



Modelling – Cathepsin D activity test

Aim

BioVision's Cathepsin-D activity detection kit is a fluorescence-based assay, using MCA-labeled cathepsin-D preferred substrate sequence GKPILFFRLK (Dnp) -D-R-NH2. After digestion of the substrate by cathepsin-D enzymes contained in cell lysates or other samples, the product may release fluorescence, which can be easily quantitatively detected by fluorometer or fluorescence microtitrator reader. This kit can test the activity of our proteins.

Materials

CD Reaction Buffer

CD Substrate

Procedure

Based on the instructions in the Cathepsin D Activity Fluorometric Assay

Kit, we designed the relevant experiments as follows:

- 1. Add 180ul CD Reaction Buffer into 15 wells(A1-A4 B1-B4, C1-C4) in a 96-well plate.
- 2. Add 0 (A1-C1), 2(A2-C2), 4 (A3-C3), 6 (A4-C4), 8 (A5-C5) of the 1mM CD Substrate into plate.





- 3. Add 20ul cathepsin D protein into each well.
- 4. Read sample in a fluorometer equipped with a 328-nm excitation filter and 460-nm emission filter. Measure every few seconds. The interval depends on the reaction rate.

