

Single IHC Staining*

| Antibody code | Antibody - primary designation | Antibody - secondary designation | Source | Cat# | Ab type | Validated species reactivity | Primary classification | Control tissue - human | Control tissue - mouse | Comments |
|---------------|--------------------------------|----------------------------------|---------------------------|----------|-------------------|------------------------------|---------------------------------------|------------------------|------------------------|--|
| AKT1 | Akt1/PKBa | | Cell Signaling Technology | 4685 | rabbit monoclonal | human | Signal transduction | Colon cancer | N/A | A Ser/Thr protein kinase whose activity plays a key role in various cellular functions, including apoptosis, glycogen synthesis, and cell growth. |
| AKT-pS473 | Akt1/PKBa (pS473) | | Abcam | Ab81283 | rabbit monoclonal | human | Signal transduction | Colon cancer | N/A | Phosphorylation on Thr308, Ser473 and Tyr474 is required for full activity. |
| Beta catenin | Beta Catenin | | Abcam | Ab 32572 | rabbit monoclonal | human | Signal transduction, Stem cell marker | colon cancer | N/A | Change of localization from cell surface to nuclear indicates Wnt signalling. Stem cells often have wnt signalling. |
| CD3 | CD3 | | Dako | A0452 | Rabbit polyclonal | Human,mouse | T cell marker | Tonsil | spleen | CD31 is non-covalently associated with T cell receptor. The CD3 components of the TCR/CD3 complex mediate signal transduction upon antigen recognition by TCR. CD3 is expressed by T cells in thymus, bone marrow, blood and lymphoid tissues. |

Core Immunohistochemical Stains

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|--------|-------|--|---------|----------|----------------------|-------|----------------------|--------|--------|---|
| CD4(H) | CD4 | | Ventana | 790-4423 | rabbit monoclonal | Human | T cell marker | Tonsil | N/A | CD4 a membrane glycoprotein of T lymphocytes that interacts with major histocompatibility complex class II antigens and is also a receptor for the human immunodeficiency virus. This antigen is expressed not only in T lymphocytes, but also in B cells, macrophages, and granulocytes. It is also expressed in specific regions of the brain. The protein functions to initiate or augment the early phase of T-cell activation. |
| CD4(M) | CD4 | | Abcam | Ab183685 | rabbit monoclonal | mouse | T cell marker | N/A | spleen | CD4 a membrane glycoprotein of T lymphocytes that interacts with major histocompatibility complex class II antigens and is also a receptor for the human immunodeficiency virus. This antigen is expressed not only in T lymphocytes, but also in B cells, macrophages, and granulocytes. It is also expressed in specific regions of the brain. The protein functions to initiate or augment the early phase of T-cell activation. |
| CD8(H) | CD8 | | Dako | M7103 | mouse monoclonal | Human | T cell marker | Tonsil | N/A | The CD8 antigen is a cell surface glycoprotein found on most cytotoxic T lymphocytes that mediates efficient cell-cell interactions within the immune system. The CD8 antigen acts as a coreceptor with the T-cell receptor on the T lymphocyte to recognize antigens displayed by an antigen presenting cell in the context of class I MHC molecules. |
| CD8(M) | CD8 | | Abcam | Ab209775 | rabbit monoclonal | mouse | T cell marker | N/A | spleen | The CD8 antigen is a cell surface glycoprotein found on most cytotoxic T lymphocytes that mediates efficient cell-cell interactions within the immune system. The CD8 antigen acts as a coreceptor with the T-cell receptor on the T lymphocyte to recognize antigens displayed by an antigen presenting cell in the context of class I MHC molecules. |
| Cd11b | CD11b | | Abcam | Ab133357 | rabbit monoclonal | Mouse | Macrophage marker | N/A | spleen | CD11b is a leukocyte-specific receptor and is regarded as a marker for monocyte/macrophages, granulocytes, and natural killer cells. |

