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International Edition

AdipoGen®
Connecting Immunology to Metabolism™

Functional Antibodies

Blocking, Neutralizing, Activation & Depletion

Antibodies are highly specific, naturally evolved molecules that recognize and eliminate pathogenic and disease antigens. The typical antibody consists of two antigen-binding fragments (Fabs), which are linked via a flexible region (the hinge) to a constant Fc region. This structure comprises two pairs of polypeptide chains, each pair containing a heavy and a light chain of different sizes. The Fc portion of the Ig serves to bind various effector molecules of the immune system, as well as molecules that determine the biodistribution of the antibody.

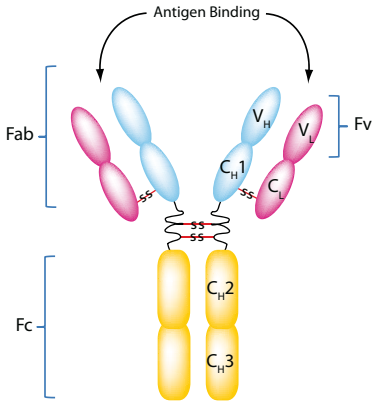
Antibodies are produced by: a) Injecting an antigen into mammals (mouse, rat, rabbit, goat, etc). Blood isolated from these animals contains **polyclonal antibodies** (multiple antibodies that bind to different epitopes of the same antigen), which are purified. b) Hybridoma technology generating **monoclonal antibodies** (epitope specific). Specific antibody-secreting lymphocytes are isolated from animals and immortalized by fusing them with a cancer cell line.

Monoclonal antibodies are routinely used in biochemistry, molecular biology, medical research and as therapeutic agents. Important advances have been made over the past decade to improve the specificity and efficacy of such antibodies by new engineering technologies, including **recombinant antibody technology**, such as antibody phage display (see page 8 for more information).

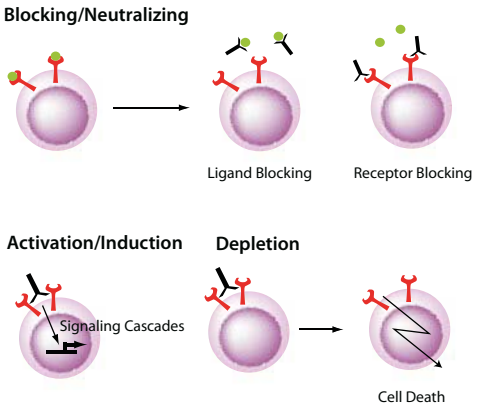
Functional Grade Antibodies (FuncAbs™):

Antibodies displaying an agonist or antagonist activity (functional grade antibodies (FuncAbs™)) are powerful tools for mimicking or blocking physiological functions *in vitro* and *in vivo*. **Functional grade antibodies** are available free of preservatives and tested for low endotoxin content and may be used for **activation, neutralizing or blocking studies, both *in vitro* or *in vivo*.**

General Antibody Schematic



Different Types of Antibody Functionality



Contents

AdipoGen®
Connecting Immunology to Metabolism™

Blocking/Neutralizing Antibodies [FuncAbs™]

anti-APRIL (mouse), mAb (rec.) (blocking) (Apry-1-1)

BULK

AG-27B-0001-C100		100 µg
AG-27B-0001PF-C100	Preservative Free	100 µg
AG-27B-0001B-C100	Biotin	100 µg

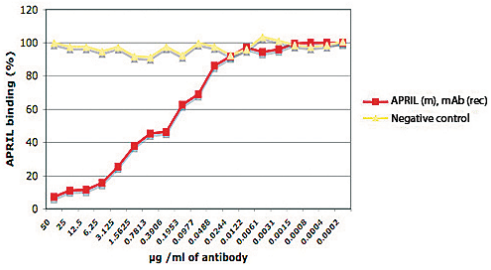
Clone: Apry-1-1

Isotype: Mouse IgG2bλ

Application: ELISA, IP, FUNC (Blocking)

Functional Application:
Inhibits binding of mouse APRIL to mouse BCMA and TACI.

LIT: Production of the plasma-cell survival factor APRIL peaks in myeloid precursor cells from human bone marrow: T. Matthes, et al.; Blood **118,** 1838 (2011)



anti-Angiopoietin-2 (human), mAb (rec.) (blocking) (Angy-1-4)

BULK

AG-27B-0015-C100		100 µg
AG-27B-0015PF	Preservative Free	100 µg | 500 µg | 1 mg

Clone: Angy-1-4

Isotype: Human IgG2λ

Application: ELISA, FUNC (Blocking)

Functional Application:
Human: Inhibits the binding of human angiopoietin-2 to human Tie-2.
*ND50** = 600-800ng/ml (for 10ng/ml of angiopoietin-2)

anti-Angiopoietin-2 mAb (rec.) (blocking) (Angy-2-1)

AG-27B-0016-C100		100 µg
AG-27B-0016PF	Preservative Free	100 µg | 500 µg | 1 mg

Clone: Angy-2-1

Isotype: Mouse IgG2bλ

Application: ELISA, FUNC (Blocking)

Functional Application:
Mouse: Inhibits the binding of mouse angiopoietin-2 to mouse Tie-2.
ND_{50}* = 50-60ng/ml (for 10ng/ml of mouse angiopoietin-2)
Human: Inhibits the binding of human angiopoietin-2 to human Tie-2.
ND_{50}* = 8-12ng/ml (for 10ng/ml of human angiopoietin-2)

**ND$_{50}$:* = 50% neutralizing dose of antibody for a given concentration of ligand.

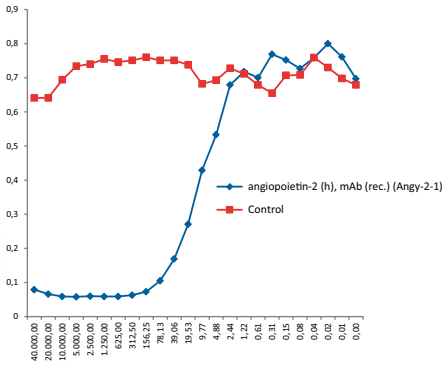


FIGURE: Binding of human Angiopoietin-2 to Tie-2 (human): Fc is inhibited by Angy-2-1. Tie-2 (human):Fc was coated on an ELISA plate at 1µg/ml. Angy-2-1 or an unrelated mAb (recombinant) (Control) were added (starting at 40µg/ml with a twofold serial dilution) together with 20ng/µl of Angiopoietin-2 (human) (Prod. No. AG-40B-0114). After incubation for 1h at RT, the binding was detected using an anti-FLAG antibody (HRP).

anti-IL-33 (mouse), mAb (rec.) (blocking) (Bondy-1-1)

BULK

AG-27B-0013-C100		100 µg
AG-27B-0013PF	Preservative Free	100 µg | 500 µg | 1 mg

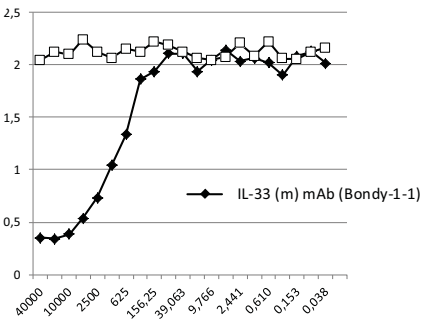
Clone: Bondy-1-1

Isotype: Mouse IgG2b

Application: ELISA, FUNC (Blocking)

Functional Application:
Inhibits the binding of mouse IL-33 to ST2/IL-1RAcP.

