Marshfield Labs™

Immunohistochemistry (IHC) Services

- Validated using 10% neutral buffered formalin.
- Unstained required for each antibody marker is 2 (1 minimum) except:
  - Kappa, Lambda, or Epstein-Barr Virus by In Situ Hybridization requires 6 (3 minimum) slides
  - Lambda/Kappa double stain by In Situ Hybridization requires 8 (4 minimum) slides
  - Estrogen, Progesterone, c-erbB2 Oncoprotein (HER2), and PD-L1 all require 2 (1 minimum) unstained each and an additional H & E.
- Age of a cut paraffin section can affect immunoreactivity; Stability thresholds vary widely among published literature and are antigen-dependent. Best practice is for paraffin sections to be cut fresh.
- Note type of tissue/specimen
- Unless specified otherwise, positive and negative controls react satisfactorily.
- Detection system is a polymer.

Available Chromogen – All markers have been validated with 3,3'-Diaminobenzidine Tetrahydrochloride (DAB) which results in a brown/black precipitate. DAB is the routine chromogen. In addition, some markers have also been validated using the Fast Red (RED), which results in a red precipitate. If available with both chromogens and one is not selected, the default will be the DAB chromogen.

*This test was developed and its performance characteristics determined by Marshfield Labs. It has not been cleared or approved by the US Food and Drug Administration. This test is used for clinical purposes. It should not be regarded as investigational or for research.

Antibody	Common Applications	Staining Characteristics
Actin (muscle specific)	Smooth, skeletal & cardiac muscle	Cytoplasmic
Actin (smooth muscle)	Smooth muscle and myoepithelial cells	Cytoplasmic and membrane
ALK Protein	ALK1 positive lymphomas	Cytoplasmic and/or nuclear
Alpha-1-Antitrypsin (A-1-AT)	Demonstrates A-1-AT in liver	Cytoplasmic
Bcl-2 Oncoprotein	Follicular lymphoma and soft tissue tumors	Cytoplasmic
Bcl-6	Follicular lymphoma	Nuclear
Ber-EP4, Epithelial Antigen	Adenocarcinoma vs. mesothelioma and epithelial tumors	Membrane and cytoplasmic. The membrane staining is preferentially basolateral.
Beta-Catenin	Desmoid type-fibromatosis, solid and pseudopapillary tumors of pancreas.	Nulcear
CD1a	Langerhan cells, thymic T-cells, thymoma	Membrane and weakly cytoplasmic
CD3	T-cells, lymphoma/leukemia typing	Membrane
CD4	Helper/Inducer T-cells mycosis fungoides vs. cutaneous inflammatory processes, lymphoma/ leukemia typing	Membrane
CD5	T-cell, lymphoma/leukemia typing	Membrane
CD8	T-cell suppressor/cytotoxic, lymphoma/leukemia typing, mycosis fungoides vs. cutaneous inflammatory processes	Membrane
CD10	Lymphoma typing, metastatic carcinoma unknown primary	Cytoplasmic and Membrane
CD15 *	Hodgkin Lymphoma Typing, mesothelioma vs. adenocarcinoma	Reed-Sternberg cells in Hodgkin's lymphoma show cell membrane and granular paranuclear staining.
CD20, B Cell	Lymphoma/leukemia typing	Cytoplasmic side of the cell surface membrane
CD21	Follicular Dendritic cell marker, found also on some B cells	Cytoplasmic & Membrane
CD23	Lymphoma/leukemia typing	Cytoplasmic/membrane
CD30	Anaplastic large cell lymphoma, Hodgkin lymphoma	Membrane and/or a dot like cytoplasmic staining