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| CD19(H) | CD19 | | Abcam | Ab134114 | rabbit monoclonal | human | B cell marker | Tonsil | N/A | CD19 is a biomarker for normal and neoplastic B cells, as well as follicular dendritic cells. CD19 is critically involved in establishing intrinsic B cell signaling thresholds through modulating both B cell receptor-dependent and independent signaling. |
| CD19(M) | CD19 | | Abcam | Ab245235 | rabbit monoclonal | mouse | B cell marker | N/A | spleen | CD19 is a biomarker for normal and neoplastic B cells, as well as follicular dendritic cells. CD19 is critically involved in establishing intrinsic B cell signaling thresholds through modulating both B cell receptor-dependent and independent signaling. |
| CD20 | CD20 | | Leica microsystems | NCL-L-CD20 | Mouse monoclonal | Human | B cell marker | tonsil | N/A | CD20 is a membrane-embedded surface molecule which plays a role in the development and differentiation of B-cells into plasma cells. |
| CD31(H) | CD31 | PECAM1 | Abcam | Ab 134168 | rabbit monoclonal | human | Leukocyte & endothelial differentiation | colon-mucosa | N/A | CD31 is found on the surface of endothelial cells, platelets and leukocytes. It plays a major role in a number of cellular interactions, particularly in adhesion between endothelial cells and leukocytes during inflammation and angiogenesis. Angiogenesis is critical to tumor growth, neoplastic progression and metastasis |
| CD31(H,M) | CD31 | PECAM1 | Abcam | ab28364 | rabbit polyclonal | human, mouse | Leukocyte & endothelial differentiation | colon | colon | CD31 is found on the surface of endothelial cells, platelets and leukocytes. It plays a major role in a number of cellular interactions, particularly in adhesion between endothelial cells and leukocytes during inflammation and angiogenesis. Angiogenesis is critical to tumor growth, neoplastic progression and metastasis |

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| CD45(M) | CD45 | | Abcam | Ab281586 | Rabbit polyclonal | mouse | Leukocyte differentiation | N/A | spleen | CD45 is a transmembrane glycoprotein which is expressed at high level on leukocytes from hematopoietic cells. CD45 isoforms play roles in T-cell and B-cell antigen receptor signaltransduction. |
| CD45(H) | CD45 | | Dako | M0701 | mouse monoclonal | human | Leukocyte differentiation | tonsil | N/A | CD45 is a transmembrane glycoprotein which is expressed at high level on leukocytes from hematopoietic cells. CD45 isoforms play roles in T-cell and B-cell antigen receptor signaltransduction. |
| CD56 | CD56 | | Cell Marque | 156R-94 | Rabbit monoclonal | human | NK cell marker | lymph node | N/A | CD56 is the archetypal phenotypic marker of natural killer cells but can actually be expressed by many more immune cells, including alpha beta T cells, gamma delta T cells, dendritic cells, and monocytes |
| CD68(H) | CD68 | | Novus Biologicals | NB100-683 | mouse monoclonal | human | Macrophages | Tonsil | N/A | Glycosylated transmembrane protein which is mainly located in lysosomes. It reacts with myeloid precursors and peripheral blood granulocytes. |
| CD68(M) | CD68 | | Abcam | Ab125212 | Rabbit polyclonal | mouse | Macrophages | N/A | spleen | Glycosylated transmembrane protein which is mainly located in lysosomes. It reacts with myeloid precursors and peripheral blood granulocytes. |

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|--------------|-----------------------------------|--|-------------------|--------------------|-------------------|---------------|---|----------------------|--------|--|
| Cl Caspase-3 | Cleaved Caspase-3 | | Cell Signaling | 9661 | rabbit polyclonal | human, mouse] | Apoptosis | lymph node | thymus | Caspase 3 is one of the key excutioners of apoptosis, as it is either partially or toatlly responsible for the proteolytic cleavage of many key proteins such as PARP. |
| Ecad | E-cadherin | | Abcam | Ab40772 | rabbit monoclonal | human | Cell-cell interaction, epithelial differentiation | normal breast tissue | N/a | E-cadherin is a glycoprotein with an extracellular domain that interacts with other E-cadherin molecules on adjacent cells, thereby establishing adhesion between epithelial cells. |
| EGFR | Epidermal Growth Factor Receptor | | Novus Biologicals | NBP1-84815 | rabbit polyclonal | human | Signal transduction, epithelial differentiation | Placenta | N/A | EGFR is type I receptor tyrosine kinase, activated by the EGF family of ligands. EGFR is overexpressed or mutated in many common forms of carcinoma. |
| pEGFR | Phospho EGFR (phospho-Tyr845) | | Nanotools | 0116-100/EGFR-12A3 | mouse monoclonal | human | Signal transduction, epithelial differentiation | Brest cancer | N/A | EGFR/erbB receptors are activated upon binding of EGF and EGF-related growth factors such as TGF alpha, beta-cellulin, Hb-EGF, HRG, or NRG. Binding of these ligands leads to receptor homo- and heterodimerization followed by autophosphorylation and activation of downstream signal transduction pathways (MAPK, PI3K/PKB, and STAT). In addition, EGFR becomes fully activated after phosphorylation of Y845 by src family kinases. |
| EpCAM | Epithelial cell adhesion molecule | GA733-2, EGP, KSA, KS 1/4, Trop-1, CD326 | Abcam | Ab32392 | rabbit monoclonal | human, mouse | Epithelial differentiation | colon mucosa | colon | A monomeric membrane glycoprotein expressed on virtually all epithelial cells |