FIGURE: Binding of IL-33 (mouse) to ST2/IL-1RAcP is inhibited by Bondy-1-1. IL-33 (mouse) was coated on an ELISA plate at 1µg/ml. Bondy-1-1 or an unrelated mAb (recombinant) (Control) were added (starting at 40µg/ml with a two-fold serial dilution) together with 100µl of supernatant of cells containing ST2 (human):Fc/IL-1RAcP (human):Fc. After incubation for 1 h at RT, the binding was detected using an anti-Fc human antibody (HRP).



APPLICATIONS: FACS: Flow Cytometry; **FUNC:** Functional Application; **ICC:** Immunocytochemistry; **IHC:** Immunohistochemistry **IP:** Immunoprecipitation; **WB:** Western blot

FORMULATION: PF = Preservative free
SPECIES: Hu = Human; **Ms** = Mouse; **Rt** = Rat; **Rb** = Rabbit; **Prm** = Primate

Other Blocking/Neutralizing FuncAbs™

anti-BAFF (human), mAb (blocking) (4.62)

BULK

AG-20B-0017-C100		100 µg
AG-20B-0017B-C100	Biotin	100 µg

Application: ELISA, IP, FUNC (Neutralizing)

Functional Application:
Inhibition/Neutralizing of human BAFF binding to Raji cells.

anti-TRAIL-R1 (human), mAb (HS101)

BULK

AG-20B-0022PF-C100	Preservative Free	100 µg

Different labels available.

Application: FACS, IP, ICC, FUNC (Neutralizing)

Functional Application:
Inhibition/Neutralizing (blocks TRAIL-R1 mediated killing if applied in solution).

anti-TRAIL-R2 (human), mAb (HS201)

AG-20B-0023PF-C100	Preservative Free	100 µg

Different labels available.

Application: FACS, IP, ICC, FUNC (Neutralizing)

Functional Application:
Inhibition/Neutralizing (blocks TRAIL-R2 mediated killing if applied in solution).

LIT (FOR HS101 AND HS201): IFN-alpha-stimulated neutrophils and monocytes release a soluble form of TNF-related apoptosis-inducing ligand (TRAIL/Apo-2 ligand) displaying apoptotic activity on leukemic cells: C. Tecchio, et al.; Blood **103,** 3837 (2004)

anti-NS5B (HCV), mAb (blocking) (5B-12B7)

AG-20B-0003-C100		100 µg

Application: ICC, IP, FUNC (Blocking)

Functional Application:
Blocks the RNA-dependent RNA polymerase activity *in vitro*.

LIT: Functional properties of a monoclonal antibody inhibiting the hepatitis C virus RNA-dependent RNA polymerase: D. Moradpour, et al.; J. Biol. Chem. **277,** 593 (2002)

anti-BTLA (human), mAb (6F4)

AG-20B-0049-C100		100 µg

Application: ELISA, FACS, FUNC (Blocking)

Functional Application:
Inhibits interaction of BTLA to HVEM or UL144.

anti-LAG-3, mAb (blocking) (11E3)

BULK

AG-20B-0011-C100		100 µg
AG-20B-0011PF-C100	Preservative Free	100 µg

Application: ELISA, ICC, IHC, IP, WB, FUNC (Blocking)

Functional Application:
Blocks LAG-3/MHC class II interactions.

LIT: Cellular expression and tissue distribution of the human LAG-3-encoded protein, an MHC class II ligand: B. Huard, et al.; Immunogenetics **39,** 213 (1994)

anti-LAG-3 (human), mAb (blocking) (17B4)

AG-20B-0012-C100		100 µg
AG-20B-0012PF-C100	Preservative Free	100 µg

Different labels available.

Application: ICC, IHC, IP, WB, FUNC (Blocking)

Functional Application:
Blocks LAG-3/MHC class II interactions.

LIT: The negative regulatory function of the lymphocyte-activation gene-3 co-receptor (CD223) on human T cells: L. Macon-Lemaitre and F. Triebel; Immunology **115,** 170 (2005)

anti-VEGF-A (human), mAb (3(6D3))

AG-20T-0105-C200		200 µg

Application: ELISA, WB, FUNC (Neutralizing)

Functional Application:
Inhibits VEGF-A signaling.

LIT: DLL1-mediated Notch activation regulates endothelial identity in mouse fetal arteries: I. Sörensen, et al.; Blood **113,** 5680 (2009)

Inducing/Activating Antibodies [FuncAbs™]

anti-LTβR (mouse), mAb (4H8 WH2)

AG-20B-0008-C100		100 µg
AG-20B-0008PF-C100	Preservative Free	100 µg
Clone:		4H8 WH2
Isotype:		Rat IgG2a
Application:		FACS, FUNC (Activation)

anti-LTβR (mouse), mAb (3C8)

AG-20B-0041-C100		100 µg
AG-20B-0041PF-C100	Preservative Free	100 µg
Clone:		3C8
Isotype:		Rat IgG1κ
Application:		FUNC (Activation)

Functional Application for 4H8 WH2 and 3C8:
Agonists inducing BAFF, chemokines and integrins *in vitro* and *in vivo*.

