



SPECTROZYME® FXIIa
Chromogenic substrate

Product No. ADG312

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Description

Highly sensitive chromogenic peptide substrate for factor XIIa. Determination of factor XIIa activity for research, in-process 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 μ mol/min

Assay Conditions

Enzyme activity is determined by measuring the increase in absorbance of the free chromophore (pNA) generated per unit time at λ 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, dist. H₂O, KALLISTOP™, FXII activator

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

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 Δ OD/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 (KALLISTOP™):

0.100 ml citrated plasma (diluted 1:5 with NaCl)
0.200 ml Kalliplastin (20 μ g/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 Δ OD/min at 405 nm

Presentation

ADG213 - amber glass vials containing 10 μ moles 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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