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|-----------|---|--|---------------------------|------------|-------------------|---------------------|--------------------------------|---------------|-------------|---|
| Fox p3(H) | Fox p3 | | Leica Bon | PA0263 | Mouse monoclonal | human | Regulatory T cell | Lymph node | N/A | The FOXP3 protein, also known as scurfin, is essential for normal immune homeostasis. Specifically, FOXP3 represses transcription through a DNA binding forkhead domain, thereby regulating T cell activation. |
| Fox p3(M) | Fox p3 | | Thermo Fisher | 14-5773-82 | Rat | mouse | Regulatory T cell | N/A | Lymph node | The FOXP3 protein, also known as scurfin, is essential for normal immune homeostasis. Specifically, FOXP3 represses transcription through a DNA binding forkhead domain, thereby regulating T cell activation. |
| pERK | Phospho-p44/42 MAPK (ERK1/2)(Thr202/Tyr204) | | Cell Signaling | 9101 | rabbit polyclonal | human,mouse | Signal transduction | colon cancer | mouse tumor | MAPKs are a widely conserved family of serine/threonine protein kinases involved in many cellular programs such as cell proliferation, differentiation, motility, and death. The p44/42 MAPK (Erk1/2) signaling pathway can be activated in response to a diverse range of extracellular stimuli including mitogens, growth factors, and cytokines (1-3) and is an important target in the diagnosis and treatment of cancer |
| GFP | Green Fluorescent Protein | | Novus Biologicals | NB600-303 | Rabbit polyclonal | GFP tagging protein | Fluorescence marker | GFP control | GFP control | The green fluorescent protein (GFP) is a protein that exhibits bright green fluorescence when exposed to light in the blue to ultraviolet range |
| Her2 | c-erbB-2,neu | | Cell Signaling Technology | CS 4290 | Rabbit monoclonal | human | Signal transduction / oncogene | Breast cancer | N/A | This protein is a member of the epidermal growth factor (EGF) receptor family of receptor tyrosine kinases. This protein is involved in kinase-mediated activation of downstream signalling pathways, such as those involving mitogen-activated protein kinase and phosphatidylinositol-3 kinase. Amplification and/or overexpression of this gene has been reported in numerous cancers, including breast and ovarian tumors |

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|--------------|-------------------------------------|--|----------------|---------|-------------------|---------------------------|----------------------------|-------------------------------------|-------------|---|
| Pan CK | Pan Cytokeratin | AE1/AE3 | Cell Marque | 313M-1 | mouse monoclonal | Human | Epithelial differentiation | Colon | N/A | a broad-spectrum keratin antibody cocktail.1-3 It is composed of mouse monoclonal antibody AE1 that recognizes acidic type I keratins 10, 14, 15, 16, 19, and AE3 that reacts with basic type II keratins 1, 2, 3, 4, 5, 6, 7, and 8. B This antibody is useful for identifying cells of epithelial origin and detects most carcinomas of different primary sites |
| Ki67 | Ki67 | | Abcam | Ab16667 | rabbit monoclonal | human, mouse | Proliferation / Cell cycle | tonsil | thymus | Present in active phases of the cell cycle (late G1, S, G2, and mitosis), but absent in resting cells. |
| NFKB-p65 | Nuclear factor kappa B, p65 subunit | | Abcam | Ab32536 | rabbit monoclonal | human | Signal transduction | colon cancer | N/A | Inducible transcription factor involved in apoptosis resistance. Bound to inhibitor (IKB) and retained in cytoplasm. During activation, the p65 subunit is released from IKB and translocates to the nucleus. |
| pMEK(Ser221) | Phospho-MEK 1/2(Ser221) | MAPK | Cell Signaling | 2338 | rabbit monoclonal | human, Mouse, Rabbit, Dog | Signal transduction | Pancreatic cancer | mouse tumor | MEK1 and MEK2 are dual-specificity protein kinases that function in a mitogen activated protein kinase cascade controlling cell growth and differentiation |
| p16 | p16 | cyclin-dependent kinase inhibitor 2A, CDKN2A | BD Pharmingen | 550834 | mouse monoclonal | human | Tumor suppressor gene | Severe squamous dysplasia of cervix | N/A | p16 protein is a specific inhibitor of cdk4, which regulates Rb and Rb related proteins, p107 and p130. p16 is inactivated in many tumor types by gene mutation or silencing, and its protein level is influenced by papillomavirus oncoproteins. |

