## PENTAPHARM

## Pefachrome® TH 8198 (corresp. S-2238)

Application: Chromogenic peptide substrate for the determination of thrombin and antithrombin III

Formula: H-D-Phe-Pip-Arg-pNA · 2HCl

**Principle:** H-D-Phe-Pip-Arg-pNA + E ==> H-D-Phe-Pip-Arg-OH + pNA + E

E = Enzyme

 $K_M$ : 7 μM (human thrombin)  $V = 1.7 \cdot 10^{-7}$  mol/min · NIH-U  $K_M$ :  $V = 2.2 \cdot 10^{-7}$  mol/min · NIH-U

**Solubility:**  $> 10 \text{ mM in H}_2\text{O}$  **MW:** 625.6

**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 micro-organisms.

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

## Material required but not provided:

Buffer, α-thrombin (3 NIH units/ml in 300 mM NaCl)

**Buffer:** 50 mM Tris-Imidazole pH 8.4, 300 mM NaCl

**Assay 1:** Suggested protocol for the determination of **thrombin** activity:

1 0.730 ml buffer 0.070 ml α-thrombin

0.200 ml Pefachrome®TH 8198, 4 mM in dist.H2O

=> Determination of ΔOD/min at 405 nm

Assay 2: Suggested protocol for the determination of antithrombin III in citrated plasma:

Inactivation of thrombin by plasma AT III

1.000 ml 4 NIH unit/ml thrombin and 10 USP unit/ml heparin / ml buffer

0.010 ml human citrated plasma => incubate for 4 min at 37° C

Assay of residual thrombin activity

1.700 ml buffer

0.100 ml solution step 1

0.200 ml Pefachrome®TH 8198, 2 mM in dist.H2O

=> Determination of  $\Delta$ OD/2 min at 405 nm

Package size: Vial containing 25mg Pefachrome® TH 8198 and 120 mg mannitol Code: 081-66

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