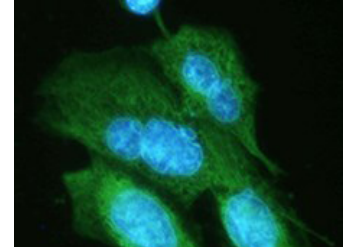


# Validated Antibodies in IHC and ICC

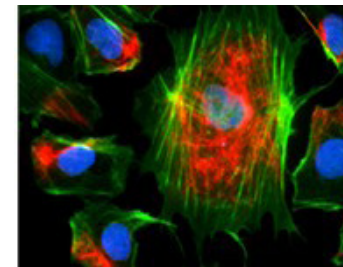
## for Cell Biology, Immunology, and Neuroscience

Antibody validation is of the utmost importance to all areas of biological research. In applications like immunohistochemistry (IHC) and immunocytochemistry (ICC), it can be complicated to assess the effect of sample type and preparation on affinity and specificity of antibodies. In an on-going effort to increase the confident use of our antibodies across all assay platforms, BioLegend has been undertaking a program of extensive antibody validation of existing clones using chromogenic methods like DAB amplification or detection with fluorophores commonly used in microscopy, like Brilliant Violet™ 421 (BV421™), Alexa Fluor® 488 (AF488), Alexa Fluor® 594 (AF594) and Alexa Fluor® 647 (AF647).

Since the need for species-based secondary detection is often a limitation for the number of antibody/fluorophore combinations that can be applied to a sample at the same time and thus the number of parameters that can be imaged at once, BioLegend offers an ever growing number of antibodies directly conjugated to these fluorophores in a focused effort to enable multicolor microscopy. Although not exhaustive, here is a product listing of antibodies that have been validated in-house, either for immunocytochemistry or immunohistochemistry of formalin-fixed paraffin-embedded, frozen acetone-fixed, and OCT embedded tissues. Protocols used for this validation can be found on our website at: [biolegend.com/technical\\_protocols](http://biolegend.com/technical_protocols)



CHO cells transfected with HA-tagged protein were stained with anti-HA.11 Epitope Tag Antibody (clone 16B12) Alexa Fluor® 488 (green) and DAPI (blue).



NTERA-2 cells were stained with anti-Nestin (10C2)/DyLight™ 594 goat anti-mouse IgG (red), Alexa Fluor® 488 Phalloidin (green) and DAPI (blue).

Learn more at: [biolegend.com/microscopy](http://biolegend.com/microscopy)

### Cell Biology

Antibody Specificity	Clone	Validated Applications	Reported Applications	BioLegend Formats Available						
				Purified	Biotin	BV421™	FITC	AF488	AF594	AF647
β-actin	2F1-1	IHC-P, ICC		•		•		•	•	•
Cytochrome c	6H2.B4	ICC		•	•		•	•	•	•
DYKDDDDK (FLAG® tag)	L5	IHC-P, ICC		•				•	•	•
GATA3	16E10A23	ICC		•		•		•	•	•
GFAP	2E1.E9	IHC-P, IHC-F, ICC		•		•		•	•	•
HA.11 Epitope Tag	16B12	ICC		•	•	•	•	•	•	•
H2A.X Phospho (Ser139)	2F3	ICC	IHC-P	•			•	•	•	•
Ki-67	16A8	IHC-F		•		•	•	•		•
Nanog	16H3A48	IHC-P, ICC		•		•		•	•	•
Nestin	10C2	ICC	IHC-F	•		•		•	•	•
OCT4 (OCT3)	3A2A20	ICC		•		•		•	•	•
SOX2	14A6A34	ICC		•		•		•	•	•
T-bet	4B10	ICC	IHC-F	•		•	•	•	•	•
Tubulin Beta 3 (TUBB3)	AA10	IHC-P, IHC-F, ICC		•		•		•	•	•

IHC-F: Immunohistochemistry (frozen)

IHC-P: Immunohistochemistry (paraffin-embedded)

ICC: Immunocytochemistry

BioLegend is ISO 13485:2003 Certified

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Tel: 1.858.768.5800

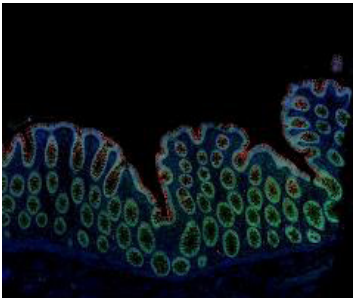
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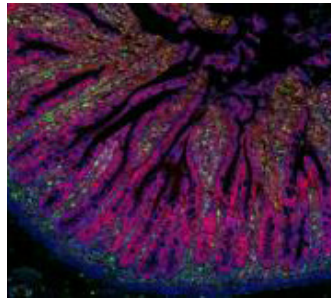