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| p21 | p21 | WAF1/Cip1 | Santa Cruz | sc-6246 | mouse monoclonal | human, mouse | Proliferation/ cell cycle | Colon cancer | N/A | Promotes cell cycle transitions. |
| p53 | p53 | | Dako Cytomation | M7001 | mouse monoclonal | Human | Tumor suppressor gene | Colon cancer | N/A | p53 plays a major role in the cellular response to DNA damage and other genomic aberrations. Activation of p53 can lead to either cell cycle arrest and DNA repair, or apoptosis. |
| p63 | p63 | | Biocare Medical | CM 163A | mouse monoclonal | human, mouse | Epithelial differentiation | prostate | N/A | Expressed in many types of basal epithelium. Differentiates between basal and surface/luminal epithelium. |
| PARP1 | PARP-1 (Cleaved p25) | | Abcam | Ab 32064 | rabbit monoclonal | human, mouse | Apoptosis | tonsil | thymus | Cleaved in vivo by caspase 3, hence is a marker of activated apoptotic pathway. |
| Plectin | Plectin 1 | | Abcam | ab32528 | rabbit monoclonal | human, mouse | Epithelial differentiation / cancer biomarker | cancer-pancreas | N/A | An intermediate filament binding protein, Could also bind muscle proteins such as actin to membrane complexes in muscle. |

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| SARS -CoV2-NP | SARS-CoV-2 (COVID-19) Nucleocapsid | | ProSci | 9099 | Rabbit polyclonal | Covid-19 NP | Covid-19 NP marker | Covid 19 infected tissue | Covid-19 mouse model | Predicted reactivity based on immunogen sequence: SARS-CoV Nucleocapsid proteins: (100%) |
| SMA | smooth muscle actin | alpha actin | Abcam | Ab32575 | rabbit monoclonal | human, mouse | Dsmooth muscle differentiation | colon-smooth muscle | colon | Contractile protein that makes up the cytoskeleton. SMA is restricted to smooth muscle cells (including vascular smooth muscle) and myoepithelial cells. |
| Vim(H) | Vimentin | | Abcam | Ab16700 | rabbit monoclonal | human | Mesenchymal differentiation | colon-smooth muscle | N/A | Most common member of intermediate filament family and a main component of cytoskeleton structure. Most strongly expressed in mesenchymal cells and other cell types derived from mesoderm. |
| Vim(H,M) | Vimentin | | Abcam | ab45939 | rabbit polyclonal | human, mouse | Mesenchymal differentiation | colon | colon | Most common member of intermediate filament family and a main component of cytoskeleton structure. Most strongly expressed in mesenchymal cells and other cell types derived from mesoderm. |

Multiplex IHC staining*

Double stain

- CD68/Pan CK(Human)
- CK18/CD45(Human)
- cleaved Caspase3/Ki67 (Human &Mouse)
- CD8 /Ki67(Human & Mouse)
- Choices of 2 markers for immune cells: CD4/CD8/CD68/CD19/FoxP3 (Human &Mouse)

Triple stain

CK18/CD45/SMA (Human)

Ki67/CD45/CK18(Human)

Choices of 3 markers for immune cells: CD4/CD8/CD68/CD19/FoxP3 (Human & Mouse)

Quadruplex(4-plex) stain

Choices of 4 markers for immune cells: CD4/CD8/CD68/CD19/FoxP3 (Human & Mouse)

Quintuplex(5-plex) stain

Immune cell markers: CD4/CD8/CD68/CD19/FoxP3 (Human & Mouse)

*** Customized IHC is available, please contact Pat Pramoontjago, Ph: 434-982-0487, email : pp6f@virginia.edu**