

COULTER CLONE®

Pit-1-FITC

	Pit-1-FITC
Specificity	CD41
Clone	69 (Pit-1) ¹
Hybridoma	NS-1 x BALB/c
Immunogen	Adherent mononuclear cells
Ig Chain	IgM
Species	Mouse
Source	Ascites fluid or Conditioned Medium
Purification	Gel filtration chromatography
Fluorescence	FITC (Green): Excites at 468-509 nm Emits at 504-541 nm
Conjugation	FITC (Fluorescein Isothiocyanate)
Molar Ratio	FITC/Protein 20-30

REF 6602993 - 100 tests

PN 4235540-H



ANALYTE SPECIFIC REAGENT

Analytical and performance characteristics are not established.

ANTIBODY SPECIFICITY

The Pit-1 antibody identifies the CD41 antigen,¹ which is the integrin $\alpha IIb\beta$ of the $gpIIb/IIIa$ complex on the surface of platelets. In association with CD61, CD41 is the receptor for fibrinogen, fibronectin, von Willebrand factor and thrombospondin.² The molecular weight of CD41 is 140 kD.² Among hematopoietic elements, the Pit-1 antigen $Ib/IIIa$ is found on human peripheral blood platelets and bone marrow megakaryocytes.³ It is not expressed by lymphoid cells (T, B or null [NK, LGL]), monocytes, neutrophils or erythrocytes.³

REAGENT

See table above.

REAGENT CONTENTS

The antibody concentration is 5.0 μ g/test. The final concentration of nonantibody reagent when reconstituted is 0.2% gelatin, 0.01 M potassium phosphate, 0.15 M NaCl and 0.1% Na₂S₂O₃.

STATEMENT OF WARNINGS

1. This reagent contains 0.1% sodium azide. Sodium azide under acid conditions yields hydrazoic acid, an extremely toxic compound. Azide compounds should be flushed with running water while being discarded. These precautions are recommended to avoid deposits in metal piping in which explosive conditions can develop. If skin or eye contact occurs, wash excessively with water.
2. Specimens, samples and all material coming in contact with them should be handled as if capable of transmitting infection and disposed of with proper precautions.
3. Never pipet by mouth and avoid contact of samples with skin and mucous membranes.
4. Do not use reagent beyond the expiration date on the vial label.
5. Minimize exposure of reagent to light during storage or incubation.
6. Avoid microbial contamination of reagent or erroneous results may occur.
7. Use Good Laboratory Practices (GLP) when handling this reagent.
8. Harmful if swallowed.
9. After contact with skin, wash immediately with plenty of water.

STORAGE CONDITIONS AND STABILITY

Unreconstituted lyophilized reagent is stable to the expiration date on the vial label when stored at 2-8°C. Do not freeze. Minimize exposure to light.

The stock solution of reconstituted lyophilized reagent is stable as follows:

- 6 months when stored at 2-8°C or 0 to -20°C when reconstituted using the Reconstitution Procedure described in the Reagent Preparation section. If all of a reconstituted reagent is not to be used within 6 months, follow the Freezing Procedure.
- 1 year when stored at -70°C using the Freezing Procedure.

Freezing Procedure

Materials Required But Not Supplied:

PBS - Phosphate Buffered Saline (pH=7.2) PN 6603369
PBS containing 2% heat-inactivated fetal or newborn calf serum (FCS). Dilute 2 mL of calf serum to 100 mL with PBS.

1. Dilute the reconstituted stock solution of the COULTER CLONE reagent with PBS containing 2% FCS prior to freezing as follows:

Add 5 μ L of reconstituted stock solution (1 test*) to 100 μ L PBS with 2% FCS**.
*These may be frozen in multiple test volume aliquots.
**This yields 2X the concentration of the working solution.
2. Prior to use, allow the frozen aliquot to reach 20-25°C.
3. The frozen aliquot, at 2X the final concentration, must be further diluted to equal the total volume as calculated in the REAGENT PREPARATION section. Dilute each aliquot with the appropriate volume of PBS without 2% FCS and mix well.
4. Avoid repeated freeze/thaw cycles. This will denature the antibody protein.
5. Do not store in a self-defrosting freezer.

EVIDENCE OF DETERIORATION

Any change in the physical appearance of this reagent*, or any major variation in values obtained for control samples may indicate deterioration and the reagent should not be used. If the lyophilized material appears moist, do not use.

*Normal Appearance of Reagent

FITC labeled: Lyophilized-white to yellow-orange plug
Reconstituted-clear, colorless to yellow-green liquid

REAGENT PREPARATION

Reconstitute the lyophilized COULTER CLONE Pit-1-FITC reagent by adding 500 μ L of distilled water to the vial. This is the stock solution. Centrifuge the stock solution at 20-25°C at 100,000 x g for 10 minutes to optimize staining results. Use this liquid reagent directly from the vial as the stock solution to prepare the reagent working solution.

The reagent working solution* is prepared as follows (volume listed is on a per test basis):

Add 5 μ L stock solution to 195 μ L PBS**.

*Diluted reagent working solution is good for day of preparation only.

**PBS - Phosphate Buffered Saline (pH=7.2).

Bring reagent to 20-25°C prior to use.

USAGE

This reagent is for use with standard flow cytometry and/or fluorescence microscopy methodologies.

The use of Pit-1-FITC in this reagent is not intended for enumeration of CD41 cells in clinical diagnostic applications.

SELECTED RESEARCH REFERENCES

1. Schlossman SF, Boumsell L, Gilks W, Harlan JM, Silverstein R, Springer R, Tedder TF and Todd RF, eds: 1995. Leukocyte Typing V. Oxford University Press, Oxford UK.
2. Barclay AN, Birkeland ML, Brown MH, Beyers AD, Davis SJ, Somoza C, William AF, eds. 1993. The Leukocyte Antigens Facts Book. London: Academic Press.
3. Lawrence LB and Galnick HR. 1987. Monoclonal antibodies to the glycoprotein $Ib/IIIa$ epitopes involved in adhesive protein binding: Effects on platelet spreading and ultrastructure on human arterial subendothelium. J Lab Clin Med 109:495.


PRODUCT AVAILABILITY

COULTER CLONE Pit-1-FITC
PN 6602993 - 100 tests (0.5 mL)

For additional information or if damaged product is received, call Beckman Coulter Customer Service at 800-526-7694 (USA or Canada) or contact your local Beckman Coulter Representative.

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