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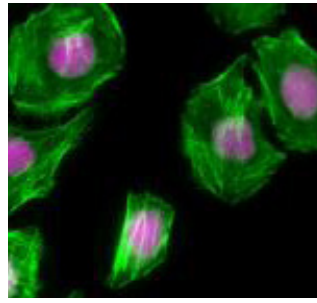




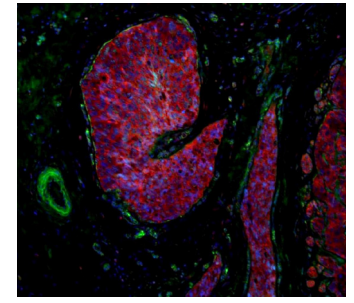
Human paraffin-embedded colon tissue slices were stained with anti-human CD66a/c/e (clone ASL-32) Alexa Fluor® 594 (red), anti-human Galectin-9 (clone 9M1-3) Alexa Fluor® 647 (green) and DAPI (blue).



Human paraffin-embedded small intestine tissue slices were stained with anti-human CD133 (clone 7) Alexa Fluor® 594 (red), anti-human Vimentin (clone O91D3) Alexa Fluor® 647 (green) and DAPI (blue).



HeLa cells were stained with Ki-67 (clone Ki-67) Alexa Fluor® 647 (red), Alexa Fluor® 488 Phalloidin (green) and DAPI (blue).



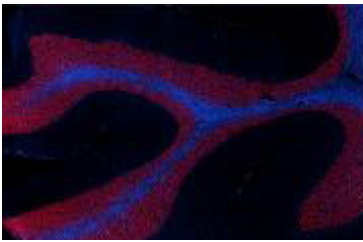
Human paraffin-embedded breast cancer tissue slices were stained with anti-human DcR3 (clone B08/035) Alexa Fluor® 594 (red), anti-Nestin (clone 10C2) Brilliant Violet 421™ (green) and DAPI (blue).

## Human Immunology

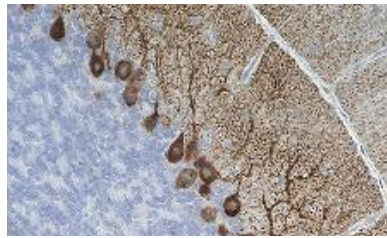
Anti-human Specificity	Clone	Validated Applications	Reported Applications	BioLegend Formats Available							
				Purified	Biotin	BV421™	BV510™	FITC	AF488	AF594	AF647
CD3	UCHT1	IHC-F, ICC		*	*	*	*	*	*	*	*
CD4	RPA-T4	ICC		*	*	*	*	*	*	*	*
CD8	C8/144B	IHC-P		*					*	*	
CD11a	HI111	ICC	IHC-F	*	*			*	*	*	*
CD11b	ICRF44	ICC	IHC-F	*	*	*	*	*	*	*	*
CD14	HCD14	ICC		*	*	*		*	*	*	*
CD15 (SSEA-1)	HI98	IHC-P	IHC-F	*	*			*	*	*	
CD19	HIB19	IHC-F		*	*	*	*	*	*	*	*
CD20	2H7	IHC-F, ICC		*	*	*	*	*	*	*	*
CD31	WM59	ICC	IHC-F	*				*	*	*	*
CD39	A1	IHC-P		*	*	*	*			*	
CD45	HI30	IHC-P	IHC-F, ICC	*	*	*	*	*	*	*	*
CD45RA	HI100	IHC-P	IHC-F, ICC	*	*	*	*	*	*		*
CD45RB	C363-16A	IHC-P		*							
CD45RO	UCHL1	IHC-P	IHC-F	*	*	*	*	*	*	*	*
CD51/61	23C6	ICC	IHC-F	*	*			*	*		*
CD66a/c/e	ASL-32	IHC-P		*		*			*	*	*
CD66b	6/40c	IHC-P		*							
CD66d/e	308/3-3	IHC-P		*							
CD90 (Thy1)	5E10	IHC-F		*	*	*	*	*	*		*
CD95 (Fas)	DX2	ICC	IHC-P, IHC-F	*	*	*	*	*	*		*
CD107a (LAMP-1)	H4A3	ICC	IHC-P		*	*	*	*	*		*
CD107b (LAMP-2)	H4B4	IHC-P		*		*		*		*	*
CD133	clone 7	IHC-P	IHC-F, ICC	*	*					*	
CD138 (Syndecan-1)	MI15	ICC	IHC-P	*		*	*	*		*	*
CD147	HIM6	ICC	IHC-P, IHC-F	*				*	*		*
CD274 (B7-H1, PD-L1)	29E.2A3	IHC-P	IHC-F	*	*	*	*			*	
CD278 (ICOS)	C398.4A	IHC-F			*	*	*	*	*		*
CD279 (PD-1)	NAT105	IHC-P		*	*	*	*		*		*
CD309 (VEGFR2)	A16085I	IHC-P		*							
CD324 (E-Cadherin)	67A4	IHC-P, ICC		*	*			*	*	*	*
CD326 (Ep-CAM)	9C4	IHC-P, ICC		*	*	*	*	*	*	*	*
EGFR	AY13	IHC-P, ICC		*		*			**	*	*
Galectin-9	9M1-3	IHC-P		*		*			*	*	*
Ki-67	Ki-67	IHC-F, ICC		*		*		*	*	*	*
PCNA	PC10	IHC-P, ICC	IHC-F	*	*				*	*	*
Podoplanin	NC-08	ICC		*	*				*	*	*
SSEA-3	MC-631	ICC		*	*			*	*	*	*

