



ATLAS ANTIBODIES IN COLORECTAL CANCER RESEARCH

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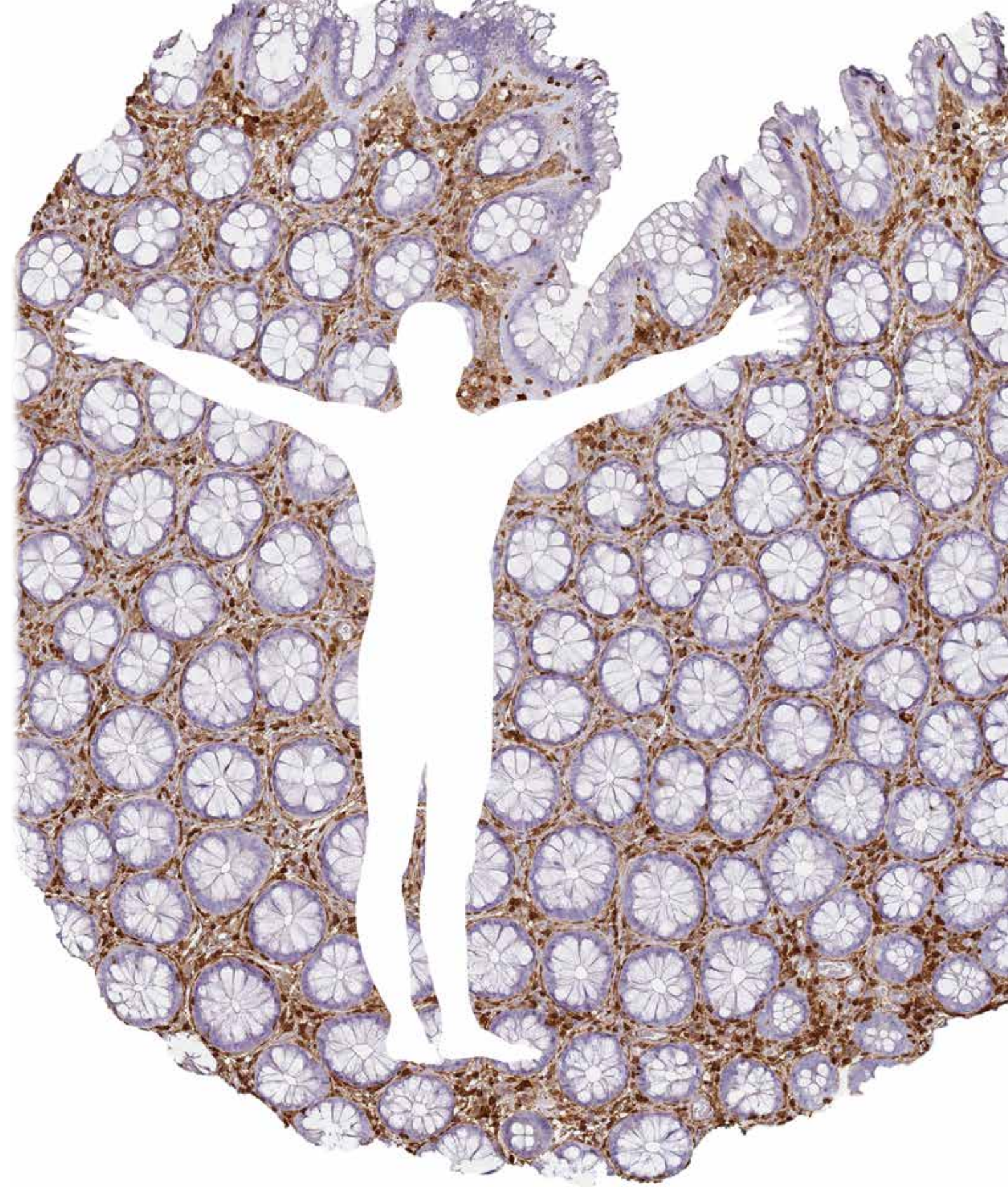
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THE HUMAN PROTEIN ATLAS



proteinaatlas.org

The Human Protein Atlas is Characterizing the Human Proteome

The Human Protein Atlas project has created a complete map of protein expression in all major organs and tissues in the human body^{1,2}. To accomplish this, highly specific antibodies have been developed to all protein coding human genes and protein profiling is established in a multitude of tissues and cells using tissue arrays. Applications applied are immunohistochemistry (IHC), Western blot (WB) analysis, protein array assay and immunofluorescent based confocal microscopy (ICC-IF).

The antibodies developed within the Human Protein Atlas project are carefully designed and manufactured to achieve the very highest level of specificity, reproducibility and versatility. You will find them in our catalog as Triple A Polyclonals.

The Human Protein Atlas (HPA) project was initiated in 2003 by Swedish researchers, headed by Professor Mathias Uhlen, and funded by the Knut and Alice Wallenberg foundation. It is a unique world leading effort performing systematic exploration of the human proteome using antibodies.

The Human Protein Atlas is divided into three major parts, the Tissue Atlas, Cell Atlas and Cancer Atlas. In different ways, the atlases show gene and protein expression data and make it easy to access, search and navigate.

The Tissue Atlas

For all proteins represented in the Tissue Atlas, the expression profiles are based on IHC analysis on a large number of human tissues. All IHC images can be viewed in high resolution on the Tissue Atlas. The presentation of protein expression data in correlation to RNA sequencing data for each gene is included.

Tissue microarrays containing samples from 44 different normal human tissues and from 20 different cancer types are utilized within the project. The 44 normal tissues are present in triplicate samples and annotated in 76 different cell types. All normal tissue images have undergone pathology-based annotation of expression levels and are displayed on the normal Tissue Atlas presenting information regarding the expression profiles of human genes both on mRNA and protein level. The mRNA expression data is derived from deep sequencing of RNA (RNASeq) from 37 major different normal tissue types.

The Cell Atlas

The Cell Atlas presents subcellular localization by confocal microscopy. The results are displayed as high resolution, multicolor images of immunofluorescently stained cells. Three human cell lines for each antibody are selected for the immunofluorescence analysis. Two cell lines from a cell line panel are chosen based on RNA sequencing data and the third cell line is always U-2 OS.

The Cancer Atlas

The Cancer Atlas contains gene expression data based on protein expression patterns in a multitude of human cancer specimens. Altogether 216 different cancer samples, corresponding to the 20 most common forms of human cancer, have been analyzed for all included genes. All cancer tissue images have been manually annotated by pathologists and just as for the normal Tissue Atlas, protein data includes protein expression levels corresponding to over 15,000 genes for which there are available antibodies.

Validation in Normal GI tract Tissue and Colorectal Cancer Tissue Samples

IHC images from normal gastrointestinal tract samples from three different individuals are available for each antibody in the normal Tissue Atlas. The gastrointestinal tract tissues include colon, small intestine, duodenum, rectum, stomach, esophagus, salivary gland and oral mucosa.

In addition, for each antibody, colorectal tumor samples from up to 12 patients in duplicates are presented in the Cancer Atlas.

References:

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Triple A Polyclonals

Triple A Polyclonals - the Building Blocks of HPA

The uniqueness and specificity of Triple A Polyclonals are due to a thorough selection of antigen regions, affinity purification on the recombinant antigen, validation using several methods and a stringent approval process.

Development

The Triple A Polyclonals are developed against recombinant human Protein Epitope Signature Tags (PrESTs) of approximately 50 to 150 amino acids. These protein fragments are designed, using a proprietary software, to contain unique epitopes present in the native protein suitable for triggering the generation of antibodies

of high specificity. This is achieved by a complete human genome scanning to ensure that PrESTs with the lowest homology to other human proteins are used as antigens.

Approval

The approval of the Triple A Polyclonals relies on a combined validation of the experimental results using IHC, WB or ICC-IF, from RNA sequencing and from information obtained via bioinformatics prediction methods and literature. Since the literature is often inconclusive, an important objective of the HPA project has been to generate paired antibodies with non-overlapping epitopes towards the same protein target, allowing the results and validation of one

antibody to be used to validate the other one.

Triple A Polyclonal catalog

Today, there are more than 21,000 Triple A Polyclonals.

The antibodies developed and characterized within the Human Protein Atlas project are made available to the scientific community by Atlas Antibodies under the brand name Triple A Polyclonals. The product numbers of Triple A Polyclonals start with "HPA".

PrecisA Monoclonals

Atlas Antibodies also provides a selected number of mouse monoclonal antibodies, under the brand name PrecisA Monoclonals. The PrecisA Monoclonal catalog is regularly expanding with hundreds of new products every year.

Unique Features

Special care is taken in offering clones recognizing only unique non-overlapping epitopes and/or isotypes. Using the same stringent PrEST production process and characterization procedure as for the Triple A Polyclonals, the PrecisA Monoclonals offer outstanding performance in approved applications, together with defined specificity, secured continuity and stable supply. In general they also permit high working dilutions and contribute to standardized assay procedures.

Clone Selection

Functional characterization is performed on a large number of ELISA positive cell supernatants to select the optimal clones for each application prior to subcloning and expansion of selected hybridomas.

Epitope Mapping

Clones are epitope-mapped using synthetic overlapping peptides in a bead-based array format for selection of clones with non-overlapping epitopes only.

Isotyping

All PrecisA Monoclonals antibodies are isotyped to allow for multiplexing using isotype-specific secondary antibodies.

Hybridoma Cell Cultivation

Atlas Antibodies use in-vitro methods for the production scale-up phase.

Antibody Characterization

The characterization of PrecisA Monoclonals starts with a thorough selection of the most relevant and clinically significant tissues to use for IHC characterization. In addition to positive stained tissues, a negative control tissue staining is also displayed and if relevant, clinical cancer tissue staining.

The Western blot (WB) characterization includes results from endogenous human cell or tissue protein

lysates or optionally recombinant full-length human protein lysates.

Each PrecisA Monoclonal is thus supplied with the most relevant characterization data for its specific target.

PrecisA Monoclonals are developed by Atlas Antibodies, based on the knowledge from the Human Protein Atlas with careful antigen design and extended validation of antibody performance. With precise epitope information following all monoclonals, these precise, accurate and targeted antibodies are denoted PrecisA Monoclonals. The product numbers of PrecisA Monoclonals start with "AMAb".

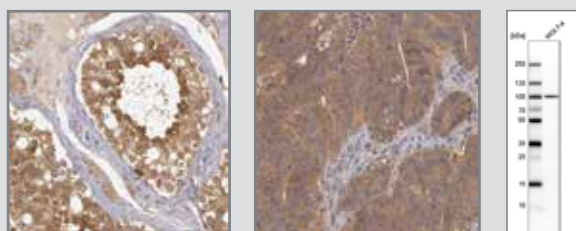


Antibodies Used In Colorectal Cancer Research

In this section, antibodies are selected either on a reference/article-basis or on colon cancer relevance for the corresponding target protein.

