Technical Data Sheet

Purified Rat anti-Human Lgr5 (Central LRR)

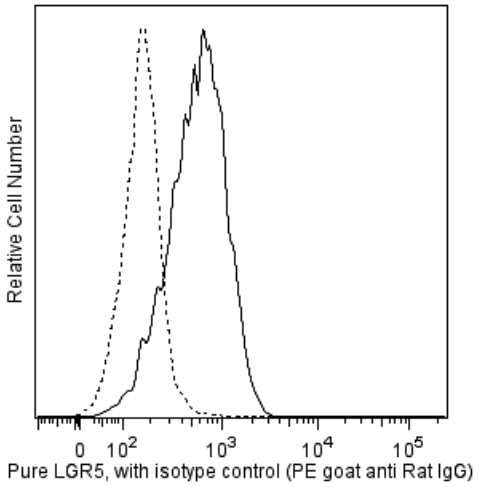
Product Information

Material Number:	**562732**
Alternate Name:	GPR49, GPR67, HG38
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	4D11F8 (also known as 4D11)
Immunogen:	Human LGR5 DNA
Isotype:	Rat IgG2b, λ
Reactivity:	QC Tested: Human Not Reactive: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

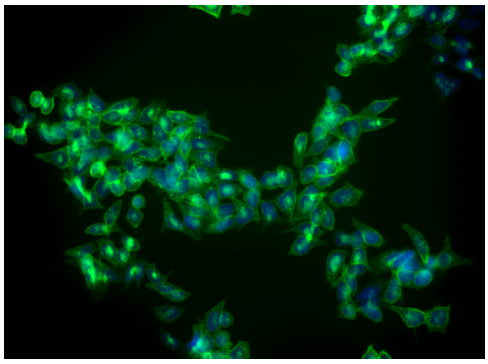
Description

Lgr5 (leucine-rich-repeat-containing G-protein-coupled receptor 5) is a seven transmembrane-domain receptor that is a target gene for Wnt and marks stem cells in the small intestine, colon, stomach, and hair follicle. Lgr5 was initially identified as a potential stem cell marker due to restricted expression of Lgr5 in the intestinal crypt and labeling of rapidly cycling cells of the colon and intestine. Using both lineage tracing and organoid culture experiments, Lgr5 positive cells are capable of generating all types of the small intestine epithelium hence indicating that Lgr5 marks stem cells of the small intestine and colon. R-spondin growth factors, which are secreted agonists of the Wnt pathway, bind Lgr5. The binding of R-spondins to Lgr5 leads to recruitment of the Frizzled/LRP Wnt receptor complex, which binds to Wnt ligands and leads to downstream Wnt signaling. Lgr5 is up-regulated in colon and ovarian cancers and has been implicated in promotion of tumor growth and metastasis.

The 4D11F8 monoclonal antibody recognizes an epitope in the center of the leucine-rich repeat (LRR) region of Human Lgr5.



Flow cytometric analysis of human LGR5. *Colorectal adenocarcinoma cells LS 174T (ATCC CL-188) were stained with either Purified Rat IgG2b isotype control (Cat. No. 553986) or Purified Rat anti-human LGR5 (Central LRR) monoclonal antibody (solid line) at matched concentrations. Cells were harvested with Accutase™ Cell Detachment Solution (Cat. No. 561527).The second step reagent was PE goat anti-Rat Ig (Cat. No. 550767). The histograms were derived from gated events based on light scattering characteristics of the LS 174T cells. Flow cytometry was performed on a BD FACSCanto™ II flow cytometry system.*



Immunoflourescent staining of human LGR5. *Colorectal adenocarcinoma cells LS 174T transfected with human LGR5 (Cells from Dr. Hans Clevers, Hubrecht Institute) were fixed and stained with Purified Rat anti-human LGR5 (Central LRR) monoclonal antibody (pseudo colored green) at 2.5 µg/mL. The second-step reagent was Alexa Fluor® 488 goat anti-rat Ig (Life Technologies) and counter-staining was with DAPI (pseudo-colored blue). The images were captured on a BD Pathway™ 435 Cell Analyzer and merged using BD Attovision™ Software.*

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Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at 4°C.

Application Notes

Application

Flow cytometry	Routinely Tested
Bioimaging	Tested During Development
Immunofluorescence	Tested During Development
Immunohistochemistry-formalin (antigen retrieval required)	Not Recommended
Western blot	Not Recommended
Immunohistochemistry-frozen	Not Recommended

Suggested Companion Products

Catalog Number	Name	Size	Clone
553986	Purified Rat IgG2b, κ Isotype Control	0.5 mg	A95-1
561527	Accutase™ Cell Detachment Solution	100 ml	(none)
550767	PE Goat Anti-Rat Ig	0.2 mg	Polyclonal
554656	Stain Buffer (FBS)	500 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
4. 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.
5. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.

References

Barker N, Huch M, Kujala P, et al. Lgr5(+ve) stem cells drive self-renewal in the stomach and build long-lived gastric units in vitro. *Cell Stem Cell.* 2010; 6(1):25-36. (Biology)

Barker N, van Es JH, Kuipers J, Kujala P, van den Born M, et al. Identification of stem cells in small intestine and colon by marker gene Lgr5. *Nature.* 2007; 449(7165):1003-1007. (Biology)

Carmon KS, Gong X, Lin Q, Thomas A, Liu Q. R-spondins function as ligands of the orphan receptors LGR4 and LGR5 to regulate Wnt/{beta}-catenin signaling. *Proc Natl Acad Sci U S A.* 2011; 108(28):11452-11457. (Biology)

de Lau W, Barker N, Low TY, et al. Lgr5 homologues associate with Wnt receptors and mediate R-spondin signalling. *Nature.* 2011; 476(7360):293-297. (Clone-specific)

Jaks V, Barker N, Kasper M, et al. Lgr5 marks cycling, yet long-lived, hair follicle stem cells. *Nat Genet.* 2008; 40(11):1291-1299. (Biology)

Merlos-Suárez A, Barriga FM, Jung P et al. The intestinal stem cell signature identifies colorectal cancer stem cells and predicts disease relapse. *Cell Stem Cell.* 2011; 8(5):511-524. (Biology)

Yui S, Nakamura T, Sato T, et al. Functional engraftment of colon epithelium expanded in vitro from a single adult Lgr5(+) stem cell. *Nat Med.* 2012; 18(4):618-623. (Biology)

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