LIT: LTβR Signaling Induces Cytokine Expression and Up-Regulates Lymphangiogenic Factors in Lymph Node Anlagen. M.F. Vondenhoff, et al.; J. Immunol. **182,** 5439 (2009)

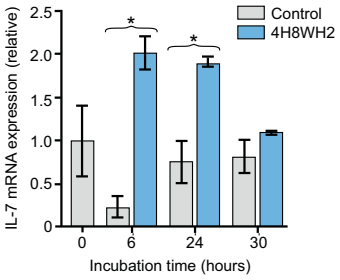


FIGURE: Treatment of cultured WT MEFs with agonistic LTβR mAb (4H8 WH2), but not with an isotype matched control mAb, results in the up-regulation of IL-7 mRNA expression. MEFs were collected at 6, 24, and 30 h after stimulation with 4H8 WH2. Relative expression levels at t = 0 were set at 1,0. Experiments were performed three times. *, $p < 0.05$.

anti-CD40 (mouse), mAb (FGK45)

BULK THE STANDARD

AG-20B-0036		100 µg | 500 µg
AG-20B-0036PF	Preservative Free	100 µg | 500 µg
Clone:		FGK45
Isotype:		Rat IgG2a
Application:		FACS, FUNC (Activation)

Functional Application:
Activates B and NK cells *in vivo* and *in vitro*.

LIT: Ovarian insufficiency and early pregnancy loss induced by activation of the innate immune system: A. Erlebacher, et al.; J. Clin. Invest. **114,** 39 (2004)

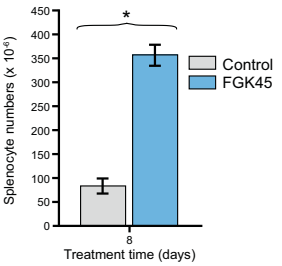


FIGURE: Systemic immune activation by CD40 ligation. Mice were sacrificed on day 8 after daily treatment on day 4-7 with FGK45 or control. FGK45 treatment, elevated splenocyte numbers in both groups. *$P < 0.005$. Data represent mean ± SD for three to four mice per group.

anti-Fas (human), mAb (APO-1-3)

THE STANDARD

AG-20B-0062PF-C050	Preservative Free	50 µg
Clone:		APO-1-3
Isotype:		Mouse IgG3
Application:		FACS, IP, WB, FUNC (Activation)

Functional Application:
Induces apoptosis with or without cross-linking (Protein A), depending on cell type.

LIT: Monoclonal antibody-mediated tumor regression by induction of apoptosis: B.C. Trauth, et al.; Science **245,** 301 (1989)

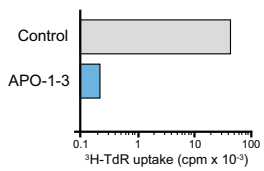


FIGURE: : Induction of growth Inhibition by apoptosis by APO-1-3 or control medium. SKW6.4 cells were pre-incubated with APO-1-3 (100 nglml). ['H)TdR incorporation was measured.

APPLICATIONS: FACS: Flow Cytometry; **FUNC:** Functional Application; **EM:** Electron Microscopy; **ICC:** Immunocytochemistry; **IHC:** Immunohistochemistry **IP:** Immunoprecipitation; **WB:** Western blot

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SPECIES: Hu = Human; **Ms** = Mouse; **Rt** = Rat; **Rb** = Rabbit; **Prm** = Primate

Depleting Antibodies [FuncAbs™]

anti-BAFF-R (mouse), mAb (9B9)

BULK

AG-20B-0034-C100		100 µg
AG-20B-0034PF-C100	Preservative Free	100 µg
AG-20B-0034B-C100	Biotin	100 µg

Different labels available.

Clone:	9B9
Isotype:	Rat IgG2a
Application:	ELISA, IP, FUNC (Depletion)

Functional Application:
Depletes B cells *in vivo*.

LIT: Crucial role for BAFF-BAFF-R signaling in the survival and maintenance of mature B cells: M. Rauch, et al.; PLoS ONE **4,** e5456 (2009)

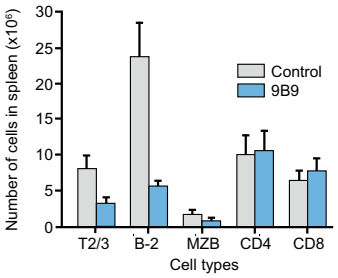


FIGURE: C57BL/6 mice were injected i.v. at day 0 with 0.5mg of 9B9. Absolute numbers of splenic T1 and T2/3 immature B cells, B-2 and MZ B cells, CD4 and CD8 T cells in controls (black bars) and 9B9 injected C57BL/6 mice at day 14 after injection (white bars). 5 mice were analyzed for each group.

The best depleting antibody for neutrophils in mice!

anti-Neutrophils (mouse), mAb (blocking) (Nimp-R14)

BULK

AG-20B-0043-C100		100 µg
AG-20B-0043PF	Preservative Free	500 µg | 2 mg | 10 mg
AG-20B-0043B-C100	Biotin	100 µg

Different labels available.

Clone:	Nimp-R14
Isotype:	Rat IgG2a
Application:	FACS, IHC, ICC, FUNC (Depletion)

Functional Application:
Optimal reagent to deplete neutrophils *in vivo* (250 µg/mouse).

LIT: An immunomodulatory function for neutrophils during the induction of a CD4+ Th2 response in BALB/c mice infected with Leishmania major: F. Tacchini-Cottier, et al.; J. Immunol. **165,** 628 (2000)

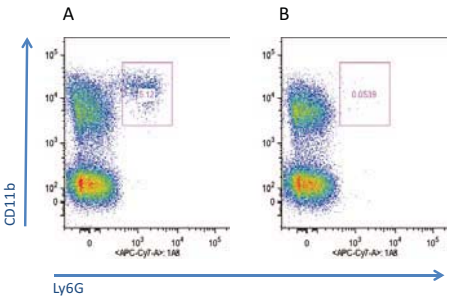


FIGURE: Mouse neutrophils are depleted *in vivo* by Nimp-R14. Mice were injected i.p. with 250µg of Nimp-R14 (B) or with Control mAb (A) in BALB/c mouse 6 h prior to *Leishmania major* infection (3x106 parasites injected in the hind footpad). 3 days later, blood (100µl) was subjected to flow cytometric analysis after staining with APC/CY7-labeled anti-Ly6G antibody (clone 1A8).

Custom Recombinant Monoclonal Antibodies [RecMAbs™]

AdipoGen®
CUSTOM PRODUCTION

AdipoGen® offers a very efficient custom service for the production of recombinant monoclonal antibodies.