Target Protein	Product Name	Product Number	Validated Applications
ABCB1/CD243	Anti-ABCB1	HPA002199 ¹⁻²	IHC,ICC-IF
ALCAM/CD166 antigen	Anti-ALCAM	HPA010926 ³⁻⁵	IHC
AOC3/HPAO	ANTI-AOC3	HPA000980 ⁶⁻⁸	IHC,WB
APC	Anti-APC	HPA013349	IHC
AXL	Anti-AXL	HPA037422 ⁹⁻¹⁰	IHC,WB
B-Raf	Anti-BRAF	HPA001328 ¹¹⁻¹³	IHC
B-Raf	Anti-BRAF	HPA071048	ICC-IF,WB
B-Raf	Anti-BRAF	AMAb91257	IHC,WB
B-Raf	Anti-BRAF	AMAb91258	IHC,WB
BCL9	Anti-BCL9	HPA020274	IHC,ICC-IF
Bloom syndrome prot	Anti-BLM	HPA005689 ¹⁴⁻¹⁵	IHC,ICC-IF
Cadherin-17	Anti-CDH17	HPA023616 ¹⁶	IHC,WB,ICC-IF
Cadherin-17	Anti-CDH17	HPA026556	IHC,WB,ICC-IF
Caldesmon	Anti-CALD1	HPA008066 ¹⁷⁻²⁰	IHC,WB,ICC-IF
Caspase-3	Anti-CASP3	HPA002643 ²¹⁻²²	IHC,WB,ICC-IF
Catenin beta-1	Anti-CTNNB1	HPA029159	IHC,WB,ICC-IF
Catenin beta-1	Anti-CTNNB1	HPA029160	IHC,ICC-IF
Catenin beta-1	Anti-CTNNB1	AMAb91209	IHC,WB
Catenin beta-1	Anti-CTNNB1	AMAb91210	IHC,WB
CDX-2	Anti-CDX2	HPA045669	ICC-IF
CDX-2	Anti-CDX2	HPA049580	ICC-IF
CEACAM1/3/5/6	Anti-CEACAM1	HPA011041	IHC,WB
Chromogranin-A	Anti-CHGA	HPA017369 ²³⁻²⁵	IHC,WB,ICC-IF
Cytokeratin 18	Anti-KRT18	HPA001605	IHC,WB,ICC-IF

Anti-BRAF (HPA001328)



The Anti-BRAF antibody (HPA001328) shows cytoplasmic positivity in cells in seminiferous ducts in normal human testis as well as in tumor cells in colorectal cancer using immunohistochemistry. The HPA001328 antibody detects BRAF in human cell line MOLT-4 lysate using Western Blot analysis.

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Target Protein	Product Name	Product Number	Validated Applications
Cytokeratin 19	Anti-KRT19	HPA002465	IHC,WB,ICC-IF
Cytokeratin 20	Anti-KRT20	HPA024309	IHC,WB
Cytokeratin 20	Anti-KRT20	HPA024684	IHC,WB,ICC-IF
Cytokeratin 20	Anti-KRT20	HPA027236	IHC,WB
Cytokeratin 8	Anti-KRT8	HPA049866	IHC,WB,ICC-IF
DACH1	Anti-DACH1	HPA012672 ²⁶⁻²⁸	IHC,ICC-IF
DCC	Anti-DCC	HPA055376	ICC-IF
DCC	Anti-DCC	HPA069552	IHC
DTL	Anti-DTL	HPA028016 ²⁹	IHC,WB
EGFR	Anti-EGFR	HPA001200 ³⁰	IHC
EGFR	Anti-EGFR	HPA018530 ³¹⁻³²	IHC,ICC-IF
EGFR	Anti-EGFR	AMAb90816	IHC,WB
FCGRT	Anti-FCGRT	HPA012122 ³³⁻³⁵	IHC,WB
Fibronectin	Anti-FN1	HPA027066 ³⁶	IHC,WB
FOXRED1	Anti-FOXRED1	HPA046192 ³⁷	IHC
GDF15	Anti-GDF15	HPA011191 ³⁸⁻⁴²	IHC,WB,ICC-IF
GPA33	Anti-GPA33	HPA018858 ⁴³⁻⁴⁴	IHC
GRHL2	Anti-GRHL2	HPA004820 ⁴⁵⁻⁴⁹	IHC,WB

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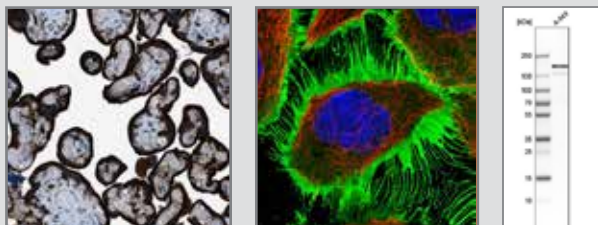
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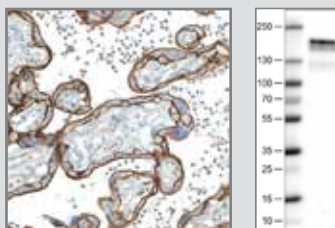
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Anti-EGFR (HPA018530)



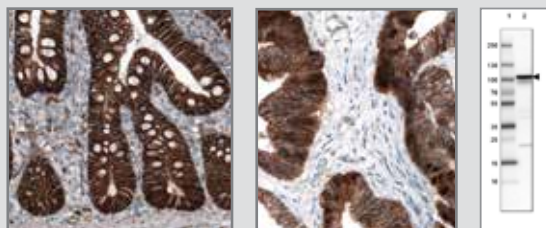
The Anti-EGFR antibody (HPA018530) shows strong cytoplasmic positivity in trophoblastic cells in human placenta tissue using IHC. By ICC-IF, the antibody shows strong positivity in plasma membrane in human cell line A-431 and in Western blot analysis, EGFR is detected in human cell line A-549 lysate.

Anti-EGFR (AMAb90816)



The Anti-EGFR (AMAb90816) shows strong membranous positivity in trophoblasts in human placenta tissue using IHC and by Western Blot analysis, EGFR is detected in human A-431 cell line lysate.

Anti-CTNNB1 (AMAb91210)



IHC staining using the Anti-CTNNB1 (AMAb91210) antibody shows strong membranous immunoreactivity in epithelial cells in normal small intestine and in tumor cells in colorectal cancer tissues. By WB analysis, Catenin beta-1 is detected in human cell line A-431.



Target Protein	Product Name	Product Number	Validated Applications
Guanylin	Anti-GUCA2A	HPA018215 ⁵⁰⁻⁵²	IHC,WB
HMGR	Anti-HMGR	HPA008338 ⁵³⁻⁵⁵	IHC
HRH4	Anti-HRH4	HPA035009 ⁵⁶	IHC
HTRB	Anti-HTRB	HPA012867 ⁵⁷⁻⁵⁹	IHC,ICC-IF
IDH1	Anti-IDH1	HPA035248 ⁶⁰	IHC,WB
IGFBP7/IBP-7	Anti-IGFBP7	HPA002196 ⁶¹⁻⁶³	IHC,WB
Integrin alpha-6	Anti-ITGA6	HPA012696 ⁶⁴⁻⁶⁵	IHC,WB
IRF2BP1	Anti-IRF2BP1	HPA042164 ⁶⁶	IHC,WB
KIT	Anti-KIT	AMAb90901	IHC,WB
KIT	Anti-KIT	AMAb90904	IHC,WB
KIT	Anti-KIT	HPA004471	IHC
KIT	Anti-KIT	HPA073252	ICC-IF

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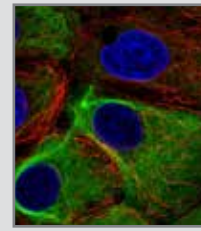
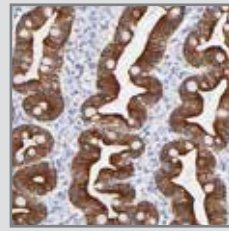
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Anti-KRT19 (HPA002465)



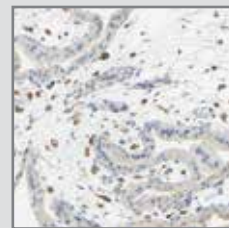
The Anti-KRT19 antibody (HPA002465) shows strong cytoplasmic and membranous positivity in glandular cells in human duodenum tissue using IHC. ICC-IF staining of human cell line MCF7 shows localization to intermediate filaments.

Anti-KRT20 (HPA024309)



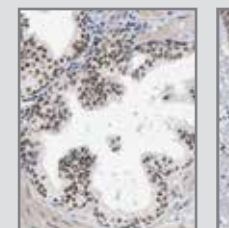
The Anti-KRT20 antibody (HPA024309) shows strong cytoplasmic and membranous positivity in glandular cells in colon tissue using IHC. Cytokeratin 20 is detected using Western blot analysis in small intestine tissue lysate.

Anti-FCGRT (HPA012122)



By IHC, the Anti-FCGRT antibody (HPA012122) shows cytoplasmic positivity in Hofbauer cells in human placenta tissue. In Western blot analysis, FCGRT is detected in human cell line THP1 lysate.

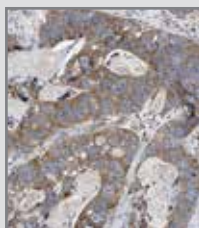
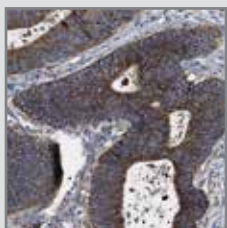
Anti-GRHL2 (HPA004820)



The Anti-GRHL2 antibody (HPA004820) shows strong nuclear positivity in glandular cells in human prostate tissue as well as in squamous epithelial cells in human skin tissue using IHC.

