IMMBIOMED

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SPECTROZYME® FXIIa Chromogenic substrate

Product No. ADG312

Description

Highly sensitive chromogenic peptide substrate for factor XIIa. Determination of factor XIIa activity for research, inprocess and quality control.

Chemistry

Formula: H-D-CHA-Gly-Arg-pNA · 2AcOH

MW: 624.7 Dalton

Purity: < 0.5% free para-nitroaniline

Biochemical Characteristics:

 $k_{cat} = 4.4/sec$ $V_{max} = 3.14 \mu mol/min$

Assay Conditions

Enzyme activity is determined by measuring the increase in absorbance of the free chromophore (pNA) generated per unit time at $\lambda 405$ nm. At excess substrate concentrations, the rate at which the absorbance increases due to the amount of chromophore released is linearly related to 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).

Material required but not provided:

Buffer, NaCl, reference material, KALLISTOPTM, FXII activator, dist. H_2O .

Buffer:

50 mM Tris-imidazole buffer, 150 mM NaCl, pH 7.9

Assay 1: Suggested protocol for the determination of factor XIIa activity:

0.700 ml buffer

0.100 ml factor XIIa (0.2 units/ml)

0.200 ml SPECTROZYME® FXIIa (4 mM)

=> Determination of ΔOD/min at 405 nm

Assay 2: Suggested protocol for the determination of factor XIIa activity (FXII activated by Kalliplastin, Dextran, or ACTIN) using SPECTROZYME® FXIIa and a selective synthetic inhibitor of plasma kallikrein (KALLISTOPTM):

0.100 ml citrated plasma (diluted 1:5 with NaCl)

0.200 ml Kalliplastin (20 µg/ml)

- incubate for 1 min at 37 °C

0.500 ml buffer

0.100 ml KALLISTOP™ (0.2 mM, ADG503)

0.100 ml SPECTROZYME® FXIIa (2 mM)

=> Determination of ΔOD/min at 405 nm

Warnings and Precautions

Contains: para-nitroanilide diacetate salt

Hazard pictograms



Signal word

Warning

Hazard statements

H315 - Causes skin irritation. H319 - Causes serious eye irritation. H335 - May cause respiratory irritation.

Precautionary statements

P261 Avoid breathing dust.

P264 Wash thoroughly after handling.

P280 Wear protective gloves, wear eye/face protection. P302+P352 IF ON SKIN: Wash with plenty of water.

P304+P340 IF INHALED: Remove person to fresh air and keep comfortable for breathing.

P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

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.

P362+P364 Take off contaminated clothing and wash it before reuse

Presentation

Amber glass vial containing 10 µmoles of lyophilized substrate.

Reconstitution

Reconstitute with filtered deionized water to create a 10 mM solution. Shake gently before use.

Storage

May be used by the expiry date given on the label when stored unopened, protected from moisture, in the dark, 2-8°C. Avoid contamination of the reagents by microorganisms.

Shipment of product does not require cooling during the time of transportation.

References

A new assay for the determination of factor XII in plasma using a chromogenic substrate and a selective inhibitor of plasma kallikrein. Stürzebecher J et al., Thromb Res 1989; 55: 709-15.





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