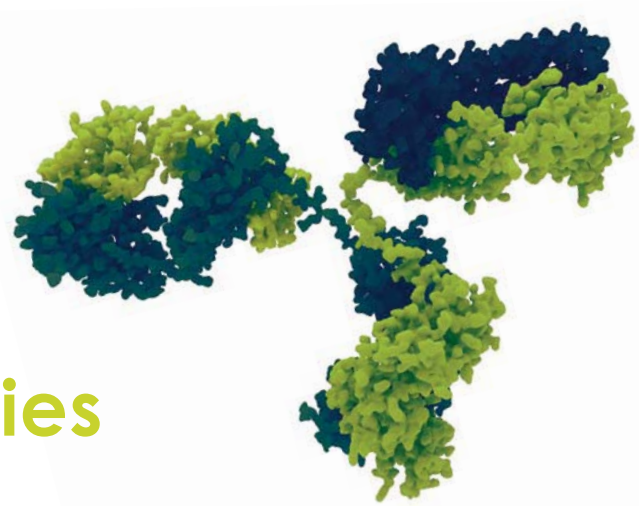
FEATURES:

- Produced in CHO cells.
- Isolated from phages and produced in bacterial (no animals used).
- Ideal for conserved antigens (which are poorly immunogenic in animals).
- Ideal for the development of antibodies against activated forms of proteins.
- Ideal for the development of blocking (inhibitory) antibodies.

See page 8–11 for more RecMAbs™

Ancell™

THE SPECIALIST FOR IMMUNOLOGY
HIGH QUALITY RESEARCH REAGENTS



Functional Antibodies

Blocking/Neutralizing Antibodies [FuncAbs™]

PRODUCT NAME	PID (*)	APPLICATIONS	FUNCTIONAL APPLICATION
CD4 (human), mAb (QS4120)	ANC-147	FUNC, FACS, ELISA	Blocks binding of HIV-1 gp120 protein to CD4 and also blocks HLA Class II rosette formation.
CD11a (human), mAb (38)	ANC-158	FUNC, FACS, WB	Blocks binding of ICAM-1 and ICAM-3 to LFA-1 at 5-10 µg/ml.
CD11b (human), mAb (ICRF44)	ANC-159	FUNC, FACS	Blocks homotypic neutrophil and monocyte (FMLP induced) aggregation.
CD16 (human), mAb (3G8)	ANC-165	FUNC, FACS	Blocks binding of complexed IgG to CD16.
CD18 (human), mAb (IB4)	ANC-167	FUNC, FACS	Blocks binding of ICAM-1 and ICAM-3 to LFA-1.
CD20 (human), mAb (2H7)	ANC-169	FUNC, FACS	Inhibits B-lymphocyte differentation and induced Ig secretion.
CD21 (human), mAb (BU33)	ANC-170	FUNC, FACS, WB	Inhibits binding to CD23.
CD31 (human), mAb (158-2B3)	ANC-180	FUNC, FACS	Blocks homophilic interaction and heterophilic transendothelial migration.
CD32 (human), mAb (7.3)	ANC-181	FUNC, FACS	Blocks immune complex binding.
CD40L [CD154] (human), mAb (24-31)	ANC-353	FUNC, FACS, ELISA, IHC, WB	Blocks MLR, sgp39 induced human B cell proliferation and T cell dependent B cell differentiation.
CD44 (human), mAb (BU75)	ANC-352	FUNC, FACS, WB	Blocks binding of HA to CD44.
CD49d (human), mAb (BU49)	ANC-200	FUNC, FACS	Blocks VLA-4 binding to VCAM-1. It can be used to aid in purification of FoxP3$^+$ Treg cells.
			Induces IL-8 production by U-937 cells.
CD50 (human), mAb (186-2G9)	ANC-201	FUNC, FACS	Blocks binding of CD11a (LFA-1) to CD50 (ICAM-3).
CD54 (D1) (human), mAb (15.2)	ANC-205	FUNC, FACS, ELISA, WB	Inhibits CD54 binding to LFA-1.
CD54 (D2) (human), mAb (8.4A6)	ANC-206	FUNC, FACS, ELISA	Inhibits CD54 binding to LFA-1.
CD58 (human), mAb (TS2)	ANC-210	FUNC, FACS	Inhibits HLA-DR mediated T cell cytotoxicity.
CD64 (human), mAb (10.1)	ANC-216	FUNC, FACS, WB	Blocks binding of FcγRI to immunoglobulin opsonized cells.
CD70 (human), mAb (BU69)	ANC-222	FUNC, FACS, ELISA, ICC, IHC	Inhibits T cell proliferation induced by dendritic cells.
CD62E (human), mAb (HAE-1f)	ANC-240	FUNC, FACS	Blocks the function of CD62E.
CD62P (human), mAb (G1)	ANC-252	FUNC, FACS	Blocks the activated endothelium or platelet-neutrophil interaction.
CD62L (human), mAb (LAM 1-116)	ANC-261	FUNC, FACS	Blocks CD62L function and induces expression of β-1 and β-2 integrins.
CD80 (human), mAb (BB1)	ANC-100	FUNC, FACS, ELISA, WB	Blocks Th induced B cell Ig synthesis and blocks binding of soluble CD152 Ig fusion protein to CD80.
CD80 (human), mAb (P1.H1.A1.A1)	ANC-110	FUNC, FACS, ELISA	Blocks binding of soluble CD152 Ig fusion protein to CD80.
CD86 (human), mAb (BU63)	ANC-307	FUNC, FACS	Blocks MLR and blocks binding of soluble CD152-mouse Ig fusion protein to CD86.

(*) The Ancell Product # is build by the prefix (ANC-), main PID (3 digits) and a suffix (3 digits). The last 3 digits define the labels:
-020 = Preservatives | -820 = Preservative Free | -030 = Biotin | -040 = FITC | -050 = R-PE | -060 = APC | -520 = F(ab')2 | -580 = Fab | -070 = PE-Cy7 | -350 = DyLight350
FAB: Fragment Antigen Binding; **FACS:** Flow Cytometry; **FUNC:** Functional Application; **ICC:** Immunocytochemistry; **IHC:** Immunohistochemistry; **IP:** Immunoprecipitation; **WB:** Western Blot