Antibody	Common Applications	Staining Characteristics
CD31	Endothelial Cells	Predominately cell membrane, with weaker cytoplasmic staining.
CD34	Soft tissue tumor classification, leukemia typing	Membrane/cytoplasmic
CD43	T-cell, lymphoma/leukemia typing	Predominantly confined to the cell surface
CD45 (LCA)	Lymphohematopoetic tumors	Membrane, but cytoplasmic may also occur
CD56	NK cells, tumors derived from neuroectodermal tumors such as neuroendocrine and neuroblastomas, etc.	Membrane
CD68, PG-M1, Macrophage	Histiocytic/monocytic marker	Cells of monocyte/macrophage lineage stain diffuse or granular cytoplasmic. Mast cells are negative.
CD79a	B-cell, lymphoma/leukemia typing	Cell Membrane and/or cytoplasm
CD99, SEE MIC2		
CD117 (c-kit)	Gastrointestinal Stromal Tumors (GIST), Mast Cells, Stains approx..75% of mesenteric fibromatosis tumors.	Membrane and/or cytoplasmic
CD138 Syndecan-1	Plasma Cells (also stains endothelial cells, fibroblasts, keratinocytes, and normal hepatocytes)	Cell Membrane, pre-B cell and plasma cell marker, but is absent from mature B cells. It is a selective marker for B cell lymphoblastic leukemia and lymphoplasmocytoid leukemia. It is lost from the apoptotic myeloma cells; hence is a useful marker for viable myeloma cells.
CD163	Histiocytic Lesions	Membrane
CDX2	Colon and other GI cancers are strong & diffusely positive. Also, mucinous ovarian cancers are positive. Neg for HCC, breast, lung, head, and neck ca.	Nucleus in normal and neoplastic intestinal epithelial cells.
Calcitonin	Medullary Thyroid Carcinoma	Cytoplasmic
Calretinin	Mesothelioma vs. adenocarcinoma, sex cord stromal, adrenal tumors, & Hirschprung's cases. Please specificy Meso or Hirsch	Cytoplasmic and nuclear
Carbonic Anhydrase IX (CAIX)	Differential diagnosis of the various types of renal cell carcinomas (clear cell renal cell carcinoma and clear cell papillary renal cell carcinoma vs. other renal cell carcinomas)	Membranous box-like staining pattern (clear cell renal cell carcinoma) cup-like staining pattern (clear cell papillary renal cell carcinoma)
Carcinoembryonic Antigen, CEA/M	Mesothelioma vs. Adenocarcinoma, metastatic carcinoma of unknown primary	Cytoplasmic. Normal colon-CEA is mainly localized at the apical border of the epithelial cells. Colon carcinoma-CEA is mainly localized at the apical border of glandular structures, whereas cytoplasmic labeling predominates in more solid parts of the tumor.
Carcinoembryonic Antigen, CEA/P	Hepatoma vs. adenocarcinoma	Cytoplasmic. In liver, predominantly bilary canaliculi are labeled. In colonic adenocarcinoma the staining reaction is localized diffusely in the cytoplasm and in the gland lumina.
Chromogranin A	Neuroendocrine differentiation	Cytoplasmic
Cyclin D1	Mantle cell lymphoma	Nuclear
Cytokeratin 5/6	Mesothelioma vs. Adenocarcinoma	Cytoplasmic

Antibody	Common Applications	Staining Characteristics
Cytokeratin 7	Metastatic carcinoma of unknown primary	Cytoplasmic. Exceptions to reactivity may occur e.g. CK-7 positive hepatocytes have been observed in patients with acute and chronic cholestasis.

