Gly-Pro-AMC *CAS 115035-46-6*

Catalog number: 13450
Unit size: 5 mg

Component	Storage	Amount
Gly-Pro-AMC *CAS 115035-46-6*	Freeze (<-15 °C), Minimize light exposure	5 mg

OVERVIEW

Gly-Pro-AMC is a sensitive fluorogenic substrate for detecting dipeptidyl peptidase IV (DPPIV), a serine protease that cleaves N-terminal dipeptides from polypeptides with L-proline or L-alanine at the penultimate position. DPPIV is a multifunctional protein expressed on the surface of several cell types including epithelial, endothelial and lymphoid cells. It is identical to the T cell activation antigen CD26 and the adenosine deaminase binding protein, and it is also released as a soluble form in plasma. The substrates of CD26/DPPIV include a wide variety of proline-containing peptides such as growth factors, chemokines, neuropeptides, and vasoactive peptides. DPPIV is involved in immune regulation, signal transduction, and apoptosis, and appears to play an important role in tumor progression. Importantly, DPPIV is a therapeutic target for type II diabetes due to its role as a serum protease that cleaves incretin hormones of the glucagon family of peptides and thus regulates glucose homeostasis. Studies indicate that a DPPIV inhibitor improves impaired glucose tolerance. The assay of DPPIV with Gly-Pro-AMC is a simple, fast and flexible assay that is ideal for high-through put screening. The assay demonstrates improved sensitivity compared to colorimetric DPPIV assays, allowing the researcher to use less enzyme while still achieving appropriate Z' values. The homogeneous coupled-enzyme format is convenient, requiring only a single reagent addition to the test samples. Maximal sensitivity is achieved in 20-30 minutes, and the signal generated is very stable.

AT A GLANCE

Important The following protocol is recommended procedure, the optimum conditions must be determined experimentally for each test.

KEY PARAMETERS

Instrument:	Fluorescence microplate reader
Excitation:	380 nm
Emission:	500 nm
Cutoff:	435 nm
Recommended plate:	Solid black

PREPARATION OF STOCK SOLUTIONS

Unless otherwise noted, all unused stock solutions should be divided into single-use aliquots and stored at -20 °C after preparation. Avoid repeated freeze-thaw cycles.

1. Gly-Pro-AMC stock solution:
Make a 10 to 25 mM Gly-Pro-AMC stock solution in DMSO. Keep from light.

Note The stock solution should be used promptly; any unused solution should be aliquoted and frozen at -20 ° C.

Note Avoid repeated freeze-thaw cycles and keep from light.

PREPARATION OF WORKING SOLUTION

Gly-Pro-AMC working solution:
Take one vial out, and make 50 to 100 µM assay solution by diluting the stock solution with an assay buffer of your choice (such as 50 mM Tris-HCl, pH 7.5, 1.0 mM DTT).

SAMPLE EXPERIMENTAL PROTOCOL

1. Mix equal volume of the DPPIV standards or samples with the assay solution, and incubate at room temperature for at least 1 hour.
2. Monitor the fluorescence increase at Ex/Em = 380/500 nm.

EXAMPLE DATA ANALYSIS AND FIGURES

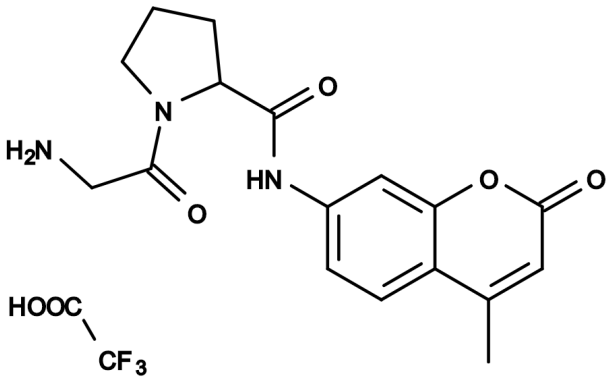

Figure 1. Chemical structure for Gly-Pro-AMC *CAS 115035-46-6*

DISCLAIMER