

# Pefachrome<sup>®</sup>TG

**Application:** Pefachrome<sup>®</sup>TG is a chromogenic peptide substrate that is specifically cleaved by thrombin but at a slow rate. For the continuous registration of thrombin formation in plasma, such a selective thrombin substrate is required, that shows moderate binding affinities (high  $K_m$ ) and a low turnover rate ( $k_{cat}$ ).

**Formula:** H- $\beta$ -Ala-Gly-Arg-pNA·2AcOH **MW:** 542.6

**Principle:** H- $\beta$ -Ala-Gly-Arg -pNA + E ==> H- $\beta$ -Ala-Gly-Arg -OH + pNA + E  
E = Enzyme (Thrombin)

**$K_M$**  1.95 mM  **$k_{cat}$ :** 1.91 s<sup>-1</sup>

**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.

**Stability after reconstitution:**

+ 2 to + 8°C	1 week
-20°C	1 month

**Material required but not provided:**

Reference material, buffer, Pefabloc<sup>®</sup>FG, Innovin<sup>®</sup> or DAPTIN<sup>®</sup>

**Buffer:** 50 mM Tris-HCl pH 7.4 , 100 mM NaCl, 0.5% HSA

Suggested protocols for the determination of thrombin generation in human plasma:

**Macro assay:**

0.600 ml platelet-free human plasma 0.075 ml buffer 0.075 ml Pefabloc <sup>®</sup> FG 36 mg/ml (Pentapharm Ltd.) 0.090 ml Pefachrome <sup>®</sup> TG (5 mM) => incubate at 37 °C 60 $\mu$ l Innovin <sup>®</sup> in 250 mM CaCl <sub>2</sub> or 60 $\mu$ l DAPTIN <sup>®</sup> in 250 mM CaCl <sub>2</sub> => Measure release of pNA at 405 nm , 15 minutes
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**Micro assay:**

0.200 ml platelet-free human plasma 0.050 ml Pefabloc <sup>®</sup> FG 18 mg/ml in buffer 0.030 ml Pefachrome <sup>®</sup> TG (5 mM) => incubate at 37 °C 20 $\mu$ l Innovin <sup>®</sup> in 250 mM CaCl <sub>2</sub> or 20 $\mu$ l DAPTIN <sup>®</sup> in 250 mM CaCl <sub>2</sub> => Measure release of pNA at 405 nm , 15 minutes
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**References:** Prasa D, Svendsen L, Stürzebecher J. The ability of thrombin inhibitors to reduce the thrombin activity generated in plasma on extrinsic and intrinsic activation. Thromb and Haemost 1997; 77: 498-503.

Prasa D, Svendsen L, Stürzebecher J. Inhibition of Thrombin Generation in Plasma by Inhibitors of Factor Xa. Thromb and Haemost 1997; 78: 1215-1220.

**Package size:** Vial containing 10  $\mu$ mol **Code:** 081-40  
Bulk [g] 081-17

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