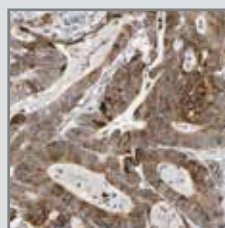
Target Protein	Product Name	Product Number	Validated Applications
KRAS/HRAS/NRAS	Anti-KRAS	HPA049830	IHC
LAMB2/S-LAM beta	Anti-LAMB2	HPA001895 ⁶⁷	IHC,WB
LCN2/NGAL/p25	Anti-LCN2	HPA002695 ⁶⁸⁻⁷⁰	IHC,WB
LGR5	Anti-LGR5	HPA012530 ⁷¹⁻⁷⁴	IHC,WB
LMAN1/ERGIC-53	Anti-LMAN1	HPA002320 ⁷⁵	IHC,WB
LPAR2	Anti-LPAR2	HPA019616 ⁷⁶⁻⁷⁸	IHC,WB
LPAR3	Anti-LPAR3	HPA013421 ⁷⁹	IHC,WB,ICC-IF
MACC1	Anti-MACC1	HPA020103 ⁸⁰⁻⁸¹	IHC,WB
MAP1B	Anti-MAP1B	HPA022275 ⁸²⁻⁸³	IHC,ICC-IF
MCAM/MUC18	Anti-MCAM	HPA008848 ⁸⁴	IHC
MGAT5/GNT-V	Anti-MGAT5	HPA010919	IHC,WB
MLH1/COCA2	Anti-MLH1	HPA052707	IHC,WB,ICC-IF
MLH1/COCA2	Anti-MLH1	HPA060714	ICC-IF
MSH6/GTBP	Anti-MSH6	HPA028376	IHC,WB,ICC-IF
MSH6/GTBP	Anti-MSH6	HPA028446	IHC
MUC3/MUC3A	Anti-MUC3A	HPA010871	IHC
Mucin-1	Anti-MUC1	HPA004179	IHC,WB
Mucin-1	Anti-MUC1	HPA007235	IHC
Mucin-1	Anti-MUC1	HPA008855 ⁸⁵	IHC,ICC-IF
NDRG1	Anti-NDRG1	HPA006881 ⁸⁶⁻⁸⁹	IHC,WB,ICC-IF

Anti-MACC1 (HPA020103)



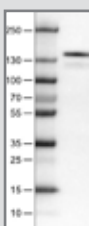
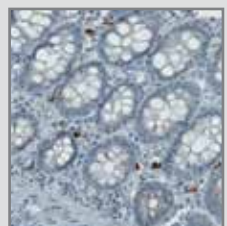
The Anti-MACC1 antibody (HPA020103) shows cytoplasmic positivity in tumor cells in colorectal and stomach cancer using IHC.

Anti-MUC1 (HPA004179)



The Anti-MUC1 antibody (HPA004179) shows cytoplasmic and membranous positivity in glandular cells in normal stomach and in tumor cells in colorectal cancer.

Anti-KIT (AMAb90901)



The Anti-KIT antibody (AMAb90901) shows strong immunoreactivity in a subset of lymphoid cells (macrophages) in colon tissue. In Western blot analysis, KIT is detected in human cell line RT-4 lysate.

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Target Protein	Product Name	Product Number	Validated Applications
Nucleophosmin	Anti-NPM1	HPA011384 ⁹⁰⁻⁹¹	IHC,WB,ICC-IF
P53	Anti-P53 Antibody	AMAb90956 ⁹²	IHC,WB,ICC-IF
PARP6	Anti-PARP6	HPA026991 ⁹³⁻⁹⁴	IHC,ICC-IF
Periostin	Anti-POSTN	HPA012306 ⁹⁵⁻⁹⁶	IHC,WB
PIK3CA	Anti-PIK3CA	HPA009985 ⁹⁷⁻⁹⁹	IHC,ICC-IF
Plexin-B1	Anti-PLXNB1	HPA040586 ¹⁰⁰	IHC
PMS2/PMSL2	Anti-PMS2	HPA070310	ICC-IF
PMS2/PMSL2	Anti-PMS2	HPA066490	ICC-IF
Podocalyxin	Anti-PODXL	HPA002110 ¹⁰¹⁻¹⁰⁵	IHC,WB,ICC-IF
PTP4A1/2/3	Anti-PTP4A1	HPA003281	IHC,WB
RBM3	Anti-RBM3	AMAb90655 ¹⁰⁶⁻¹¹⁰	IHC,WB,ICC-IF
RBM3	Anti-RBM3	HPA003624 ¹¹¹⁻¹¹³	IHC,WB,ICC-IF
RECQL5	Anti-RECQL5	HPA029971 ¹¹⁴⁻¹¹⁶	IHC,WB,ICC-IF
RET	Anti-RET	HPA008356 ¹¹⁷⁻¹¹⁸	IHC,ICC-IF
RIBC2	Anti-RIBC2	HPA003210 ¹¹⁹	IHC
ROBO2	Anti-ROBO2	HPA013371 ¹²⁰	IHC,WB,ICC-IF

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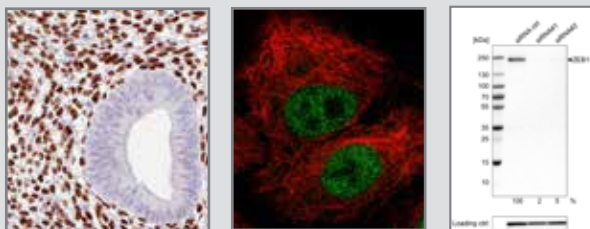
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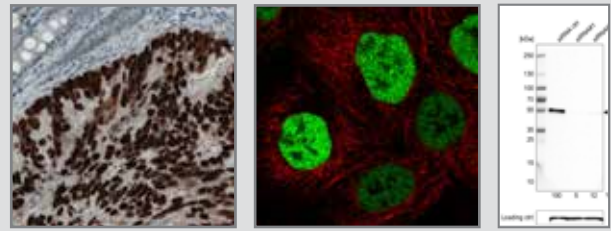
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Anti-ZEB1 (AMAb90510)



The Anti-ZEB1 antibody (AMAb90510) shows strong nuclear positivity in the stromal, but not glandular cells in endometrium tissue using IHC. ICC-IF shows nuclear staining in the human A-549 cell line. The antibody signal is downregulated using ZEB1-specific siRNA probes in extracts from RH-30 cells, shown by WB analysis.

ANTI-P53 (AMAb90956)



The Anti-P53 antibody shows strong nuclear immunoreactivity in tumor cells in human colorectal cancer using IHC. ICC-IF staining in A431 cell line shows cell cycle dependent nuclear (without nucleoli) staining. The antibody signal is downregulated using p53-specific siRNA probes in extracts from U-251 cells, shown by WB analysis.

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Target Protein	Product Name	Product Number	Validated Applications
SATB2	Anti-SATB2	HPA029543 ¹²¹	IHC, ICC-IF
SATB2	Anti-SATB2	AMAb90679	IHC, WB
Semaphorin-4D	Anti-SEMA4D	HPA015662	IHC, WB
Serpin A1	Anti-SERPINA1	HPA000927	IHC, WB, ICC-IF
SIX1	Anti-SIX1	HPA001893	IHC, WB, ICC-IF
SORD	Anti-SORD	HPA040260	IHC, WB, ICC-IF
SOX9	Anti-SOX9	HPA001758	IHC, WB, ICC-IF
SOX21	Anti-SOX21	AMAb91309	IHC, WB
SOX21	Anti-SOX21	AMAb91311	IHC, WB
SRC	Anti-SRC	HPA030875	IHC, WB, ICC-IF
SRSF5	Anti-SRSF5	HPA043484 ¹²²	IHC, WB, ICC-IF
SRSF6	Anti-SRSF6	HPA029005 ¹²³	IHC, WB
SRSF7	Anti-SRSF7	HPA043850 ¹²⁴	IHC, WB, ICC-IF
STK4/MST-1	Anti-STK4	HPA015270 ¹²⁵	IHC, WB, ICC-IF
SUSD2	Anti-SUSD2	HPA004117 ¹²⁶⁻¹²⁷	IHC, ICC-IF
TAK1/TR4/NR2C2	Anti-NR2C2	HPA006313	IHC, WB, ICC-IF
TET1	Anti-TET1	HPA019032 ¹²⁸⁻¹²⁹	IHC, ICC-IF
TFAP4	Anti-TFAP4	HPA001912 ¹³⁰	IHC, WB, ICC-IF
TGFB1	Anti-TGFB1	HPA008612 ¹³¹⁻¹³³	IHC, WB
TIMP1/EPA	Anti-TIMP1	HPA053417	IHC
TJP1	Anti-TJP1	HPA001636 ¹³⁴⁻¹³⁵	IHC, WB, ICC-IF
TNIK	Anti-TNIK	HPA012128 ¹³⁶⁻¹³⁷	IHC, ICC-IF
TRAF6	Anti-TRAF6	HPA019805	IHC, WB, ICC-IF
TRAF6	Anti-TRAF6	HPA020599	IHC, WB
Transgelin	Anti-TAGLN	HPA019467 ¹³⁸	IHC, WB, ICC-IF
Willin/FRMD6	Anti-FRMD6	HPA001297 ¹³⁹	IHC, WB, ICC-IF
YWHAB/KCIP-1	Anti-YWHAB	HPA011212 ¹⁴⁰	IHC, WB, ICC-IF
ZEB1/TCF-8	Anti-ZEB1	HPA027524 ¹⁴¹⁻¹⁴⁵	IHC, WB, ICC-IF
ZEB1/TCF-8	Anti-ZEB1	AMAb90510	IHC, WB, ICC-IF
Zyxin	Anti-ZYX	HPA004835 ¹⁴⁶	IHC, WB, ICC-IF

121. Nodin B *et al.* Molecular correlates and prognostic significance of SATB1 expression in colorectal cancer. *Diagn Pathol* 2012 Aug 30; 7:115. Epub 2012 Aug 30.

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141. Isella C *et al.* Stromal contribution to the colorectal cancer transcriptome. *Nature Genetics* February 23, 2015.

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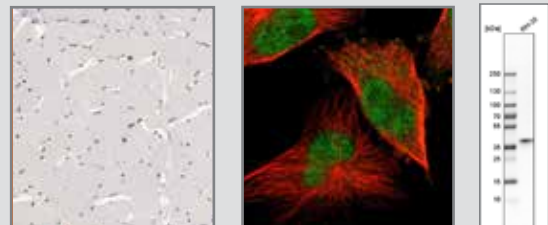
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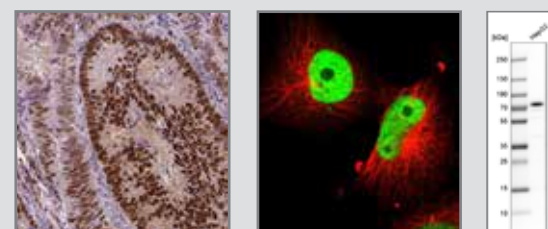
146. Fukumoto M *et al.* α-Actinin-4 Enhances Colorectal Cancer Cell Invasion by Suppressing Focal Adhesion Maturation. *PLoS One* 2015; 10(4):e0120616. Epub 2015 Apr 10.

ANTI-SIX1 (HPA001893)



The Anti-SIX1 antibody (HPA001893) shows strong nuclear positivity in striated muscle fibers in human skeletal muscle using IHC. ICC-IF staining of human cell line U-251 MG shows positivity in nucleus but excluded from the nucleoli. Western blot analysis detects SIX1 in human cell line RH-30.

ANTI-SOX9 (HPA001758)



Anti-SOX9 (HPA001758) shows moderate to strong nuclear positivity in tumor cells in human colorectal cancer using IHC. ICC-IF staining of human cell line U-251 MG shows positivity in nucleus but excluded from the nucleoli. Western blot analysis detects SOX9 in human cell line HepG2.

