Technical Data Sheet

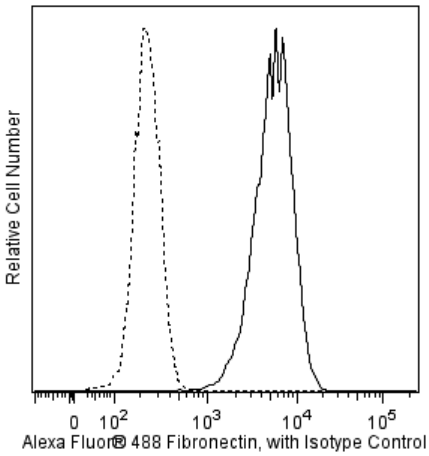
Alexa Fluor® 488 Mouse Anti-Fibronectin

Product Information

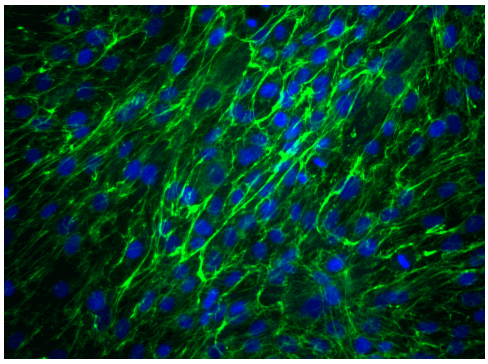
Material Number:	**563100**
Alternate Name:	FN; LETS
Size:	50 tests
Vol. per Test:	5 µl
Clone:	10/Fibronectin
Immunogen:	Human Fibronectin
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human Confirmed in Development: Mouse, Rat, Dog, Chicken, Bovine
Storage Buffer:	Aqueous buffered solution containing BSA, protein stabilizer, and ≤0.09% sodium azide.

Description

The 240-kDa dimeric fibronectin protein exists in two forms: a soluble protomer in body fluids and an insoluble multimer in the extracellular matrix. The latter is the primary functional form and creates a substrate for cell migration, a role which makes fibronectin vital to embryogenesis and wound response. Fibronectin mediates cytoskeletal organization, cell attachment, and cellular signaling through interactions with cellular receptors. Although various isoforms of fibronectin are derived by alternative splicing, they share a common N-terminus which is a critical region for cell surface binding in an initial step of multimer assembly. Further polymerization steps are regulated by fibronectin/integrin interactions and result in generation of the complex fibrils that constitute the fibronectin matrix.



Flow cytometric analysis of fibronectin in human mesenchymal stem cells (MSC). *MSC (Lonza), passage 6, were dissociated and fixed in BD Cytofix™ Fixation Buffer (Cat. No. 554655), and permeabilized with BD Phosflow™ Perm Buffer III (Cat. No. 558050). The cells were stained with either Alexa Fluor® 488 Mouse IgG1, κ isotype control (dashed line, Cat. No. 557782) or Alexa Fluor® 488 Anti-Fibronectin monoclonal antibody (solid line) at matched concentrations. Histograms were derived from gated events based on light scattering characteristics of MSC. Flow cytometry was performed on a BD LSRFortessa™ II flow cytometry system. BD Phosflow™ Perm/Wash Buffer I (Cat. No. 557885) is also suitable for permeabilization.*



Immunofluorescent analysis of fibronectin in human mesenchymal stem cells (MSC). *MSC (Lonza), passage 6, were fixed in BD Cytofix™ Fixation Buffer (Cat. No. 554655), permeabilized with BD Phosflow™ Perm Buffer III (Cat. No. 558050), and stained with Alexa Fluor® 488 Mouse Anti-Fibronectin monoclonal antibody (pseudo-colored green) at 1.2 µg/ml. Cell nuclei were stained with DAPI (pseudo-colored blue). The images were captured on a BD Pathway™ 435 Cell Analyzer and merged using BD Attovision™Software. 0.1% Triton™ X-100 is also suitable for permeabilization.*

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated to Alexa Fluor® 488 under optimum conditions, and unreacted Alexa Fluor® 488 was removed.

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Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested
Immunofluorescence	Tested During Development
Bioimaging	Tested During Development

Suggested Companion Products

Catalog Number	Name	Size	Clone
554655	Fixation Buffer	100 ml	(none)
558050	Perm Buffer III	125 ml	(none)
557885	Perm/Wash Buffer I	125 ml	(none)
557782	Alexa Fluor® 488 Mouse IgG1 κ Isotype Control	50 tests	MOPC-21

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10^6 cells in a 100-µl experimental sample (a test).
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.
4. Alexa Fluor® 488 fluorochrome emission is collected at the same instrument settings as for fluorescein isothiocyanate (FITC).
5. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
6. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
7. Triton is a trademark of the Dow Chemical Company.
8. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
9. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
10. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
11. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Chen H, Mosher DF. Formation of sodium dodecyl sulfate-stable fibronectin multimers. Failure to detect products of thiol-disulfide exchange in cyanogen bromide or limited acid digests of stabilized matrix fibronectin. *J Biol Chem.* 1996; 271(15):9084-9089. (Biology)

Danen EH, Sonneveld P, Brakebusch C, Fassler R, Sonnenberg A. The fibronectin-binding integrins alpha5beta1 and alphavbeta3 differentially modulate RhoA-GTP loading, organization of cell matrix adhesions, and fibronectin fibrillogenesis. *J Cell Biol.* 2002; 159(6):1071-1086. (Clone-specific: Immunofluorescence)

Rhee CS, Sen M, Lu D, et al. Wnt and frizzled receptors as potential targets for immunotherapy in head and neck squamous cell carcinomas. *Oncogene.* 2002; 21(42):6598-6605. (Clone-specific: Western blot)

Sechler JL, Takada Y, Schwarzbauer JE. Altered rate of fibronectin matrix assembly by deletion of the first type III repeats. *J Cell Biol.* 1996; 134(2):573-583. (Biology)

Zuk A, Bonventre JV, Brown D, Matlin KS. Polarity, integrin, and extracellular matrix dynamics in the postischemic rat kidney. *Am J Physiol.* 1998; 275(3):C711-C731. (Clone-specific: Immunohistochemistry)

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