26 (dipeptidyl peptidase-4/DPP4) is a transmembrane glycoprotein expressed on many cell types, including immune cells, epithelial cells, and endothelial cells. Its enzymatic activity: cleaves dipeptides from the N-terminus of polypeptides when the second amino acid is proline or alanine. It is used to manage and reduce inflammation and is related to some diseases, such as Alzheimer's disease, diabetes, and cancer.

Tribioscience CD26 Activity Colorimetric Assay provides a simple and sensitive method for monitoring CD26 activity in biological samples (tissue, cells, serum, urine, stool). This assay uses a synthetic p-nitroaniline derivative (R-*p*NA) as its substrate. CD26 can specifically cleave the substrate to release *p*NA, a chromophore which can be measured at absorbance (OD 405 nm) as shown in **Fig. 1.**

Fig. 1 Assay Principal Diagram



Synonyms: Dipeptidyl peptidase IV; ADABP/ADCP2, DPP4; TP103.

APPLICATIONS

This kit is used for CD26 inhibitor screening.

KEY FEATURES

Flexible: can be used for 96 wells and 384 wells plate
Simple: Just one-step: add-incubate-read model
Time saving: a 30-minute reaction at 37˚C

KIT CONTENTS

Component	100 Assays
CD26 Substrate	0.18 mL
Inhibitor	50 µL
Enzyme Mix	12 µL
Assay Buffer	12 ml

STORAGE CONDITIONS

The kit is shipped on ice and should be stored at -20°C for long-term storage. Shelf life of 12 months after receipt.

PROCEDURES

1. Equilibrate all the kit components until room temperature before starting the experiment.
2. Prepare the screening test compounds (test inhibitors): Dissolve the test compounds in an appropriate solvent, further diluting the compounds in the CD26 assay buffer. The effect of the solvent on the CD26 activity should be considered by including solvent control to the assay. Add 10 µL of the different diluted test compounds to the microplate. Add all components as shown in Table 1.

Table.1: Assay preparation

	Test Inhibitor	Inhibitor Control	Enzyme Control	Blank Control	Solvent Control
Test Inhibitor	10 µL				
Diluted Inhibitor control		10 µL			
Assay Buffer			10 µL	20 µL	
Solvent Control					10 µL

3. Prepare Enzyme working solution: Add 10 µL of the Enzyme stock to 990 µL of the CD26 assay buffer for 100 wells. Please adjust the volume as needed (Note: The diluted Enzyme control working solution is fresh use only, cannot be stored for future use).
4. Add 10 µL enzyme working solution to all wells except of blank control (already add assay buffer as table.1).
5. Dilute substrate stock with assay buffer for 50 times. For 100 well plate, 7.84 mL Assay buffer + 0.16mL substrate stock (50x). Mix well gently.
6. Add 80 µL of the substrate solution to all wells. Tap plate briefly to mix.
7. Incubate at 37°C for 30-60 minutes.
8. Read plate at OD 405nm in the endpoint mode.

Calculating CD26 inhibitor screening

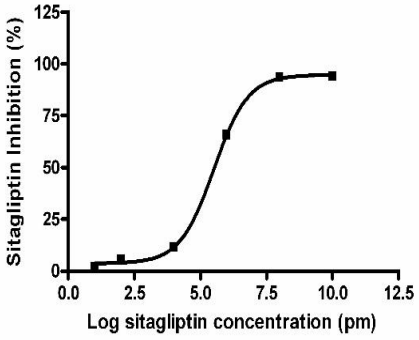
Subtract the Blank Control reading from all readings (Compound, Enzyme Control, and Inhibitor Control). Set the ΔOD of Enzyme Control (EC) as 100%.

% of Inhibition = 100* [ΔOD (EC)- ΔOD(TC)] /ΔOD(EC)

Here EC=Enzyme Control; TC= Test Compound.

Typical data displayed on Fig.2 as a reference for the assay.

Fig.2: Sitagliptin Inhibition of CD26



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RELATED PRODUCTS

Caspase-3 Fluorometric Assay kit (TBS3230)
Tryptase Activity Assay (TBS2101)
Cytochrome C Oxidase Activity Assay (TBS2115)
Fast Glucose Determination Colorimetric/Fluorometric Assay (TBS2087)
Glucose Oxidase Activity Colorimetric/Fluorometric Assay (TBS2088)
Non-esterified Fatty Acid Assy (TBS2203)
Glycerol Colorimetric / Fluorometric Assy (TBS2204)
Protein Assay Kits (TBS2005)
Cell Nuclear Extract kit (TBS6025)

Research use only.