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For Research Use Only. Not for use in diagnostic procedures.



# Anti-PS/PT (Human) mAb

CODE No.	D350-3
CLONALITY	Monoclonal
CLONE	231D
ISOTYPE	Mouse IgG1 κ
QUANTITY	100 µL, 1 mg/mL
SOURCE	Purified IgG from hybridoma supernatant
IMMUNOGEN	Human prothrombin
FORMULATION	PBS containing 50% Glycerol (pH 7.2). No preservative is contained.
STORAGE	This antibody solution is stable for one year from the date of purchase when stored at -20°C.

### **APPLICATIONS-CONFIRMED**

ELISA	1:10,201
LA test	6.25-50 μg/mL (dRVVT)

## SPECIES CROSS REACTIVITY on LA

Species	Human	Mouse	Rat	Hamster
Sample	Plasma	Not tested	Not tested	Not tested
Reactivity	+			

Entrez Gene ID 2147 (Human)

**REFERENCE** 1) Sakai, Y., *et al.*, *Arthritis Rheum.* **60**, 2457-2467 (2009)

For more information, please visit our web site https://ivd.mbl.co.jp/

The descriptions of the following protocols are examples. Each user should determine the appropriate condition.

# <u>ELISA</u>

- Add 100 μL/well of Anti-PS/PT (Human) mAb (MBL; code no. D350-3) diluted with Assay diluent\* as suggested in the APPLICATIONS to a PS/PT-coated micro plate\*. Incubate for 1 hr. at room temperature. (The concentration of antibody will depend on the conditions.)
- 2) Wash the plate with Wash buffer\* (4 times).
- 3) Add 100 µL/well of Anti-IgG (H+L chain) (Mouse) pAb-HRP (MBL; code no. 330). Incubate for 1 hr. at room temperature.
- 4) Wash the plate with Wash buffer\* (4 times).
- 5) Add 100 µL/well of substrate solution (ex. TMB). Incubate for 30 min. at room temperature.
- 6) Add 100  $\mu L/well$  of stop solution (ex. 0.5 M  $\rm H_2SO_4).$
- 7) Read absorbance at 450 nm and the reference at 620 nm.
- \* Components of PS/PT ELISA Kit (MBL; code no. 7803)

## Experimental example

	Optical Density					
Dilution factor	Anti-PS/PT mAb (D350-3)			Mouse IgG1 (isotype control) (MBL; code no. M075-3)		
	1	2	Average	1	2	Average
1*	2.226	2.180	2.203	0.009	0.007	0.008
1/2	1.541	1.490	1.516	0.001	0.001	0.001
1/4	0.891	0.825	0.858	0.001	0.004	0.003
1/8	0.539	0.509	0.524	0.007	0.006	0.007
1/16	0.266	0.249	0.258	0.003	0.002	0.003
1/32	0.134	0.116	0.125	0.005	0.006	0.006
1/64	0.059	0.056	0.058	0.003	0.004	0.004
1/128	0.027	0.027	0.027	0.003	0.001	0.002

Blank	0.007	0.002	0.005

\* Dilution factor 1; 1/10,201 (1/101 x 1/101)

# LA test (dRVVT)

Preparation of plasma

- 1) Mix fresh whole blood and 120 mM trisodium citrate at the ratio of 9:1 in a plastic tube.
- 2) Centrifuge the tube at 1,500 x g for 15 min. and transfer the supernatant to another tube.

#### dRVVT assay

- 1) Heat Reagent 1\* to  $37^{\circ}C \pm 1^{\circ}C$ .
- 2) Add Anti-PS/PT (Human) mAb (MBL; code no. D350-3) to pooled normal human plasma (prepared described above) as suggested in the **APPLICATIONS**, and heat for 1 min. at 37°C±1°C in a glass tube.
- 3) Mix 200 µL of the reagent (prepared from step 1) and 200 µL of the plasma added antibody (prepared from step 2) in a test tube.

4) Measure the clotting time.

- 5) Repeat step 1) 4) using Reagent 2\* in place of Reagent 1\*.
- \* Components of LAtest "Gradipore" (MBL; code no. 4150)

#### **Experimental example** (pooled normal human plasma)

Final antibody concentration (μg/mL)	<a> Clotting time (sec.) Reagent 1</a>	<b> Clotting time (sec.) Reagent 2</b>	Ratio (A/B)
0	36.0	33.5	1.07
6.25	39.5	34.5	1.14
12.5	44.7	35.0	1.28
25.0	54.5	36.9	1.48
50.0	70.7	41.7	1.70