Antibodies against gene products in Oncotype and Coloprint tests

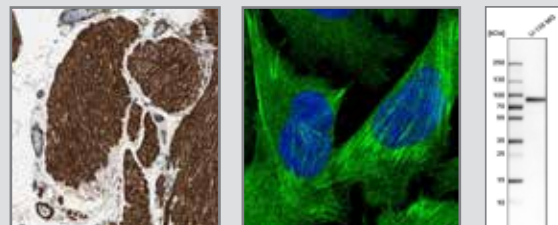
This section presents antibodies in Atlas Antibodies' product catalog against gene products included in the diagnostic Oncotype and Coloprint tests.

Oncotype DX (developed by Genomic Health) is the most frequently used gene expression profile in clinical practice in the United States analyzing a panel of 21 genes within a tumor to determine a Recurrence Score.

Target Protein	Product Name	Product Number	Validated Applications
AKAP12	Anti-AKAP12	HPA006344 ¹	IHC,ICC-IF
AKAP12	Anti-AKAP12	HPA056230	IHC,ICC-IF
AKT3	Anti-AKT3	HPA026441 ²⁻⁴	IHC,WB
Caldesmon	Anti-CALD1	HPA008066 ⁵⁻⁸	IHC,WB,ICC-IF
Caldesmon	Anti-CALD1	HPA017330 ⁹⁻¹¹	IHC,WB,ICC-IF
ANTXR1	Anti-ANTXR1	HPA052046	IHC
Biglycan	Anti-BGN	HPA003157 ¹²⁻¹³	IHC,WB,ICC-IF
Collagen alpha-1	Anti-COL1A1	HPA008405	IHC
Collagen alpha-1	Anti-COL1A1	HPA011795	IHC,ICC-IF
SPARC	Anti-SPARC	HPA002989 ¹⁴	IHC,WB
SPARC	Anti-SPARC	HPA003020 ¹⁵	IHC,WB
CTHRC1/NMTC1	Anti-CTHRC1	HPA059806	IHC,WB
FAP	Anti-FAP	HPA059739	IHC
Inhibin beta A chain	Anti-INHBA	HPA020031 ¹⁶⁻¹⁸	IHC
LOXL2	Anti-LOXL2	HPA036257	ICC-IF
LOXL2	Anti-LOXL2	HPA056542	ICC-IF
TGFB3	Anti-TGFB3	HPA063582	ICC-IF
PDGFC	Anti-PDGFC	HPA009134	IHC,ICC-IF
IGFBP7	Anti-IGFBP7	HPA002196 ¹⁹⁻²¹	IHC,WB
SFRP4	Anti-SFRP4	HPA009712	IHC,WB
SFRP4	Anti-SFRP4	HPA050585	IHC,WB
DLC1	Anti-DLC1	HPA017753 ²²	IHC,WB,ICC-IF
EGR1	Anti-EGR1	HPA029938	ICC-IF
EGR1	Anti-EGR1	HPA029937	ICC-IF
GADD45B	Anti-GADD45B	HPA029816 ²³	IHC,ICC-IF
SERPINE1/PAI1	Anti-SERPINE1	HPA050039 ²⁴	IHC,ICC-IF
SPP1/OPN	Anti-SPP1	HPA027541 ²⁵	IHC,WB
S100A4	Anti-S100A4	HPA007973 ²⁶⁻²⁷	IHC,WB

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Anti-CALD1 (HPA008066)



IHC staining of human smooth muscle tissue using the Anti-CALD1 antibody (HPA008066) shows cytoplasmic positivity in smooth muscle cells. Using ICC-IF in U2-OS cells, the antibody stains actin filament and plasma membrane. CALD1 is detected in cell line U-138 MG cell line.

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Target Protein	Product Name	Product Number	Validated Applications
S100A4	Anti-S100A4	AMAb90596	IHC,WB,ICC-IF
S100A4	Anti-S100A4	AMAb90598	IHC,WB,ICC-IF
S100A4	Anti-S100A4	AMAb90599	IHC,WB,ICC-IF
HSPA1A	Anti-HSPA1A	HPA052504	IHC,WB
TGFBI	Anti-TGFBI	HPA008612 ²⁸⁻³⁰	IHC,WB
TGFBI	Anti-TGFBI	HPA017019	IHC,WB
GRB10	Anti-GRB10	HPA027502	IHC
LAMC2	Anti-LAMC2	HPA024638	IHC
LAMC2	Anti-LAMC2	AMAb91098	IHC,WB
CDKN2A/P14ARF	Anti-CDKN2A	HPA047838	ICC-IF
CDC20	Anti-CDC20	HPA055288	IHC,WB
CDC20	Anti-CDC20	HPA045842	ICC-IF
Ki-67/MKI67	Anti-MKI67	AMAb90870	IHC
Ki-67/MKI67	Anti-MKI67	HPA000451 ³¹⁻³²	IHC,ICC-IF
Ki-67/MKI67	Anti-MKI67	HPA001164 ³³	IHC,ICC-IF
MCM2	Anti-MCM2	HPA031495	IHC,WB,ICC-IF
MCM2	Anti-MCM2	HPA031496	IHC,WB,ICC-IF
RRM1	Anti-RRM1	HPA057265	IHC,ICC-IF
RRM1	Anti-RRM1	HPA064297	IHC
RRM2	Anti-RRM2	HPA056994	IHC,WB,ICC-IF
SKP2	Anti-SKP2	HPA051196	WB,ICC-IF
SKP2	Anti-SKP2	HPA054633	IHC,WB
MYC/CMYC	Anti-MYC	HPA055893	IHC,ICC-IF
MYC/CMYC	Anti-MYC	HPA066556	ICC-IF
CSEL1/CSE1L	Anti-CSE1L	HPA038059	IHC,WB,ICC-IF
CSEL1/CSE1L	Anti-CSE1L	HPA038060	IHC,ICC-IF
MYBL2	Anti-MYBL2	HPA030530	IHC,WB
MYBL2	Anti-MYBL2	HPA055416	ICC-IF
NME1/GAAD	Anti-NME1	HPA008467 ³⁴	IHC,WB,ICC-IF
NME1/GAAD	Anti-NME1	HPA041113	IHC,WB
UMPS/OPRT	Anti-UMPS	HPA036178	IHC,WB,ICC-IF
UMPS/OPRT	Anti-UMPS	HPA036179	IHC
HNRPD	Anti-HNRPD	HPA004911	IHC,WB,ICC-IF
MCTP1	Anti-MCTP1	HPA019018	IHC,WB,ICC-IF
LAMA3	Anti-LAMA3	HPA009309	IHC
LAMA3	Anti-LAMA3	AMAb91123	IHC,WB
CTSC/Cathepsin C	Anti-CTSC	HPA066610	WB,ICC-IF
PYROXD1	Anti-PYROXD1	HPA038319	IHC,WB
PYROXD1	Anti-PYROXD1	HPA038320	IHC,WB,ICC-IF
EDEM1	Anti-EDEM1	HPA029565	IHC,ICC-IF
IL2RB/CD122	Anti-IL2RB	HPA062657	IHC,WB
ZNF697	Anti-ZNF697	HPA049933	IHC,ICC-IF
SLC6A11/GAT-3	Anti-SLC6A11	HPA037981	IHC,WB
IL2RA/CD25	Anti-IL2RA	HPA054622	IHC
CYFIP2	Anti-CYFIP1	HPA068106	IHC,WB
PIM3	Anti-PIM3	HPA068758	ICC-IF
LIF	Anti-LIF	HPA018844	IHC,ICC-IF
Perilipin-3/PLIN3	Anti-PLIN3	HPA006427 ³⁵	IHC,WB,ICC-IF
Perilipin-3/PLIN3	Anti-PLIN3	HPA066538	IHC,WB,ICC-IF
HSD3B1	Anti-HSD3B1	HPA043261	IHC
HSD3B1	Anti-HSD3B1	HPA043264	IHC
HSD3B1	Anti-HSD3B1	HPA044028	IHC
ZBED4	Anti-ZBED4	HPA045341	IHC,ICC-IF
PPARA	Anti-PPARA	HPA067049	WB,ICC-IF
THNSL2	Anti-THNSL2	HPA035395	IHC

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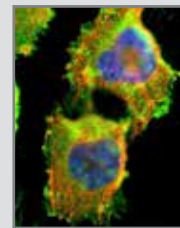
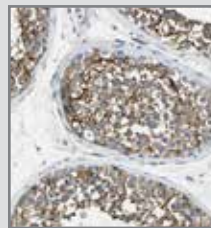
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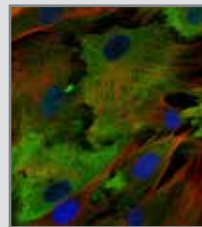
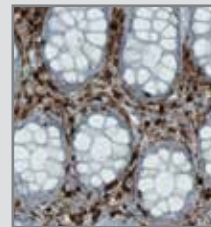
35. Akil A *et al.* Septin 9 induces lipid droplets growth by a phosphatidylinositol-5-phosphate and microtubule-dependent mechanism hijacked by HCV. *Nat Commun* 2016 Jul 15; 7:12203. Epub 2016 Jul 15.

Anti-AKAP12 (HPA006344)



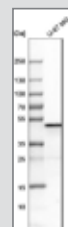
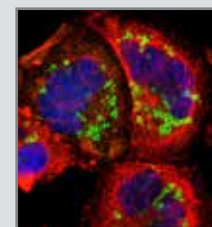
IHC staining of human testis tissue using the Anti-AKAP12 antibody (HPA006344) shows cytoplasmic and membranous positivity in seminiferous ducts. ICC-IF staining of human cell line U-251 MG shows localization to plasma membrane and cytosol.

Anti-S100A4 (AMAb90598)



IHC staining of human rectum tissue using the Anti-S100A4 antibody (AMAb90598) shows strong immunoreactivity in a subset of lymphoid cells. In ICC-IF, in BJ cells, plasma membrane is stained.

Anti-PLIN3 (HPA006427)



IHC staining of human small intestine tissue using the Anti-PLIN3 antibody (HPA006427) shows positivity in glandular cells. ICC-IF in A-431 cell line shows positivity in lipid droplets. PLIN3 is detected in cell line U-87 MG lysate using WB analysis.

Antibodies against gene products elevated in colon identified in the Human Protein Atlas

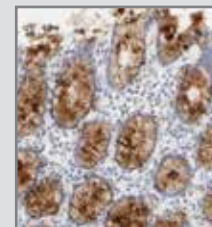
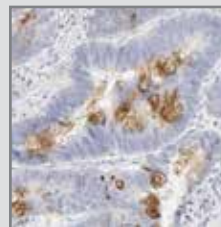
The genes included in this section show at least 5-fold higher mRNA levels in colon tissue compared to all other human tissues. Antibodies against these gene products are presented.