# IHC and ICC Antibodies

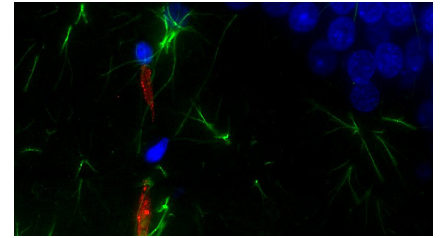
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Paraffin-embedded human cerebellum tissue slices were stained with anti-GFAP Antibody (clone 2E1.E9) Brilliant Violet 421™ (blue) and DRAQ5™.



Formalin-fixed paraffin-embedded mouse brain tissue stained with anti-IP3R1 (clone L24/18) and Ultra-Streptavidin (USA) HRP kit (Multi-species, DAB), followed by hematoxylin staining.



Formalin-fixed paraffin-embedded rat brain tissue co-stained with Alexa Fluor® 594 anti-Rat Blood-Brain Barrier (clone SMI 71, red) and Alexa Fluor® 488 anti-GFAP (clone SMI 25, green) antibodies and counterstained with DAPI (blue).

## Neuroscience

Antibody Specificity	Clone	Validated Applications	Reported Applications	BioLegend Formats Available					
				Purified	HRP	Biotin	AF488	AF594	AF647
α-Synuclein, aggregated	Syn-O2	IHC-P		•					
β-Amyloid, 1-15	3A1	IHC-F	IHC-P	•					
β-Amyloid, 1-16	6E10	IHC-P		•	•	•	•		
β-Amyloid, 17-24	4G8	IHC-P		•	•	•	•	•	•
ATG5	177.19	IHC-P, ICC		•					
Clusterin	A15113A	IHC-P							
Cx3CR1	8E10.D9	IHC-P		•		•			
GFAP	SMI 25	IHC-P		•	•		•	•	
KCC2	N1/66	IHC-P		•					
LC3	A15143K	IHC-P, ICC		•					
MAP2	SMI 52	IHC-P	ICC	•	•		•	•	•
MMP-9	L51/82	IHC-P		•	•				
Myelin CNPase	SMI 91	IHC-P		•	•				
Nicastrin	9C3	IHC-P		•					
Neurofilament H (NF-H), Nonphosphorylated	SMI 32	IHC-P, IHC-F, ICC	Array Tomography	•	•		•	•	
Neurofilament H (NF-H), Phosphorylated	SMI 31	IHC-P, ICC		•	•	•		•	
Neurogranin	NG2	IHC-P		•					
NSE	NSE-P1	IHC-P, ICC		•			•	•	•
P2RY12	S16007D	IHC-P, ICC		•					
Pan-Shank	N23B/49	IHC-P	ICC	•					
Rab7A	W16034A	IHC-P, ICC		•				•	
Rat Blood-Brain Barrier	SMI 71	IHC-P		•		•		•	
Synaptotagmin-12	N277/7	IHC-P		•					
Tau, 1-100	43D	IHC-P		•		•	•	•	•
Tau, 368-441	A16097F	IHC-P		•					
Tau Phospho (Ser262)	A15091A	IHC-P		•					
Tubulin beta 3	TUJ1	IHC-P, ICC		•	•		•	•	•
Tyrosine Hydroxylase	2/40/15	IHC-P		•					

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