Antibody	Common Applications	Staining Characteristics
Cytokeratin 20	Metastatic carcinoma of unknown primary	Cytoplasmic. May occasionally be expressed in breast and lung adenocarcinomas, and in squamous cell carcinomas. Less than 5% CK20 positive cells may be present in a number of tissues not generally considered CK 20
CK116 – MNF116	Epithelial marker	Cytoplasmic
Cytokeratin 34BE12	Prostate basal cells, squamous cells vs. adenocarcinoma	Cytoplasmic
Cytokeratin, AE1/AE3	Epithelial tumors, hepatoma vs. adenocarcinoma	Cytoplasmic
Cytokeratin, CAM5.2	Epithelial tumors	Cytoplasmic
Cytokeratin Cocktail (KerCK)	Epithelial tumors	Cytoplasmic
Cytomegalovirus/IHC *	Cytomegalovirus	Nuclear staining pattern in early HCMV infection, later stage, cytoplasmic staining might be observed. Does not x-react with adenovirus, herpes simplex virus, & varicella zoster virus.
D2-40	Lymphatic endothelium, mesothelioma vs. adenocarcinoma	Cytoplasmic and sometimes membrane
Desmin	Smooth and skeletal muscle differentiation	Cytoplasmic, may show a fibrillary aspect.
Desmin/Gata-3	See Individual Markers	Desmin Cytoplasmic (DAB) GATA-3 Nuclear (RED)
DOG-1	Gastrointestinal stromal tumors, chondroblastoma, acinic cell carcinoma, hidadenoma papilliferum	Cytoplasmic and membranous
Epstein-Barr Virus (EBV) RNA CISH *	Latent EBV infection	Nuclear. Surgical/Hematopathology consultation and review of the entire case is highly recommended.
E-Cadherin	Lobular vs. ductal breast carcinoma	Cellular membrane, some cytoplasmic
Epithelial Membrane Antigen (EMA)	Metastatic carcinoma of unknown primary, lymphoma	In neoplasms, cytoplasmic and apical luminal membrane staining are the most common patterns of immunoreactivity with peripheral membrane staining or other patterns also occurring. Plasma cells stain positive. In normal breast and other secretory epithelia, labeling is predominantly localized to apical luminal membranes.
Estrogen Receptor (ERA)	Breast carcinoma prognostic marker, metastatic carcinoma of unknown primary	Nuclear, cytoplasmic is considered non-specific Occasional lymphoid tumors and non-lymphoid neoplasms such as melanomas are labeled. Follow CAP & ASCO guidelines.
Factor VIII Related Ag (Von Willebrand Factor)	Megakaryocytic and endothelial marker	Cytoplasm as diffuse or sometimes slightly granular staining
GATA-3	Urothelial carcinoma, breast ductal epithelium, and transitional cells	Must be nuclear, strong or moderate intensity, and non-focal in urothelial carcinoma.
Glial Fibrillary Acidic Protein GFAP	Glial tumors	Cytoplasmic Acetone fixed frozen or Bouins fixed tissues label certain neuronal structures, including axons, indicting a X-rxn w/ neurofilament