Target Protein	Product Name	Product Number	Validated Applications
B3GNT6	Anti-B3GNT6	HPA039805	IHC
C10orf99	Anti-C10orf99	HPA050920	IHC
CA1	Anti-CA1	HPA006558	IHC
CA2	Anti-CA2	HPA001550 ¹	IHC,WB
CD177	Anti-CD177	HPA041820	IHC
CD177	Anti-CD177	HPA046601	IHC
CDH17	Anti-CDH17	HPA023614	IHC,WB
CDH17	Anti-CDH17	HPA023616 ²	IHC,WB,ICC-IF
CDH17	Anti-CDH17	HPA026556	IHC,WB,ICC-IF
CDHR5	Anti-CDHR5	HPA009081	IHC
CDHR5	Anti-CDHR5	HPA009173	IHC,WB
CDX1	Anti-CDX1	HPA055196	IHC
CDX2	Anti-CDX2	HPA045669	ICC-IF
CDX2	Anti-CDX2	HPA049580	ICC-IF
CEACAM1/3/5/6	Anti-CEACAM1	HPA011041	IHC,WB
CEACAM5	Anti-CEACAM5	HPA019758	IHC,WB,ICC-IF
CEACAM7	Anti-CEACAM7	HPA069621	IHC
Claudin-3	Anti-CLDN3	HPA014361	IHC,ICC-IF
CLCA1	Anti-CLCA1	HPA052787	IHC
CLCA1	Anti-CLCA1	HPA059301	IHC
DHRS11	Anti-DHRS11	HPA041226	IHC,ICC-IF
DHRS11	Anti-DHRS11	HPA048236	IHC
DHRS11	Anti-DHRS11	HPA053623	IHC
ENTPD8	Anti-ENTPD8	HPA021509	IHC,ICC-IF
FABP1	Anti-FABP1	HPA028275	IHC,WB,ICC-IF
FCGBP	Anti-FCGBP	HPA003517 ³	IHC
FCGBP	Anti-FCGBP	HPA003564	IHC,ICC-IF
FUT3/5/6	Anti-FUT3	HPA046966	IHC
FXYD3	Anti-FXYD3	HPA010856 ⁴	IHC,ICC-IF
GAL3ST2	Anti-GAL3ST2	HPA071809	IHC
Galectin-4	Anti-LGALS4	HPA031184	IHC
Galectin-4	Anti-LGALS4	HPA031185	IHC
Galectin-4	Anti-LGALS4	HPA031186	IHC,WB
GPA33	Anti-GPA33	HPA018858	IHC
GUCA2A	Anti-GUCA2A	HPA018215 ⁵⁻⁷	IHC,WB
HNF4A	Anti-HNF4A	HPA004712 ⁸⁻⁹	IHC,WB,ICC-IF
HSD11B2	Anti-HSD11B2	HPA042186	IHC
HSD11B2	Anti-HSD11B2	HPA056385	IHC,ICC-IF
INSL5	Anti-INSL5	HPA030100	IHC,WB
Keratin 20	Anti-KRT20	HPA024309	IHC,WB
Keratin 20	Anti-KRT20	HPA024684	IHC,WB,ICC-IF
Keratin 20	Anti-KRT20	HPA027236	IHC,WB
MEP1A	Anti-MEP1A	HPA029416	IHC
MISP	Anti-MISP	HPA049511	IHC,WB,ICC-IF
MISP	Anti-MISP	HPA062232	IHC,WB,ICC-IF
MS4A12	Anti-MS4A12	HPA057657	IHC
MUC13	Anti-MUC13	HPA045163	IHC,WB
NOXO1	Anti-NOXO1	HPA071540	IHC,WB
NXPE1	Anti-NXPE1	HPA049133	IHC,WB
NXPE2	Anti-NXPE2	HPA039744	IHC

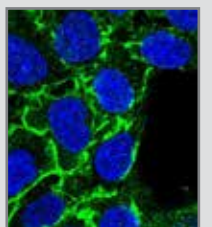
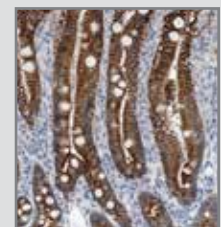
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Anti-FCGBP (HPA003564)



The Anti-FCGBP antibody (HPA003564) shows strong cytoplasmic positivity in glandular cells in human colorectal cancer and normal rectum tissue using IHC.

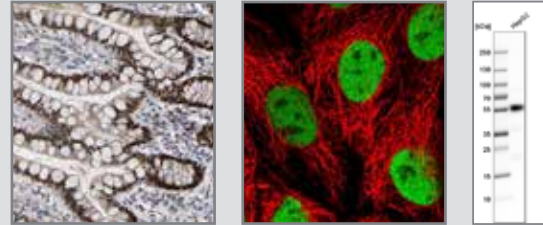
Anti-CDH17 (HPA023614)



The Anti-CDH17 antibody (HPA023614) shows membranous positivity in glandular cells in human duodenum tissue using IHC. In ICC-IF, CDH17 is localized to cell junctions in human cell line CACO-2.

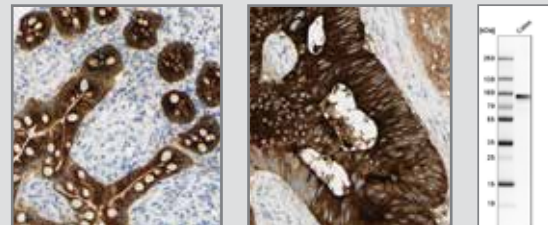
Target Protein	Product Name	Product Number	Validated Applications
NXPE2	Anti-NXPE2	HPA039876	IHC
PADI2	Anti-PADI2	HPA047735	IHC,WB
PHGR1	Anti-PHGR1	HPA068787	IHC,ICC-IF
PIGR	Anti-PIGR	HPA006154	IHC
PIGR	Anti-PIGR	HPA012012 ¹⁰⁻¹³	IHC,WB
PYY	Anti-PYY	HPA010973	IHC
REG4	Anti-REG4	HPA046555	IHC
SATB2	Anti-SATB2	HPA001042 ¹⁴⁻¹⁷	IHC
SATB2	Anti-SATB2	HPA029543 ¹⁸	IHC,ICC-IF
SATB2	Anti-SATB2	AMAb90679	IHC,ICC-IF
SLC22A18AS	Anti-SLC22A18AS	HPA068288	IHC,WB,ICC-IF
SLC26A2	Anti-SLC26A2	HPA058090	IHC,WB
SLC9A3	Anti-SLC9A3	HPA036493	IHC,ICC-IF
SLC9A3	Anti-SLC9A3	HPA036669	IHC,WB
SPINK4	Anti-SPINK4	HPA007286	IHC,ICC-IF
SULT1B1	Anti-SULT1B1	HPA002107	IHC,WB,ICC-IF
Tetraspanin 8	Anti-TSPAN8	HPA044337	IHC,ICC-IF
TFF3	Anti-TFF3	HPA035464	IHC,ICC-IF
TPH1	Anti-TPH1	HPA022483	IHC
UGT2B17	Anti-UGT2B4	HPA045108	IHC
VIL1	Anti-VIL1	HPA006884 ¹⁹	IHC,WB,ICC-IF
VIL1	Anti-VIL1	HPA006885 ²⁰	IHC,WB
VIP	Anti-VIP	HPA017324	IHC
ZG16	Anti-ZG16	HPA052066	IHC,WB
ZG16	Anti-ZG16	HPA052512	IHC,WB

Anti-HNF4A (HPA004712)



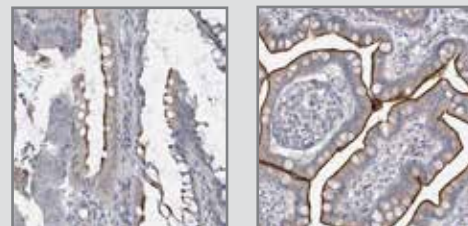
The Anti-HNF4A antibody (HPA004712) shows strong nuclear positivity in glandular cells in human small intestine using IHC. Using ICC-IF, HNF4A was localized to nucleoplasm in cell line CACO-2. HNF4A was detected in human cell line HepG2 using WB analysis.

Anti-PIGR (HPA012012)



The Anti-PIGR antibody (HPA012012) shows strong cytoplasmic and membranous positivity in glandular cells in human colon and in tumor cells in colorectal cancer tissue using IHC. PIGR is detected in colon tissue lysate using Western blot analysis.

Anti-SLC9A3 (HPA036669)



The Anti-SLC9A3 antibody (HPA036669) shows strong apical membrane positivity in glandular cells in human small intestine and in colorectal cancer tissues using immunohistochemistry.

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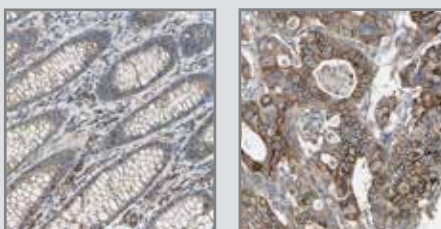
Antibodies identified in the Human Protein Atlas

In this section, antibodies are selected based on identified differential IHC staining patterns in colon and colorectal cancer samples.

Product Name	Product Number	Validated Applications
Anti-ACADSB	HPA041458	IHC,WB,ICC-IF
Anti-ACBD7	HPA043326	IHC,ICC-IF
Anti-ACSL5	HPA007162	IHC,WB,ICC-IF
Anti-ADIRF	HPA026810	IHC,ICC-IF
Anti-AGR3	HPA053942	IHC,ICC-IF
Anti-AJUBA	HPA006171 ¹	IHC,WB,ICC-IF
Anti-ALG14	HPA031829	IHC,ICC-IF
Anti-ANKRD34C	HPA045329	IHC,ICC-IF
Anti-AOAH	HPA021666	IHC,WB
Anti-AQP3	HPA014924	IHC,WB,ICC-IF
Anti-ATF6	HPA005935	IHC
Anti-ATP6V1B2	HPA008147	IHC,WB,ICC-IF
Anti-AVPR2	HPA046678	IHC
Anti-B3GNT8	HPA043669	IHC
Anti-BCL9	HPA020274	IHC,ICC-IF
Anti-CAND2	HPA005777	IHC,ICC-IF
Anti-CCDC144NL	HPA023457	IHC,WB,ICC-IF
Anti-CDH12	HPA029325	IHC
Anti-CDK6	HPA002637	IHC,WB,ICC-IF
Anti-CLDN18	HPA018446	IHC
Anti-COG7	HPA040758	IHC,WB,ICC-IF
Anti-CPE	HPA003545	IHC
Anti-CPE	HPA003819	IHC,WB
Anti-CXorf67	HPA006128	IHC,ICC-IF
Anti-DACH1	HPA012672 ^{2,4}	IHC,ICC-IF
Anti-DEFB115	HPA053160	IHC
Anti-FAM3D	HPA013844	IHC
Anti-FBXW12	HPA037491	IHC
Anti-FKBP7	HPA008707 ⁵	IHC,WB,ICC-IF
Anti-GAA	HPA026970	IHC,WB
Anti-GAK	HPA027463	IHC,ICC-IF
Anti-GALNT6	HPA011762	IHC,WB
Anti-GLB1L3	HPA039916	IHC
Anti-GLDC	HPA002318 ⁶	IHC,WB
Anti-GLUL	HPA007316 ⁷⁻⁹	IHC,WB
Anti-HEPH	HPA005824	IHC,WB
Anti-HLA-E	HPA031454	IHC,ICC-IF
Anti-HMGCS2	HPA027423	IHC,WB
Anti-HMGCS2	HPA027442	IHC,WB,ICC-IF
Anti-HNF4G	HPA005438	IHC
Anti-HPS6	HPA040687	IHC,WB
Anti-IFITM3	HPA004337 ⁹	IHC,WB
Anti-ITGBL1	HPA005676	IHC,WB
Anti-KLHL8	HPA017762	IHC,ICC-IF
Anti-MAGEB1	HPA002820	IHC
Anti-ME2	HPA008247	IHC,WB,ICC-IF
Anti-ME2	HPA008880 ¹⁰⁻¹⁴	IHC,WB

