

Technical Data Sheet

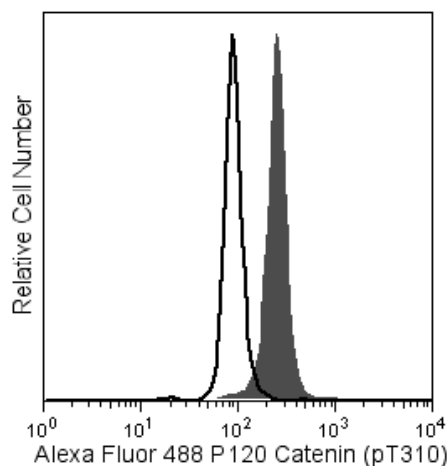
Alexa Fluor® 488 Mouse anti-p120 Catenin (pT310)**Product Information**

Material Number:	558667
Size:	50 tests
Vol. per Test:	20 µl
Clone:	22/p120 (pT310) (also known as cl.22)
Immunogen:	Phosphorylated Human p120 Catenin
Isotype:	Mouse IgG1, κ
Reactivity:	Confirmed by flow cytometry: Human Confirmed by western blot using purified antibody (Cat. No. 558203): dog, human, monkey, mouse
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The membrane-associated protein pp120 Src substrate (p120 catenin, p120cas) was identified as a tyrosine kinase substrate that is phosphorylated in Src-transformed cells. p120 catenin is localized to the E-Cadherin/catenin cell adhesion complex. Like β- and γ-catenin, p120 catenin directly associates with the cytoplasmic C terminus of E-Cadherin via its Arm domain. It exists as four isoforms that range in size from 90 to 115 kDa. Expression of these isoforms is heterogeneous in human carcinomas, suggesting that altered expression contributes to malignancy. Phosphorylation of multiple serine (S252, S268, S288, and S879) and threonine (T310 and T916) residues in p120 catenin may regulate its activity. The S879 residue is phosphorylated after PKC activation, while the S268 site is dephosphorylated after PKC activation. The latter residue is phosphorylated in vitro by p160 Rock. S252 and T310 residues are phosphorylated in vitro by GSK3b. Thus, p120 catenin function may be regulated in a complex manner through both serine and threonine phosphorylation.

The 22/p120 (pT310) monoclonal antibody recognizes the phosphorylated T310 in the regulatory domain of p120 catenin.



Analysis of p120 Catenin (pT310) in human vascular endothelium. After serum starvation overnight, EA-hy 926 cells (Edgell, McDonald, Graham, 1983) were either stimulated with 50 nM calyculin A (Calbiochem Cat. No. 208851, shaded histogram) for 30 minutes at 37°C or unstimulated (open histogram). The cells were fixed (BD Cytotfix™ buffer, Cat. No. 554655) for 10 minutes at 37°C, then permeabilized (BD™ Phosflow Perm Buffer III, Cat. No. 558050) on ice for at least 30 minutes, and then stained with Alexa Fluor® 488 Mouse anti-p120 Catenin (pT310). Flow cytometry was performed on a BD™ FACSCalibur flow cytometry system.

Preparation and Storage

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.

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

Application Notes**Application**

Intracellular staining (flow cytometry)	Routinely Tested
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Suggested Companion Products

<u>Catalog Number</u>	<u>Name</u>	<u>Size</u>	<u>Clone</u>
558050	Perm Buffer III	125 ml	(none)
554655	Fixation Buffer	100 ml	(none)

Product Notices

1. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.
2. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1×10^6 cells in a 100- μ l experimental sample (a test).
3. Alexa Fluor® 488 fluorochrome emission is collected at the same instrument settings as for fluorescein isothiocyanate (FITC).
4. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
5. 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.
6. 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.
7. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
8. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.

References

Edgell C-JS, McDonald CC, Graham JB. Permanent cell line expressing human factor VIII-related antigen established by hybridization. *Proc Natl Acad Sci U S A*. 1983; 80:3734-3737. (Biology: Controls)

Reynolds AB, Roczniak-Ferguson A. Emerging roles for p120-catenin in cell adhesion and cancer. *Oncogene*. 2004; 23:7947-7956. (Biology)

Xia X, Brooks J, Campos-Gonzalez R, Reynolds AB. Serine and threonine phospho-specific antibodies to p120-catenin. *Hybrid Hybridomics*. 2004; 23(6):343-351. (Immunogen)