IMMBIOMED

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SPECTROZYME® FVIIa Chromogenic substrate

Product No. ADG217L

Description

Chromogenic substrate for amidolytic assay of Factor VIIa and lipidated Tissue Factor/VIIa complex.

Chemistry

Formula: CH₂SO₂-D-CHA-But-Arg-pNA.AcOH

Chemical name: Methanesulphonyl-D-cyclohexylalanyl-butyl-arginine-paranitroaniline monoacetate

salt

MW: 670.8 Dalton

Solubility: > 10 mM in distilled/deionized water.

> 0.5 mM in Tris Buffer, pH 8.4

Suggested Use

SPECTROZYME® FVIIa, it is the substrate of choice in all chromogenic based methods for the determination of factor VIIa, and for the factor VIIa/Tissue Factor complex in particular.

Assay Conditions/SubstrateKinetics

Enzyme activity is determined by measuring the increase in absorbance of the free chromophore (pNA) generated, in comparison to the original substrate, per unit time at λ405 nm. At excess substrate concentration, the rate of absorbance increase due to the amount of chromophore released is linearly related enzyme concentration. Measurement can be made either through acid quenching of the reaction (end-point method), or through use of a kinetic recording spectrophotometer (initial-rate-method).

Kinetic Data for Factor VIIa and TF complex

Buffer: 880 µL Tris-Buffer, pH 8.4 Temp. 37°C (0.1M Tris. 0.1M NaCl. 5 mM CaCl₂. 0.1% BSA)

Substrate: 0.100 mL of No. ADG217 at 4 mM

Enzyme:

- human factor VIIa 0.020 mL of FVIIa at 0.025 mg/mL

 $=> K_{M}: 5.0 \text{ mM}$ V_{max}: 6.72 nmoles/min

- By adding Tissue Factor with a 10:1 ratio (TF:FVIIa) 0.100 mL of FVIIa at 0.025 mg/mL +

0.100 mL of tissue factor at 0.25 mg/mL V_{max}: 69.7 nmoles/min $=> K_{M}: 0.97 \text{ mM}$

Warnings and Precautions

Contains: para-nitroanilide acetate salt

Hazard pictograms



Signal word

Hazard statements

H315 Causes skin irritation.

H319 Causes serious eye irritation.

H335 May cause respiratory irritation.

Precautionary statements

P261 Avoid breathing dust.

P280 - Wear protective gloves.

P312 - Call a POISON CENTER/doctor if you feel unwell.

P332 + P313 - If skin irritation occurs: Get medical advice/attention.

P337 + P313 - If eye irritation persists: Get medical advice/attention.

P501 - Dispose of contents/container in accordance with local/regional/ national/international regulations.

Presentation

Glass vial containing 50 µmoles of lyophilized substrate.

Reconstitution

Reconstitute with filtered distilled/deionized water to create a stock solution with a concentration of 5.0 mM. The typical working range concentration is 4.0 mM.

Storage

Store intact vials in the dark at 2 - 8 °C for up to 5 years from the date of manufacture. Reconsituted substrate may be stored for 1 week at room temperature, 2 months at 2° - 8°C, or for up to 6 months at -20°C (Aliquot and freeze. Do not submit to freeze-thaw cycles).

Distributed by



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