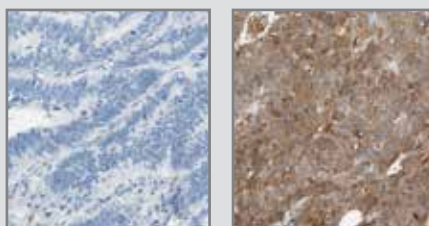
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Anti-HEPH (HPA005824)



Using IHC, the Anti-HEPH antibody (HPA005824) shows cytoplasmic and membranous positivity in glandular cells in normal human colon tissue. In some colorectal cancer samples, prominent membranous positivity could be seen.

Anti-GLUL (HPA007316)

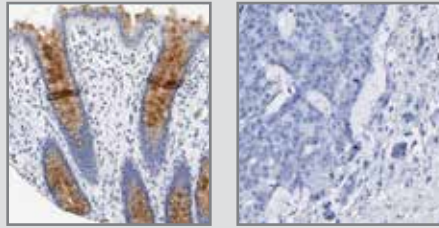


Using IHC, the Anti-GLUL antibody (HPA007316) shows either positive, or negative staining in different colorectal cancer samples.

Product Name	Product Number	Validated Applications
Anti-METTL7B	HPA038644	IHC,WB,ICC-IF
Anti-MRS2	HPA017642	IHC,WB
Anti-MYBBP1A	HPA005466	IHC,WB,ICC-IF
Anti-NAALADL2	HPA012413	IHC
Anti-NCBP3	HPA008959 ¹⁵	IHC,ICC-IF
Anti-OR9K2	HPA015808	IHC
Anti-OSBPL3	HPA000691 ¹⁶	IHC,WB,ICC-IF
Anti-P2RX6	HPA028776	IHC,ICC-IF
Anti-PFKFB2	HPA049975	IHC,ICC-IF
Anti-PHTF2	HPA012312	IHC,ICC-IF
Anti-PITX1	HPA008743	IHC,ICC-IF
Anti-PKN3	HPA045390	IHC
Anti-POMK	HPA013321	IHC,WB,ICC-IF
Anti-PPP1R35	HPA051607	IHC
Anti-PYGB	HPA031067	IHC,WB,ICC-IF
Anti-RAD18	HPA008752	IHC,WB,ICC-IF
Anti-REEP4	HPA042683	IHC,WB
Anti-REG1A	HPA045549	IHC,WB
Anti-RIPPLY2	HPA047454	IHC
Anti-RPS13	HPA005985	IHC,ICC-IF
Anti-S100A4	HPA007973 ¹⁷⁻¹⁸	IHC,WB
Anti-SATB2	HPA001042 ¹⁹⁻²²	IHC
Anti-SOCS7	HPA004475 ²³	IHC,ICC-IF
Anti-SQLE	HPA018038 ²⁴	IHC,WB
Anti-STAG3	HPA049106	IHC
Anti-SYNC	HPA028311	IHC,ICC-IF
Anti-TACC3	HPA005781 ²⁵	IHC,WB
Anti-TBXAS1	HPA031257	IHC
Anti-TBXAS1	HPA031258	IHC
Anti-TBXAS1	HPA031259	IHC,WB
Anti-TGFBI	HPA017019	IHC,WB
Anti-TMEM154	HPA019184	IHC
Anti-TMEM222	HPA016579	IHC,ICC-IF
Anti-TMEM47	HPA046658	IHC,ICC-IF
Anti-TPX2	HPA005487 ²⁶	IHC,WB,ICC-IF

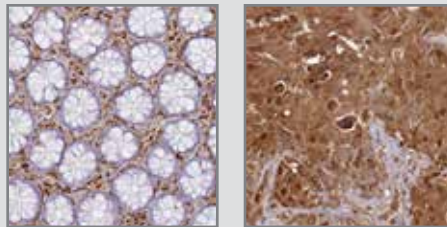


Anti-SOCS7 (HPA004475)



The Anti-SOCS7 antibody (HPA004475) shows positivity in glandular cells in normal human colon tissue, while colorectal cancer samples are negative.

Anti-S100A4 (HPA007973)



The Anti-S100A4 antibody (HPA007973) shows no positivity in glandular cells in normal human colon tissue (left image), while colorectal cancer samples are either positive (right image), or negative.

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Epithelial to Mesenchymal Transition Marker Panel

The EMT Panel

Epithelial and mesenchymal cells are fundamentally different and represent the two main cell types in the body. Epithelial cells are polarised along the apical/basal axis and are tightly connected to each other as well as to underlying basement membrane by a number of cell junction proteins. In contrast, mesenchymal cells are adhered to the extracellular matrix and have enhanced migratory capacities.

Epithelial cells can transition into mesenchymal cells – a process known as epithelial-mesenchymal transition (EMT), which leads to loss of epithelial barrier functions and changes in cell adhesion and motility¹. Normally, EMT occurs during development (embryogenesis), but it is also present in wound healing and cancer progression of epithelial tumors. In metastasis, tumor cells dissociate from the epithelial layer, penetrate through basement membrane into connective tissue and can then enter the vascular system for further dissemination and subsequent growth of distant metastases².

A number of factors drive and regulate the EMT process, including zinc finger proteins such as SNAI1, SNAI2, ZEB1 and ZNF703. These transcription factors down-regulate the expression of

epithelial cell adhesion proteins such as E-cadherin, occludin, beta-catenin and claudin. In addition, they up-regulate expression of mesenchymal proteins, including N-cadherin, fibronectin, vimentin, S100A4 and others. Taken together, EMT leads to increase motility and invasiveness of cancer cells¹.

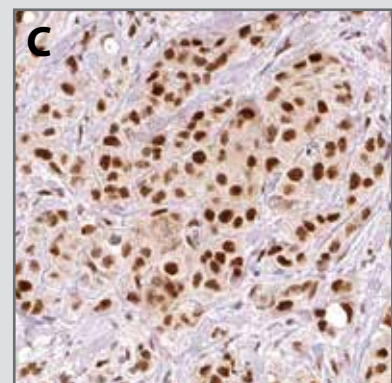
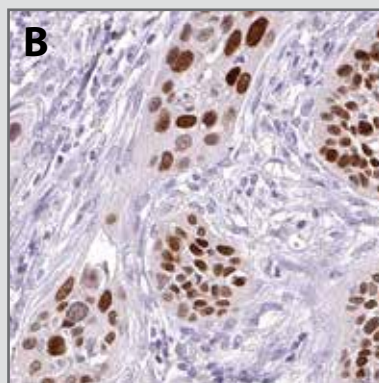
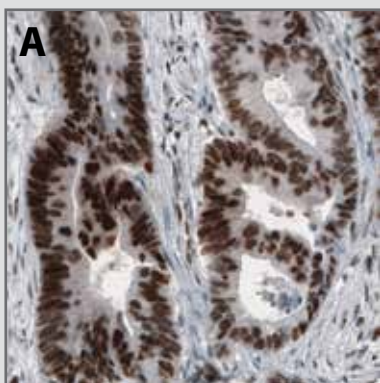
At Atlas Antibodies, we have developed a panel of monoclonal antibodies against the key EMT markers for cell junctions, cytoskeletal changes, transcription regulation and migration/motility. The antibodies targeting selected EMT marker proteins are:

- IHC-validated in relevant normal and cancer human tissues
- WB-validated in positive and negative cell lines (when available)
- Available with different isotypes, allowing for multiplexing experiments
- Supplemented with information on antigens used for immunization and precise epitope sequence (when available)

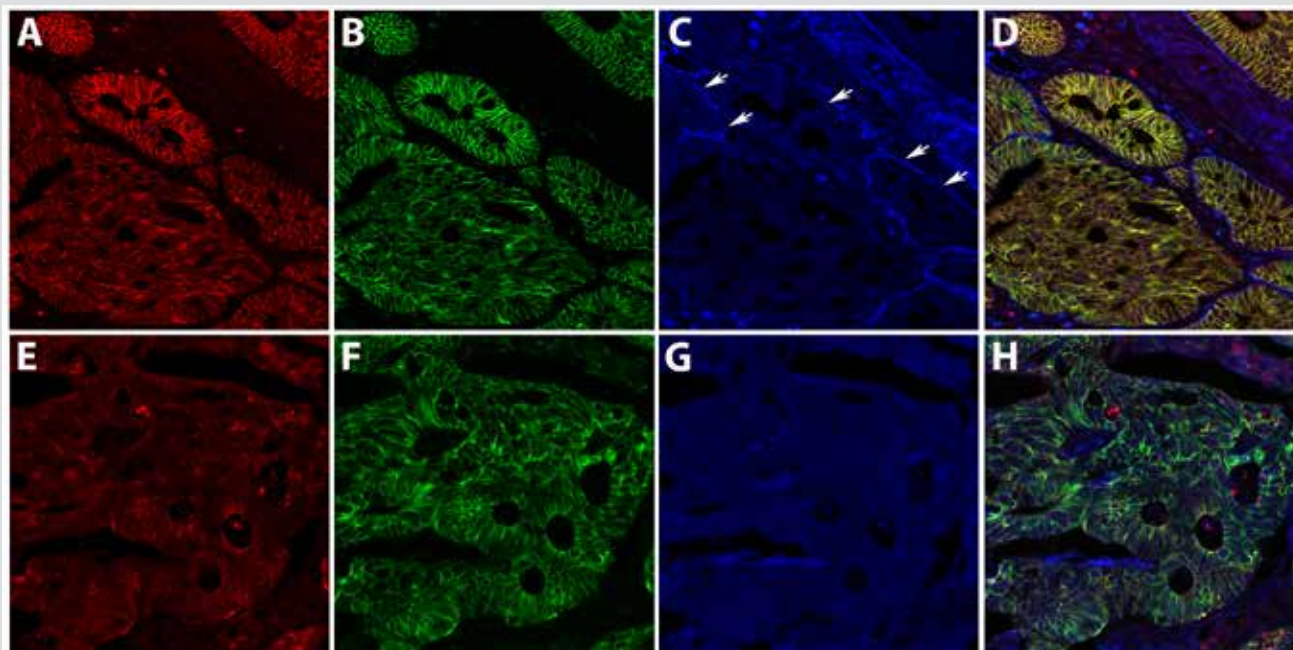
The monoclonal antibodies within the panel have been developed using the same stringent conditions as for all PrecisA Monoclonals, ensuring a secured continuity and stable supply.