Blocking/Neutralizing Antibodies *continued*

PRODUCT NAME	PID (*)	APPLICATIONS	FUNCTIONAL APPLICATION
CD94 (human), mAb (HP-3D9)	ANC-315	FUNC, FACS	Inhibits IL-2 dependent proliferation of NK cells.
CD104 (human), mAb (UMA 9)	ANC-325	FUNC, FACS, WB	Partially blocks binding to laminin.
CD106 (human), mAb (1.G11B1)	ANC-327	FUNC, FACS, ELISA, IHC, WB	Blocks leukocyte adhesion.
CD122 (human), mAb (9A2)	ANC-343	FUNC, FACS	Inhibits binding of IL-2 to IL-2Rβ (CD122).
CD137 (human), mAb (4B4-1)	ANC-360	FUNC, FACS, ELISA	Blocks binding of CD137-human Ig fusion protein to Raji cells.
CD147 (human), mAb (UM-8D6)	ANC-376	FUNC, FACS, IP, WB	Inhibits homotypic aggregation, adhesion to matrix proteins and migration through matrigel.
CD152 (human), mAb (ANC152.2/8H5)	ANC-359	FUNC, FACS, ELISA	Blocks binding of CD152 (CTLA-4)- human Ig fusion protein to its CD80/CD86 receptor.
CD162 (human), mAb (PL1)	ANC-389	FUNC, FACS, WB	Blocks binding of CD162 to CD62P.
CD165 (human), mAb (AD2)	ANC-392	FUNC, FACS	Blocks the function of CD165.
CD166 (human), mAb (3A6)	ANC-393	FUNC, FACS	Blocks binding of CD6 to CD166.
CD178 (human), mAb (ALF-2.1A)	ANC-399	FUNC, FACS, ELISA	Blocks CD178 activity.
CD252 (human), mAb (ANC10G1)	ANC-400	FUNC, FACS, ELISA	Blocks binding of recombinant CD134-mouse Ig fusion protein.
CD257 (human), mAb (ANC2H3)	ANC-266	FUNC, ELISA	Blocks binding of recombinant human CD257(BAFF) to receptors on Raji cells in flow cytometry.
CD272 (human), mAb (ANC6E9)	ANC-272	FUNC, FACS, ELISA	Blocks binding of biotinylated CD270(HVEM)-mouse Ig fusion protein to CD272-mouse Ig fusion protein in EIA.
CD278 (human), mAb (ANC6C6)	ANC-265	FUNC, FACS, ELISA	Blocks binding of recombinant GL50-mouse Ig fusion protein to HPB-MLT cells.
TNF-α (human), mAb (J1D9)	ANC-398	FUNC, FACS, WB	Neutralizes TNF-α biological activities.

Activating/Inducing Antibodies [FuncAbs™]

PRODUCT NAME	PID (*)	APPLICATIONS	FUNCTIONAL APPLICATION
CD3 (human), mAb (UCHT1)	ANC-144	FUNC, FACS, WB	Activates T cells expressing CD3ε.
CD6 (human), mAb (3F7B6)	ANC-151	FUNC, FACS, WB	Activates T cells.
CD7 (human), mAb (3A1E)	ANC-152	FUNC, FACS	Induces T cell transmembrane calcium flux.
CD15 (human), mAb (AHN1.1)	ANC-164	FUNC, FACS, IHC	Activates normal monocytes and inhibits neutrophil chemotaxis.
CD19 (human), mAb (BU12)	ANC-168	FUNC, FACS	Induces adhesion of B cells.
CD28 (human), mAb (ANC28.1/5D10)	ANC-177	FUNC, FACS, ELISA	Stimulates expression of IL-2 from $CD28^+$ cells.
CD40 (human), mAb (BE-1)	ANC-189	FUNC, FACS, ELISA, IP	Partially activates B cells.
CD40 (human), mAb (EA-5)	ANC-300	FUNC, FACS, ELISA	Partially activates B cells.
CD43 (human), mAb (DFT1)	ANC-192	FUNC, FACS, WB	Partially induces apoptosis in hemopoietic progenitor cells and also induces homopoietic aggregation.
CD49d (human), mAb (BU49)	ANC-200	FUNC, FACS	Blocks VLA-4 binding to VCAM-1. It can be used to aid in purification of $FoxP3^+$ Treg cells.
			Induces IL-8 production by U-937 cells.
CD60 (human), mAb (UM4D4)	ANC-212	FUNC, FACS, WB	Activates T cells.
CD79b (human), mAb (SN8)	ANC-301	FUNC, FACS, WB	Induces signal transduction in B cells.
CD105 (human), mAb (SN6)	ANC-326	FUNC, FACS, IHC	Augments binding of TGF-β1 to CD105 expressing leukemia cells.
IgM (human), mAb (UCHB1)	ANC-141	FUNC, FACS, ELISA	Delivers a costimulatory signal to B cells *in vitro*.

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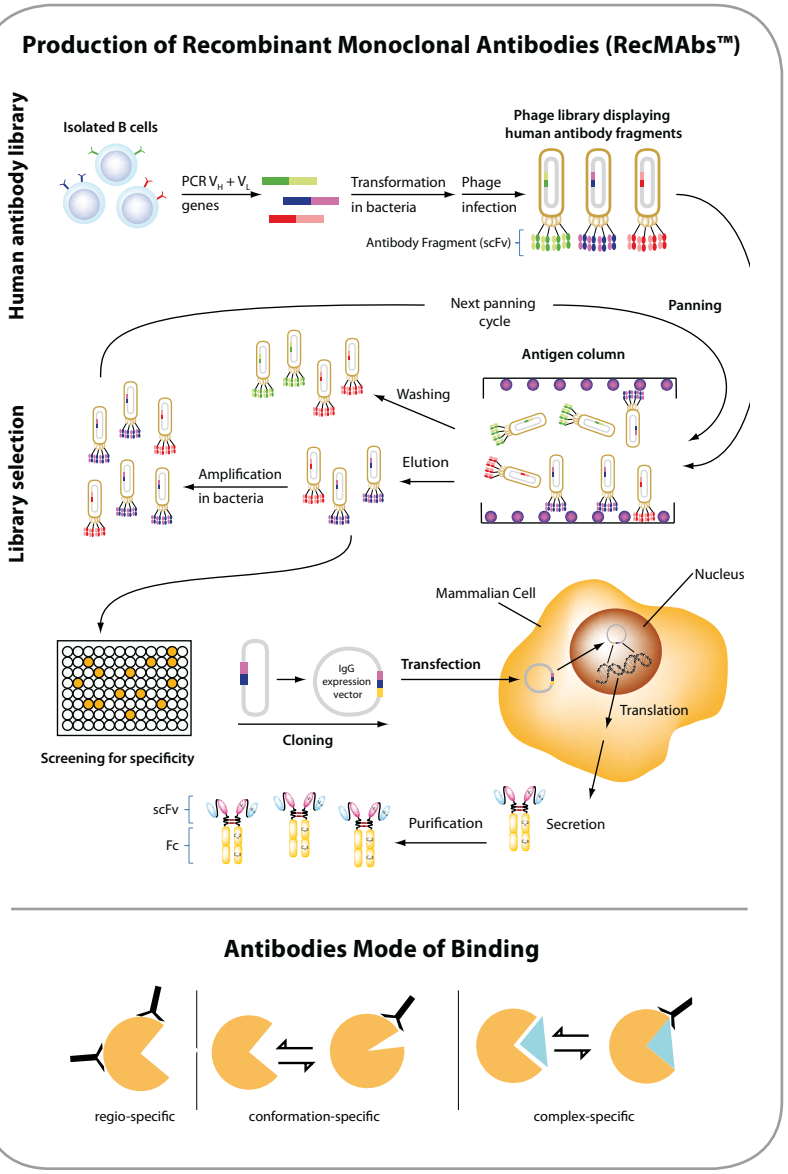
-020 = Preservatives | -820 = Preservative Free | -030 = Biotin | -040 = FITC | -050 = R-PE | -060 = APC | -520 = F(ab')2 | -580 = Fab | -070 = PE-Cy7 | -350 = DyLight350
FAB: Fragment Antigen Binding; **FACS:** Flow Cytometry; **FUNC:** Functional Application; **ICC:** Immunocytochemistry; **IHC:** Immunohistochemistry; **IP:** Immunoprecipitation; **WB:** Western Blot

