

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Lgr5/GPR49 in ELISA. Stains human and mouse Lgr5 transfected cells but not irrelevant transfectants in Flow Cytometry and Immunocytochemistry. This antibody, also known as "RD42", has been found to detect an epitope in C-terminal LRR cap of LGR5 (Ref. 1).
Source	Monoclonal Mouse IgG _{2A} Clone # 707042
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Chinese hamster ovary cell line CHO-derived recombinant human Lgr5/GPR49 Met1-Ile560 Accession # O75473
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

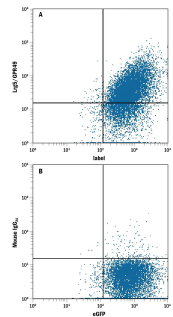
APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Flow Cytometry	2.5 µg/10 ⁶ cells	See Below
Immunocytochemistry	8-25 µg/mL	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	
Agonist Activity	Peng, W.C., <i>et al.</i> (2013) Cell Reports. 3:1885.	

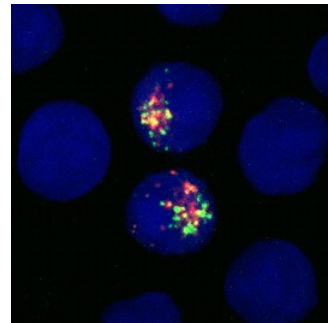
DATA

Flow Cytometry



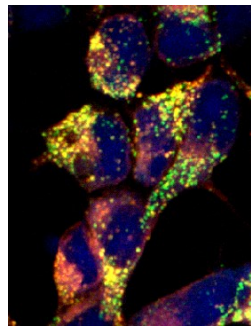
Detection of Lgr5/GPR49 in NSO Mouse Cell Line Transfected with Human Lgr5/GPR49 and eGFP by Flow Cytometry. NSO mouse myeloma cell line transfected with human Lgr5/GPR49 and eGFP was stained with either (A) Mouse Anti-Human Lgr5/GPR49 Monoclonal Antibody (Catalog # MAB8078) or (B) Mouse IgG_{2A} Isotype Control (Catalog # MAB003) followed by Allophycocyanin-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0101B).

Immunocytochemistry



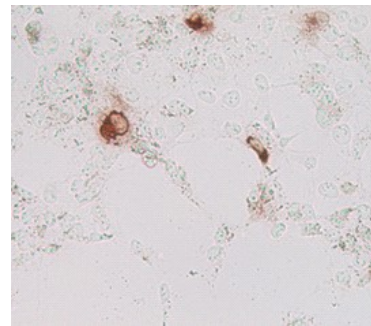
Lgr5/GPR49 in NSO Mouse Cell Line Transfected with Human Lgr5/GPR49. Lgr5/GPR49 was detected in immersion fixed NSO mouse myeloma cell line transfected with GFP (green) tagged human LGR5 using Mouse Anti-Human Lgr5/GPR49 Monoclonal Antibody (Catalog # MAB8078) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for [Fluorescent ICC Staining of Non-adherent Cells](#).

Immunocytochemistry



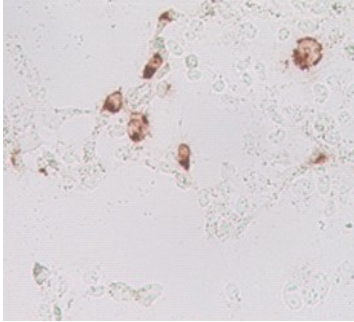
Lgr5/GPR49 in HEK293 Human Cell Line Transfected with Mouse Lgr5/GPR49. Lgr5/GPR49 was detected in immersion fixed HEK293 human embryonic kidney cell line transfected with GFP (green) tagged mouse LGR5 using Mouse Anti-Human Lgr5/GPR49 Monoclonal Antibody (Catalog # MAB8078) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cell surfaces and cytoplasm. View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

Immunocytochemistry



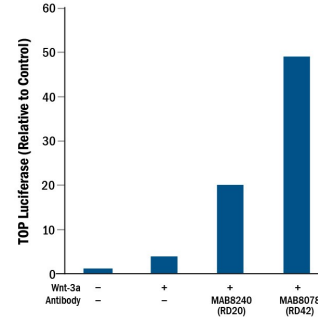
Lgr5/GPR49 in Human Cells Transfected with Human Lgr5/GPR49. Lgr5/GPR49 was detected in MYC tagged human transfectants using Mouse Anti-Human Lgr5/GPR49 Monoclonal Antibody (Catalog # MAB8078). Cells were stained using an anti-mouse HRP-conjugated secondary antibody (brown). Specific staining was localized to cytoplasm. *Image courtesy of Dr. Hans Clevers, Hubrecht Institute, The Netherlands.*

Immunocytochemistry



Lgr5/GPR49 in Mouse Cells Transfected with Mouse Lgr5/GPR49. Lgr5/GPR49 was detected in immersion fixed MYC tagged mouse transfectants using Mouse Anti-Human Lgr5/GPR49 Monoclonal Antibody (Catalog # MAB8078). Cells were stained using an anti-mouse HRP-conjugated secondary antibody (brown). Specific staining was localized to cytoplasm. *Image courtesy of Dr. Hans Clevers, Hubrecht Institute, The Netherlands.*

Agonist Activity



Human Lgr5/GPR49 Antibody Induces Activity. Mouse Anti-Human Lgr5/GPR49 Monoclonal Antibody (Catalog # MAB8078) induces TOPflash activity in the HEK293 human embryonic kidney cell line stably expressing LGR5 in the presence of Wnt-3a, but in the absence of R-Spondins. (*Data courtesy of Dr. Wim de Lau and Dr. Hans Clevers, Hubrecht Institute, The Netherlands. See Reference 1.*)

PREPARATION AND STORAGE

Reconstitution Reconstitute at 0.5 mg/mL in sterile PBS.

Shipping The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
*Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C

Stability & Storage Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Leucine-rich repeat G-protein-coupled receptor 5 (Lgr5), also called GPR49, is a 907 amino acid (aa), approximately 97 kDa (calculated), seven-transmembrane glycoprotein receptor in the Lgr family of cell surface receptors. The subfamily of Lgrs comprising Lgr4, Lgr5, and Lgr6 are G-protein-independent mediators of the potentiating effect of R-Spondins on Wnt signaling (2). Lgr5 binds and forms complexes with R-Spondins, Frizzled Wnt receptors and LRP Wnt co-receptors. The region of the human Lgr5 long extracellular domain used as an immunogen shares 90% amino acid sequence identity with mouse and rat Lgr5, respectively. Lgr5 is found on embryonic and adult epithelial stem cells (3). Lgr5⁺ stem cells can produce all epithelial cell types of the intestinal crypts (4). Abnormal LGR5 expression and regulation in stem cells might give rise to cancers such as intestinal, hepatocellular, pancreatic and ovarian carcinomas (5,6). This antibody has been referred to as "RD42" in Peng *et al.* (1).

References:

1. Peng *et al.* (2013) Cell Rep. **3(6)**:1885.
2. de Lau *et al.* (2014) Genes Dev. **28(4)**:305.
3. Barker *et al.* (2013) Development **140(12)**:2484.
4. Clevers (2013) Nature **495(7439)**:53.
5. Wu *et al.* (2014) Nat Commun **5**:3149.
6. Jang *et al.* (2013) PLoS One **8(12)**: e82390495.