Using Monoclonals of Defined Isotypes for Multiplexed Immunofluorescence

The EMT panel includes monoclonal antibodies with different isotypes, which allows for co-localization studies using immunofluorescence with isotype-specific secondary antibodies. The images on the right side show multiplexed staining of colorectal cancer tissue derived from two different patients using the Anti-CDH1 (A, E: AMAb90863, IgG1), Anti-CTNNB1 (B, F: AMAb91209, IgG2a) and LAMC1 (C, G: AMAb91138, IgG2b) monoclonal antibodies, respectively. The tumor with higher degree of differentiation (indicated by preserved basement membrane, C) shows higher expression of E-cadherin (A) as compared to the tumor with lower differentiation grade (E). Also note the absence of LAMC1 immunoreactivity in the second tumor (G). Beta-catenin (CTNNB1) expression is preserved in both tumors (B, F). Panels D and H show overlay images for the two tumors.



Transcription factors involved in regulation of EMT. IHC images show nuclear immunoreactivity in tumor cells in (A) colorectal cancer (Anti-SNAI1 antibody AMAb91215), (B) cervical cancer (Anti-SIX1 antibody AMAb90544) and (C) breast cancer (Anti-ZNF703 AMAb90789).



Multiplexed IHC-IF staining of two colorectal tumors (A-D and E-H) showing E-cadherin (A, E), beta-catenin (B, F) and laminin-gamma 1 (C, G) immunoreactivity using primary antibodies of different isotypes: Anti-CDH1 AMAb90863, IgG1 (red), Anti-CTNNB1 AMAb91209, IgG2a (green) and Anti-LAMC1 (AMAb91138), IgG2b (blue). Arrowheads in C indicate basement membrane. Alexa Fluor® 647-, 594- and 488-labelled isotype-specific secondary antibodies (Life Technologies) were used for visualisation.

Table 1.

Summary of the Precisa Monoclonals EMT Markers.

Marker for	Product Name	Product Number	Validated Applications	Epitope	Isotype
Cell junctions	Anti-CDH1	AMAb90862	IHC, WB	NWTIQYNDPTQESII	IgG2b
Cell junctions	Anti-CDH1	AMAb90863	IHC, WB	APIPEPTIF	IgG1
Cell junctions	Anti-CDH1	AMAb90865	IHC, WB	LKPKMALEVG	IgG2a
Cell junctions	Anti-OCLN	AMAb90889	IHC, WB	TSPVDDFRQPRYSSG	IgG2a
Cell junctions	Anti-OCLN	AMAb90890	IHC, WB	NDKRFYPESSYKSTP	IgG2a
Cell junctions	Anti-OCLN	AMAb90893	IHC, WB	RYSSGGNFETPSKRA	IgG1
Cell junctions	Anti-CTNNB1	AMAb91209	IHC, WB	TSQVLYEWEQGFSQS	IgG2a
Cell junctions	Anti-CTNNB1	AMAb91210	IHC, WB	TSQVLYEWEQGFSQS	IgG1
Cell junctions	Anti-CLDN1	AMAb91213	IHC, WB	KTTSYTPRPYPKPA	IgG1
Cytoskeletal changes	Anti-VIM	AMAb90516	IHC, WB	N.D.	IgG1
Cytoskeletal changes	Anti-S100A4	AMAb90596	IHC, WB	KFKLNKSELKELLTR	IgG1
Cytoskeletal changes	Anti-S100A4	AMAb90598	IHC, WB	CNEFFEGFPDKQPRKK	IgG2b
Cytoskeletal changes	Anti-S100A4	AMAb90599	IHC, WB	CNEFFEGFPD	IgG1
Transcription regulation	Anti-SNAI1	AMAb91215	IHC	N.D.	IgG1
Transcription regulation	Anti-ZEB1	AMAb90510	IHC, WB, ICC	N.D.	IgG1
Transcription regulation	Anti-SIX1	AMAb90544	IHC, WB, ICC	N.D.	IgG1
Transcription regulation	Anti-ZNF703	AMAb90789	IHC, WB	PGDKAGFRVP	IgG1
Transcription regulation	Anti-TP63	AMAb91224	IHC, WB	MQYLPQHTIETYRQQ	IgG1
Migration/Motility	Anti-CDH2	AMAb91220	IHC, WB	ENPYFAPNPK	IgG1
Migration/Motility	Anti-FN1	AMAb91223	IHC, WB	GRWKCDPVVDQ	IgG1
Migration/Motility	Anti-MMP9	AMAb90804	IHC, WB	VPDLGRFQTF	IgG1
Migration/Motility	Anti-MMP9	AMAb90805	IHC, WB	RGESKSLGPALLLLQ	IgG1
Migration/Motility	Anti-MMP9	AMAb90806	IHC	RGESKSLGPALLLLQ	IgG2b

Related Publications

1. Lamouille S *et al.* Molecular mechanisms of epithelial-mesenchymal transition. 2014 *Nat Rev Mol Cell Biol.* 15(3):178-196

2. Chambers AF *et al.* 2002. Dissemination and growth of cancer cells in metastatic sites. *Nat Rev Cancer* 2(8):563-572.

Finding biomarkers for colorectal cancer research

Colorectal Cancer

Colorectal cancer is one of the most common types of cancer. Each year, approximately one million new cases are detected, and approximately 600,000 deaths can be contributed to this disease worldwide. Today, surgery is the only curative treatment for colorectal cancer, but adjuvant treatment may significantly improve patient survival. For adjuvant treatment to be successful, however, it is important to correctly identify patients that will benefit from treatment. For colon cancer, which accounts for approximately 70 % of colorectal cancer cases, adjuvant treatment is currently recommended for patients with stage III and high-

risk stage II disease. For patients with stage II colon cancer, it is thus of utmost importance to find biomarkers that can separate high-risk disease from low-risk disease.

have been identified. These potential biomarkers have subsequently been analyzed in larger patient cohorts, and their prognostic potential evaluated. Below, some of the most promising markers are described briefly.

Colorectal Cancer Biomarkers

Within the Human Protein Atlas (HPA) project, several potential prognostic and diagnostic biomarkers have been discovered. By staining of both normal- and tumor tissue samples, proteins with a tissue specific expression have been identified. Also, proteins with a differential expression in colorectal tissue samples from different patients



RBM3

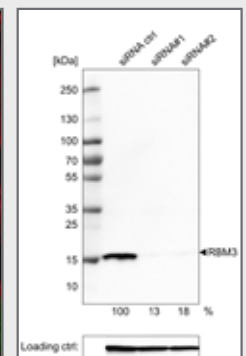
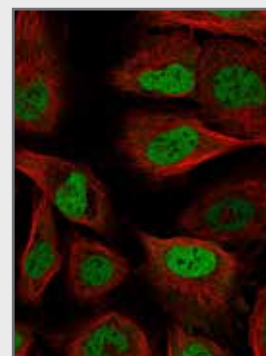
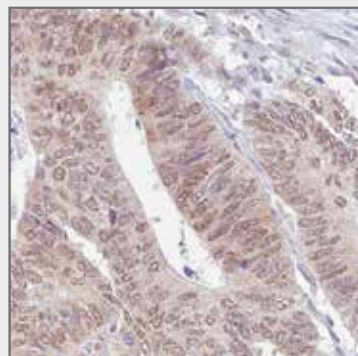
The RNA-binding motif protein 3 (RBM3) is an RNA- and DNA-binding protein, whose function has not been fully elucidated. It has been shown that the protein is expressed as an early event in mild hypothermia, and also in other conditions relating to cellular stress, such as glucose deprivation and hypoxia. During stress, RBM3 is thought to protect the cells by aiding in maintenance of protein synthesis needed for survival. Recently, it has also been shown that RBM3 attenuates stem cell-like properties in prostate cancer cells.

The RNA-binding protein RBM3 was identified via the Human Protein Atlas as an oncology biomarker through the differential expression pattern observed within several investigated cancers.

The levels of RBM3 expression were found to have a significant correlation to patient survival in breast, colon, ovarian, testicular, prostate and urothelial cancer as well as in malignant melanoma.

RBM3 as a prognostic biomarker in colon cancer

RBM3 was shown to be a prognostic marker in colorectal cancer in two independent patient cohorts, with



The Anti-RBM3 (AMAb90655) antibody shows nuclear positivity by IHC in cancer cells in human colorectal tumor samples and nuclear staining by ICC-IF in U2-OS cells. By WB, the AMAb90655 antibody signal is down regulated using target specific siRNA probes in U-251 cells.

a significantly improved survival for patients with high levels of RBM3 expression in their tumors. When analyzing stage II patients separately, similar results were obtained.

This indicates that RBM3 may be used as a biomarker for aid in deciding which stage II patients would benefit from adjuvant treatment.

RBM3 as a treatment predictive biomarker

The RBM3 protein has also been shown to be a treatment predictive marker for platinum based treatment. Chemotherapy with oxaliplatin is commonly used in colorectal cancer treatment.

Related Publications

Zeng Y *et al.* (2013) Stress response protein RBM3 attenuates the stem-like properties of prostate cancer cells by interfering with CD44 variant splicing. *Cancer Res.* Jul 1;73(13):4123-33.

Ehlén A *et al.* Expression of the RNA-binding protein RBM3 is associated with a favourable prognosis and cisplatin sensitivity in epithelial ovarian cancer. *J Transl Med.* 2010 Aug 20;8:78.

Hjelm B *et al.* High nuclear RBM3 expression is associated with an improved prognosis in colorectal cancer. *Proteomics Clin Appl.* 2011 Dec;5(11-12):624-35.

Boman K *et al.* Decreased expression of RNA-binding motif protein 3 correlates with tumour progression and poor prognosis in urothelial bladder cancer. *BMC Urol.* 2013, 13:17.