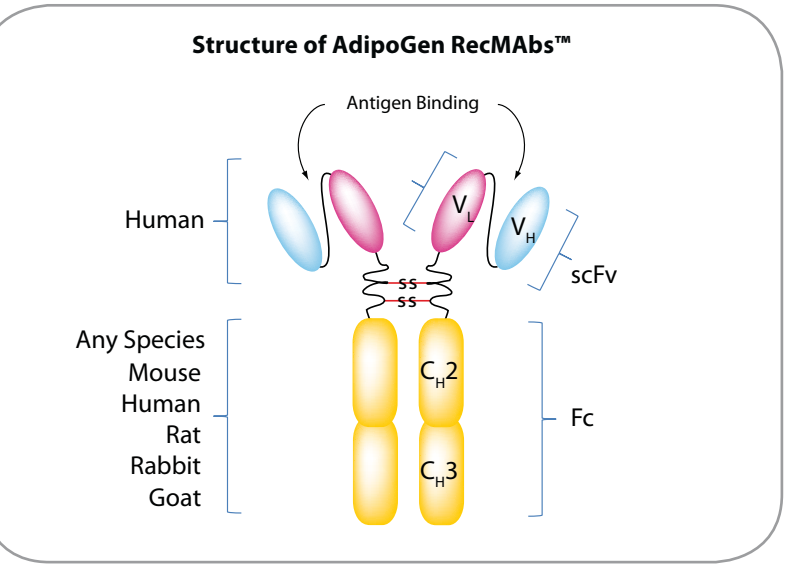
Recombinant Monoclonal Antibodies [RecMAbs™]

Antibody phage display is an *in vitro* technology to generate **recombinant monoclonal antibodies (RecMAbs™)**. It is an alternative to the hybridoma technology, since it circumvents the limitations of the immune system. Antibodies developed by **"antibody phage display technology"** use human naive antibody gene libraries. These libraries consist of billions of scFv (single chain fragment variable) composed of VH (variable domain of the human immunoglobulin heavy chain) and VL (variable domain of the human immunoglobulin light chain) connected by a polypeptide linker. The antibody fragments are fused to the coat protein pIII and displayed on the surface of filamentous bacteriophages (M13). The scFvs are selected *in vitro* by affinity selection on the antigen in a process termed panning, where the antigen of interest is coated on a vial (see Figure). Panning methods are based on four major steps: i) preparation of phage-displaying libraries; ii) adsorbing the specific binding phage, iii) removal of non-specific or low affinity phages, and recovering of target binders, that will be reamplified after bacteria infection for the next round of selection. Multiple rounds of panning are performed to enrich for the antigen-specific scFv-phages. Monoclonal antibodies are subsequently identified by screening after the last round of selection. The selected monoclonal scFv is cloned into an appropriate vector containing a Fc portion of interest and then produced in mammalian cells to generate an IgG like scFv-Fc fusion protein.

There are many advantages to use recombinant antibodies instead of classical antibodies: i) economical production and permanent storage of DNA clones are some of the assets of the recombinant antibody approach; ii) absence of requirement of sacrificing animals in large animal facility; iii) use of a single stable antibody fragment makes it straightforward to reformat a RecMAb™ into a full-length IgG construct or a single chain fragment variable (Fv).

An important attribute of the RecMAbs™ phage display approach is the ability to design selection strategies to generate antibodies with customized functions (FuncAbs™), which furthermore can be classified based on activity (see frontpage) or mode of binding. For instance, it is possible to generate RecMAbs™ that: (1) preferentially recognize a specific conformational state and thus, have the potential to induce a specified conformational change; (2) target specific regions of the surface of the target protein ("regio-specific") or (3) specifically recognize multi-protein complexes.





RecMAbs™ –
Antibodies developed from a NON-ANIMAL SOURCE using *in vitro* antibody phage display technology

AdipoGen®

FEATURES:

- Developed from a human antibody phage display library.
- Consists of scFv (single chain fragment variable) composed of VH (variable domain of the human immunoglobulin heavy chain) and VL (variable domain of the human immunoglobulin light chain) fused to a Fc region.
- Produced in mammalian cells (CHO or HEK 293).
- Similar properties compared to monoclonal antibodies developed in mice / rat (e.g. affinity in the low nanomolar range).
- Standard secondary antibodies can be used.
- Ideal for conserved antigens (which are poorly immunogenic in animals).
- Detect conformational epitopes (e.g. GTP-bound proteins).
- Detect protein modifications (e.g. phosphorylations, ubiquitinations).
- Possibility to exchange the Fc region with Fc from other species.

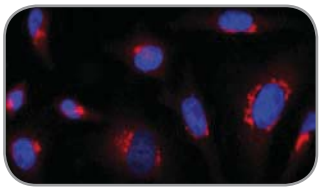
Ask for Custom Production!

LATEST REVIEW: Generating conformation-specific synthetic antibodies to trap proteins in selected functional states: M. Paduch, et al.; Methods **60,** 3 (2013)

Conformation-specific Recombinant Antibodies

anti-Rab1-GTP, mAb (rec.) (ROF7)

AG-27B-0006-C100		100 µg
Clone:		ROF7
Isotype:		Human IgG2bλ
Specificity:		Recognizes human, mouse, rat and dog Rab1a-GTP and Rab1b-GTP.
Application:		ICC, IP

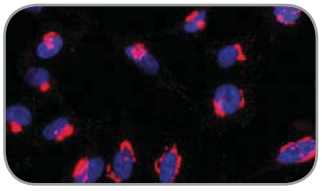


LIT: Characterization of single chain antibody targets through yeast two hybrid: O. Vielemeyer, et al.; BMC Biotechnol. **10,** 59 (2010)

