



Product Information

N-Succinyl-Ala-Ala-Pro-Phe 7-Amido-4-trifluoromethylcoumarin

Product Number **S 5189**

Storage Temperature -20°C

Product Description

Molecular formula: $\text{C}_{34}\text{H}_{36}\text{F}_3\text{N}_5\text{O}_9$

Mol. wt.: 715.7

N-Succinyl-Ala-Ala-Pro-Phe 7-amido-4-trifluoromethylcoumarin (Suc-AAPF-AFC) is a fluorogenic substrate suitable for the assay of chymotrypsin and cathepsin G.

When Suc-AAPF-AFC is hydrolyzed, the free AFC produced in the reaction can be quantified by fluorometric detection (excitation 400 nm, emission 505 nm) or by spectrophotometric detection at 380 nm (extinction coefficient = 12,600 at pH 7.2). When used in an enzyme assay with fluorescence detection, AFC has higher sensitivity than 4-methoxy-2-naphthylamide (MNA).^{1,2}

Chymotrypsin is a serine protease with preferential cleavage at Tyr-, Trp-, Phe-, and Leu- residues. Cathepsin G, a serine protease first discovered in human neutrophil leukocytes, has a specificity similar to that of chymotrypsin.^{3,4} Suc-AAPF-AFC may also be suitable for other serine proteases with the same peptide recognition sequence.

Preparation Instructions

Prepare stock 20 mM solutions in dry DMSO.

Storage/Stability

Store at -20°C . Product is stable for at least one year, if stored as recommended.

Store stock solutions in frozen aliquots at -20°C . Allow the material to warm to room temperature before use to ensure stability.

References

1. Lojda, Z., The use of substrates with 7-amino-3-trifluoromethylcoumarine (AFC) leaving group in the localization of protease activities *in situ*. *Acta Histochem.*, **98**, 215-228 (1996).
2. Johansen, H. T., et al., Colorimetric and fluorimetric microplate assays for legumain and a staining reaction for detection of the enzyme after electrophoresis. *Anal. Biochem.*, **273**, 278-283 (1999).
3. Barrett, A.J., and McDonald, J. K., "Mammalian Proteases. A Glossary and Bibliography," pp. 182-187. Academic Press, New York, 1980.
4. Barrett, A.J., *Method in Enzymology* **80**, part C 561-565 (1981).

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