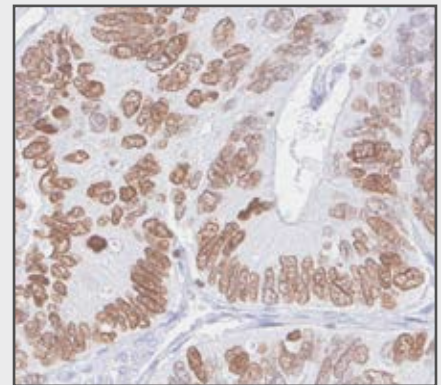
SATB2 – A diagnostic biomarker for tumors of colorectal origin

Cell- and cancer-type specific proteins are rare. The special AT-rich sequence-binding protein SATB2 has been identified as having a very selective expression pattern. In cells of epithelial lineages, SATB2 is expressed in glandular cells lining the lower gastrointestinal tract and expression is retained in a large majority of primary and metastatic colorectal cancers. Thus, SATB2 is a promising diagnostic biomarker for tumors of colorectal origin.

In a previously published study by Magnusson *et al* it was shown, by analyzing more than 1,800 tumor samples, that SATB2 expression is largely preserved in cells of colorectal cancer origin. More than 85% of all colorectal cancers showed distinct SATB2 immunostaining and when used in com-

bination with Cytokeratin 20 analysis, SATB2 identified more than 95% of all tumors with colorectal origin.

These promising data suggested that the combination of SATB2 and CK20 should be tested in an unbiased clinical study to further validate the initial findings. In a recent publication by Dragomir *et al*, the expression of SATB2 was analyzed in over 800 consecutive clinical cases for which CK20 immunostaining was considered necessary to obtain a final diagnosis. In this study, SATB2 showed 93% sensitivity and 77% specificity to determine a cancer of colorectal origin and in combination with CK7 and CK20, the specificity increased to 100%. SATB2 thus provides a new and advantageous supplement to current standards for clinical differential diagnosis.



Immunohistochemical staining of human colorectal tumor with Anti-SATB2 antibody (AMAb90635) shows strong nuclear staining in tumor cells.



Related Publications

Magnusson K *et al*. SATB2 in combination with cytokeratin 20 identifies over 95% of all colorectal carcinomas. *Am J Surg Pathol*. 2011 Jul;35(7):937-48.

Dragomir A *et al*. The role of SATB2 as a diagnostic marker for tumors of colorectal origin: results from a pathology-based clinical prospective study. *Am J Clin Pathol*. 2013 In press.

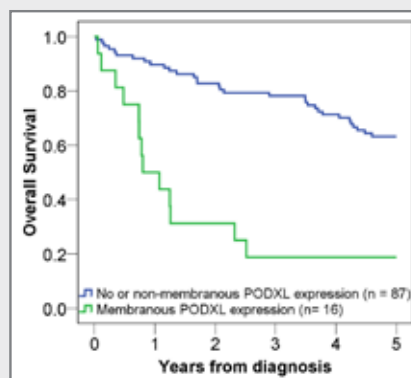
PODXL - An independent factor for poor prognosis and treatment stratification

Podocalyxin-like 1 (PODXL) is a cell-adhesion glycoprotein and stem cell marker that has been associated with aggressive tumor phenotype and adverse outcome in several cancer types.

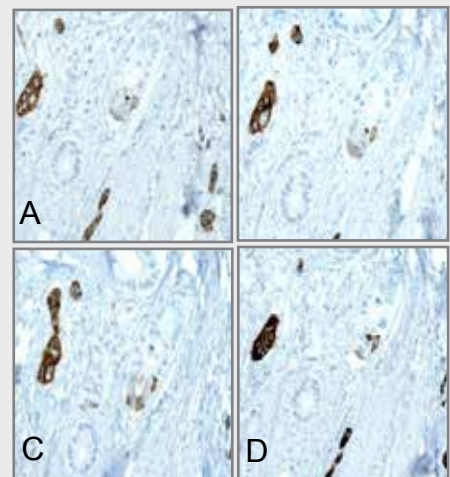
In a number of recently published papers, Larsson *et al* have demonstrated that membraneous expression of PODXL is associated with unfavourable clinicopathological characteristics and independently predicts a poor prognosis in colorectal cancer (CRC). This has been demonstrated in three independent patient cohorts in total comprising more than 1,000 patients. The results clearly demonstrate the potential utility of PODXL as a biomarker for more precise prognostication and treatment stratification in CRC.

Boman *et al* have investigated the

prognostic impact of membraneous PODXL expression in almost 500 cases of urothelial cancer. They concluded that PODXL is indeed an independent risk factor for progressive disease and death in patients with urothelial cancer and that this warrant further studies to fully evaluate the use of PODXL as a biomarker for improved treatment stratification of bladder cancer patients.



Kaplan-Meier estimates of 5-year Overall Survival (OS) according to PODXL expression in a urothelial cancer patient cohort of 110 individuals.



Immunohistochemical staining of PODXL protein in colorectal tumor tissue using A) HPA002110, B) AMAb90643, C) AMAb90644 and D) AMAb90667 antibodies.

Related Publications

Larsson A *et al*. Overexpression of podocalyxin-like protein is an independent factor of poor prognosis in colorectal cancer. *Br J Cancer* 2011 Aug 23;105(5):666-72.

Larsson A *et al*. Validation of podocalyxin-like protein as a biomarker of poor prognosis in colorectal cancer. *BMC Cancer*. 2012 Jul 8;12:282.

Boman K *et al*. Membraneous expression of podocalyxin-like protein is an independent factor of poor prognosis in urothelial bladder cancer. *Br J Cancer*. 2013 Jun 11;108(11), 2321-2328.

Co-Development Program

Research remains at the heart of Atlas Antibodies. We welcome customers to contact us for possible collaborations on both existing and future product offerings.

Atlas Antibodies invite you to participate in our Monoclonal Antibody Development Program. If you are looking for mouse monoclonal antibodies currently not available in our catalog, and if you are interested in developing the antibody together with us, please send in your project proposal to us.

Upon agreement to proceed with a collaboration, Atlas Antibodies will develop and produce the monoclonal antibody using the same procedures as for Precisa Monoclonals. As part of this procedure we epitope map all our clones to obtain only unique

clones with defined epitopes for final characterization. The selection of the optimal clones for specific applications will be done in collaboration with you. Antibodies will either be sent to you for additional characterization in your laboratory or Atlas Antibodies will make the characterization at our facilities with your expert input and/or material. Atlas Antibodies cover all other development costs. If the project results in a commercialized product it will be added to Atlas Antibodies Precisa Monoclonal product portfolio and available to you. All antibodies will be used for staining of a multitude of human tissues by the Human Protein Atlas (HPA) project and these results will be available on the HPA web portal.

Benefits of the program

Atlas Antibodies take the full development cost while you get a discounted antibody with proven functionality in your experimental set-up.

For more information and/or requests for participating in the program, you are welcome to contact us at contact@atlasantibodies.com.

We are looking forward to hearing from you.



Collaboration project for SOX11

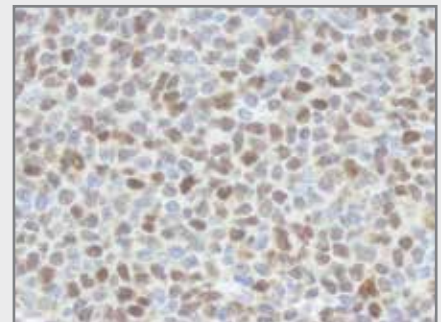
Precisa Monoclonals against SOX11 (AMAb90501 and AMAb90502) were developed in collaboration with Dr Antonio Martinez (Laboratory of Pathology, Hospital Clínic, University of Barcelona, Spain).

Dr. Martinez is involved in the study of aggressive lymphomas, mechanisms of transformation, progression and prognostic factors. He has collaborated in the description of transcription factors involved in B-cell development and lymphomagenesis with special emphasis in those related in late B-cell differentiation pathways such as IRF4, IRF8, XBP1 and SOX11. His lab has long expertise in the characterization of antibodies for clinical use in hematopathology.

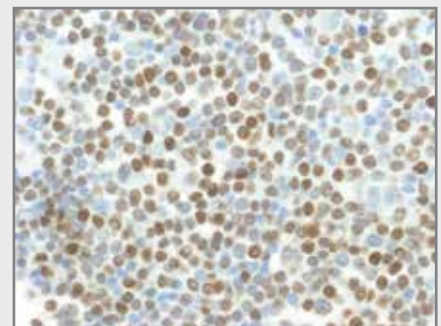
SOX11

Soldini D *et al.* Assessment of SOX11 Expression in Routine Lymphoma Tissue Sections: Characterization of New Monoclonal Antibodies for Diagnosis of Mantle Cell Lymphoma. *Am J Surg Pathol.* 2013 Oct 18.

This gene encodes a member of the group C SOX (SRY-related HMG-box) transcription factor family involved in the regulation of embryonic development and in the determination of the cell fate. The encoded protein may act as a transcriptional regulator after forming a protein complex with other proteins. The protein may function in the developing nervous system and play a role in tumorigenesis and adult neurogenesis. Diseases associated with SOX11 include mantle cell lymphoma (MCL), lymphoblastic lymphoma, Burkitt lymphoma and malignant glioma. The diagnosis of mantle cell lymphoma can be difficult, especially in Cyclin D1 negative cases and the transcription factor SOX11 may serve as an important diagnostic marker. For this purpose, there is a need of a reliable Anti-SOX11 antibody in the clinical setting.



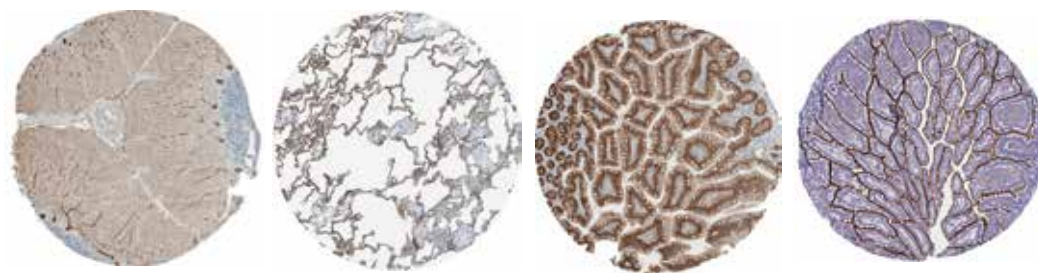
Tonsil involved by a Classical Mantle cell lymphoma, cyclin D1 negative in a 50 yo male. SOX11 staining (AMAb90501, clone CL0142; Atlas Antibodies).



Lymph node involvement by Classical Mantle cell lymphoma positive for Cyclin D1 in a 64 yo male. SOX11 is expressed in virtually all tumor cells. (AMAb90502, clone CL0143; Atlas Antibodies).



Anti-KRT20 (HPA024684) in human colorectal cancer tissue.



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Or send an e-mail to support@atlasantibodies.com to discuss any matters regarding use of antibodies. **You'll find we're Totally Human.**