FIGURE: Rab1-GTP is detected by immunocytochemistry using ROF7. *Picture courtesy of Dr. Sandrine Moutel & Dr. Franck Perez Lab, Curie Institute, Paris.*

anti-Rab6-GTP, mAb (rec.) (AA2)

AG-27B-0004-C100		100 µg
AG-27B-0004TD-C100	ATTO 488	100 µg
Clone:		AA2
Isotype:		Human IgG2bλ
Specificity:		Recognizes human, mouse and drosophila GTP-bound Rab6a and Rab6b and mutant Rab6Q72L. Does not detect Rab6-GDP.
Application:		ICC, WB

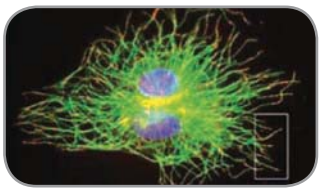


LIT: Recombinant antibodies to the small GTPase Rab6 as conformation sensors: C. Nizak, et al.; Science **300,** 984 (2003)

FIGURE: Rab6-GTP is detected by immunocytochemistry using AA2. *Picture courtesy of Dr. Sandrine Moutel & Dr. Franck Perez Lab, Curie Institute, Paris.*

anti-Tubulin-GTP, mAb (rec.) (MB11)

AG-27B-0009-C100		100 µg
Clone:		MB11
Isotype:		Human IgG2bλ
Specificity:		Recognizes human, mouse, rat and drosophila tubulin-GTP.
Application:		ICC, WB



LIT: Detection of GTP-Tubulin Conformation in Vivo Reveals a Role for GTP Remnants in Microtubule Rescues: A. Dimitrov, et al.; Science **322,** 1353 (2008)

FIGURE: Tubulin-GTP is detected by immunocytochemistry using MB11. *Picture courtesy of Dr. Sandrine Moutel & Dr. Franck Perez Lab, Curie Institute, Paris*

APPLICATIONS: **FACS:** Flow Cytometry; **FUNC:** Functional Application; **EM:** Electron Microscopy; **ICC:** Immunocytochemistry; **IHC:** Immunohistochemistry **IP:** Immunoprecipitation; **WB:** Western blot

FORMULATION: **PF** = Preservative free

SPECIES: **Hu** = Human; **Ms** = Mouse; **Rt** = Rat; **Rb** = Rabbit; **Prm** = Primate

Other Recombinant Monoclonal Antibodies [RecMAbs™]

anti-Giantin, mAb (rec.) (TA10)

AG-27B-0003-C100		100 µg
AG-27B-0003TD-C100	ATTO 488	100 µg
Clone:		TA10
Isotype:		Human IgG2bλ
Specificity:		Recognizes human and mouse giantin.
Application:		ICC

LIT: Recombinant antibodies selected against subcellular fractions to track endogenous protein dynamics in vivo: C. Nizak, et al.; Traffic **7**, 739 (2003)

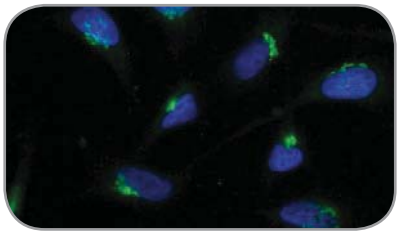
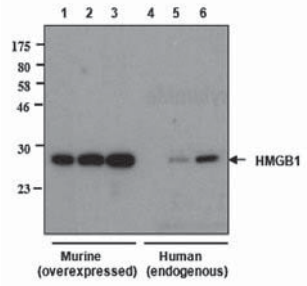


FIGURE: Human giantin is detected by immunocytochemistry using TA10 (ATTO 488) (Prod. No AG-27B-0003TD). *Picture courtesy of Dr. Sandrine Moutel & Dr. Franck Perez Lab, Curie Institute, Paris.*

anti-HMGB1, mAb (rec.) (Giby-1-4)

AG-27B-0002-C100	100 µg
Clone:	Giby-1-4
Isotype:	Human IgG2bλ
Specificity:	Recognizes human, mouse and rat HMGB1.
Application:	ELISA, WB

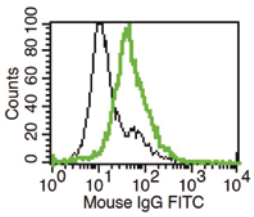
FIGURE: Western blot analysis of human and rat HMGB1 using Giby-1-4. Different amounts of cell extracts from HEK293T cells (3µg, 5µg and 30µg) either transfected with a plasmid coding for rat HMGB1 (lanes 1, 2, 3) or non-transfected (lanes 4, 5, 6), were separated by SDS-PAGE under reducing conditions, transferred to nitrocellulose and incubated with anti-HMGB1, mAb (rec.) (Giby-1-4) (1µg/ml). Proteins were visualized by a chemiluminescence detection system.



anti-IL-1R2 (mouse), mAb (rec.) (Praxy-1-1)

AG-27B-0011-C100	100 µg
Clone:	Praxy-1-1
Isotype:	Human IgG2bλ
Specificity:	Recognizes mouse IL-1R2.
Application:	ELISA, FACS

FIGURE: Detection of endogenous mouse IL-1R2 using Praxy-1-1. *In vitro*-cultivated BMN (mouse Neutrophils) (stimulated 24h with hydrocortisone) were stained with Praxy-1-1 (thick green line) or an isotype control (thin black line) at 5µg/ml each, revealed with a secondary anti-mouse antibody (FITC) and then analyzed by flow cytometry.



anti-Myosin IIA (non-muscle) (heavy chain), mAb (rec.) (SF9)

AG-27B-0010-C100	100 µg
Clone:	SF9
Isotype:	Human IgG2bλ
Specificity:	Recognizes human, mouse, rat and drosophila myosin IIA (heavy chain).
Application:	EM, ELISA, ICC, WB

LIT: Recombinant antibodies selected against subcellular fractions to track endogenous protein dynamics in vivo: C. Nizak, et al.; Traffic **7**, 739 (2003)

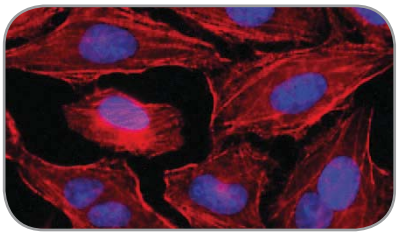


FIGURE: Human myosin IIA (non-muscle) (heavy chain) is detected by immunocytochemistry using SF9. *Picture courtesy of Dr. Sandrine Moutel & Dr. Franck Perez Lab, Curie Institute, Paris.*

APPLICATIONS: FACS: Flow Cytometry; **FUNC:** Functional Application; **EM:** Electron Microscopy; **ICC:** Immunocytochemistry; **IHC:** Immunohistochemistry **IP:** Immunoprecipitation; **WB:** Western blot

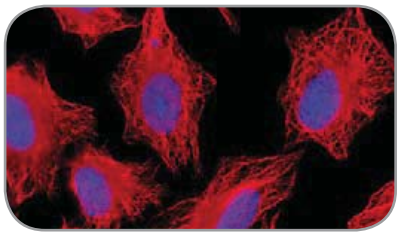
FORMULATION: PF = Preservative free
SPECIES: Hu = Human; **Ms** = Mouse; **Rt** = Rat; **Rb** = Rabbit; **Prm** = Primate

anti-α-Tubulin, mAb (rec.) (F2C)

AG-27B-0005-C100		100 µg
AG-27B-0005TD-C100	ATTO 488	100 µg

Clone:	F2C
Isotype:	Human IgG2λ
Specificity:	Recognizes mouse, bovine and human α-tubulin.
Application:	ICC, WP (only AG-27B-0005)

LIT: Recombinant antibodies selected against subcellular fractions to track endogenous protein dynamics in vivo: C. Nizak, et al.; Traffic **7,** 739 (2003)



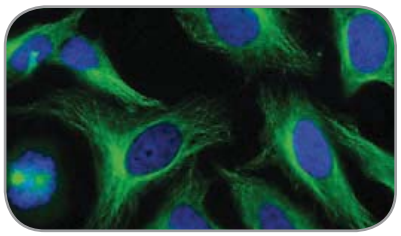


FIGURE (ABOVE): Human α-tubulin is detected by immunocytochemistry using F2C. *Picture courtesy of Dr. Sandrine Moutel & Dr. Franck Perez Lab, Curie Institute, Paris.*

FIGURE (BELOW): Human α-tubulin is detected by immunocytochemistry using F2C (ATTO488). *Picture courtesy of Dr. Sandrine Moutel & Dr. Franck Perez Lab, Curie Institute, Paris.*

anti-β-Tubulin, mAb (rec.) (S11B)

AG-27B-0008-C100	100 µg

Clone:	S11B
Isotype:	Human IgG2λ
Specificity:	Recognizes human, mouse, rat, pig, drosophila and monkey β-tubulin.
Application:	ELISA, ICC, IP

LIT: Recombinant antibodies selected against subcellular fractions to track endogenous protein dynamics in vivo: C. Nizak, et al.; Traffic **7,** 739 (2003)

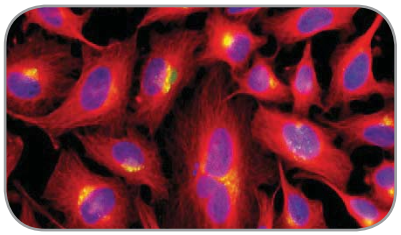


FIGURE: Human β-tubulin is detected by immunocytochemistry using S11B. *Picture courtesy of Dr. Sandrine Moutel & Dr. Franck Perez Lab, Curie Institute, Paris.*

Newly Released RecMAbs™

anti-IL-33 (mouse), mAb (rec.) (Carly-1-4)

AG-27B-0012-C100	100 µg

Clone:	Carly-1-4
Isotype:	Human IgG2λ
Specificity:	Recognizes mouse IL-33.
Application:	ELISA, WB

anti-PEDF (human), mAb (rec.) (Serpy-1-4)

AG-27B-0014-C100	100 µg

Clone:	Serpy-1-4
Isotype:	Human IgG2λ
Specificity:	Recognizes human PEDF.
Application:	ELISA, WB

Also available:

anti-EGFP, mAb (rec.) (G3)

AG-27B-0007-C100	100 µg

Clone:	G3
Isotype:	Human IgG2λ
Specificity:	Recognizes EGFP, ECFP and EYFP.
Application:	ELISA, ICC, IP

LIT: Fully in vitro selection of recombinant antibodies: S. Moutel, et al.; Biotech. J. **4,** 38 (2009)

UNIQUE

Post-translational Modification-specific Antibody for Cancer Research

Polyglutamylation is a post-translational modification in which glutamate side chains of variable lengths are added on the modified protein. It is evolutionarily conserved and the most prominent substrate is tubulin, the microtubule (MT) building block. Polyglutamylation has been proposed to be involved in the functional adaptation of MTs, as it occurs within the carboxy-terminal tubulin tails that participate directly in the binding of many structural and motor MT-associated proteins. The recent identification of new substrates of polyglutamylation indicates that this post-translational modification could be a potential regulator of diverse cellular processes and be involved in cell cycle and cell proliferation.

anti-Polyglutamylation Modification, mAb (GT335)

AG-20B-0020-C100		100 µg
AG-20B-0020B-C100	Biotin	100 µg

Clone:	GT335
Isotype:	Mouse IgG1κ
Application:	EM, IHC, IP, WB

Recognizes most forms of polyglutamylated tubulin (α- and β-tubulin), independent of the length of the glutamate side chains. No specificity to particular tubulin isoforms nor to tubulin from particular species are observed. Detects also other (poly)glutamylated proteins. Since no consensus modification site is known for protein (poly)glutamylation, the detection is not sequence-specific. However, an acidic environment of the modification site is required.

LITERATURE:

Distribution of glutamylated alpha and beta-tubulin in mouse tissues using a specific monoclonal antibody, GT335: A. Wolff, et al.; Eur. J. Cell Biol. **59,** 425 (1992)
Polyglutamylation of nucleosome assembly proteins: C. Regnard, et al.; J. Biol. Chem. **275,** 15969 (2000)
Glutamylated tubulin: diversity of expression and distribution of isoforms: M.L. Kann, et al.; Cell Motil. Cytoskeleton **55,** 14 (2003)
Polyglutamylation Is a Post-translational Modification with a Broad Range of Substrates: J. van Dijk, et al.; J. Biol. Chem. **283,** 3915 (2008)
Tubulin detyrosination promotes monolayer formation and apical trafficking in epithelial cells: S. Zink , et al.; J. Cell Sci. **125,** 5998 (2012)

THE STANDARD

Inflammasome Signaling Blocking Antibody

anti-Asc, pAb (AL177)

AG-25B-0006-C100		100 µg
AG-25B-0006PF-C100	Preservative Free	100 µg

Source:	Rabbit
Application:	ICC, IHC, IP, WB, FUNC (Blocking)

Functional Application:
Inhibits interaction between Asc and NLRP3, leading to blockade of caspase-1 processing *in vitro*.

LIT: The inflammasome: a molecular platform triggering activation of inflammatory caspases and processing of proIL-beta: F. Martinon, et al.; Mol. Cell. **10,** 417 (2002)

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