Antibody	Common Applications	Staining Characteristics
Glypican-3	Hepatocellular carcinoma vs. benign hepatocellular lesions Yolk Sac tumor, Choriocarcinoma	Granular cytoplasmic, and membranous patterns
Helicobacter Pylori *	Helicobacter pylori infection	Individual H. pylori bacterium when present on the surface of the epithelium or in the cytoplasm of the epithelial cells.
HepPar1 (Hep Ab/Hepatocyte)	Hepatoma, gastric carcinoma	Displays a distinct, granular cytoplasmic staining pattern, which is occasionally ring-like and is present diffusely throughout the hepatocyte cytoplasm without canalicular accentuation.
HER2/NEU c-erbB-2 Oncoprotein	HER-2/neu overexpression for invasive breast and gastric cancers.	Follow CAP and ASCO guidelines for interpretation.
Herpes Simplex Virus I (HSVI) *	HSV infection	Nuclear and cytoplasmic
HMB-45, Melanosome	Melanocyte marker, angiomyolipoma	Cytoplasmic. Order DAB or RED
hMLH-1	Screening for Lynch Syndrome	Nuclear – stains normal cells and non-mutant cells
hMSH-2 *	Screening for Lynch Syndrome	Nuclear – stains normal cells and non-mutant cells
hMSH-6 *	Screening for Lynch Syndrome	Nuclear – stains normal cells and non-mutant cells
IgG	Plasma Cell Marker, used in ratio with IgG4 for IgG4 related diseases	Cytoplasmic
IgG4	Plasma Cell Marker, used in ratio with IgG for IgG4 related diseases	Cytoplasmic
IDH1	Oligodendroglioma Tumors	Cytoplasmic
Inhibin, Alpha	Adrenal cortical, sex-cord stromal tumors	Cytoplasmic
Kappa Light Chains	Plasmacytomas, neoplastic lymphoid tumors vs. reactive proliferations	Cell membrane and/or cytoplasm
Kappa/Lambda Double or Single Stain RNA CISH *	Demonstrate clonality in leukemias, plasmacytomas, and certain non-Hodgkin lymphomas	Combination of Kappa (RED) and Lambda (DAB). Cytoplasmic.
Ki-67	Cell proliferation marker	Nuclear, except in mitotic cells where the chromosomes and cytoplasm are labeled Occasional labeling of tissue
KiMart	Ki-67 and Melan A. See separate antibodies.	Ki-67 nuclear DAB and Melan A cytoplasmic RED.
Lambda Light Chains	Plasmacytomas, neoplastic lymphoid tumors vs. reactive proliferations	Cell membrane and/or cytoplasm
Melan-A (A103)	Melanocyte marker, adrenal cortical, sex-cord stromal tumors	Cytoplasmic The Melan-A gene is also called MART-1. Order DAB or RED
Melanoma Cocktail (MelCK)	Melanocyte marker	Cytoplasmic. Combination of HMB-45, two clones of MART-1 and Tyrosinanse antibodies. Order DAB or RED.
MIC2, CD99	PNET/Ewings sarcoma	Cell membrane and Cytoplasmic
MOC-31	Epithelial related antigen	Membrane
MUM1	Germinal center B cells, activated T cells, plasma cells, and melanocytes	Nuclear positivity with weak to moderate cytoplasmic staining
MYC (c-Myc)	Diffuse Large B-cell lymphomas (DLBCL)	Positive is a nuclear stain in >40% of cells staining moderate to strong
Myeloperoxidase	Myeloid marker	Cytoplasmic Occasional staining of histiocytes was observed, possibly due to phagocytosed material

Antibody	Common Applications	Staining Characteristics
MyoD1	Rhabdomyosarcoma	Nuclear. Results of a study suggest that expression in rhabdomyosarcomas is inversely related to the degree of cellular differentiation of the tumor cells. Only
Myogenin	Rhabdomyosarcoma	Nuclear. Nuclear expression has been reported to be inversely related to the degree of cellular differentiation of rhabdomyosarcoma tumor cells
Myoglobin	Rhabdomyosarcoma	Cytoplasmic
Napsin A	Pulmonary Adenocarcinoma	Cytoplasmic
Neurofilament	Neural tumors & neurons	Cytoplasmic
Neuron Specific Enolase NSE	Can be found in virtually any type of neoplasm	Cytoplasmic. Neurons are labeled in both cytoplasm and processes.
NKX3.1	Glandular epithelium of prostate	Nuclear
Oct-3/4	Classic Seminoma/Dysgerminoma, Embryonal Carcinoma, Gonadoblastoma, and Intratubular	Strong nuclear reactivity, with cytoplasmic staining
p16	Cervical dysplasia	Both nuclear and cytoplasmic. Look for diffuse and intense staining. Focal and sporadic is considered negative.
p40	Pulmonary squamous cell carcinomas, more specific than p63.	Nuclear
p53 Protein	p53 tumor suppressor gene product	Usually nuclear, but cytoplasmic has been reported in some cases.
p57	For classification of molar pregnancy	Nuclear stain in cytotrophoblasts and decidual cells. Hydropic abortus and partial moles=positive stromal cells. Complete moles=negative stromal cells.
p63 Antibody	Basal cells in the prostate gland, myoepithelial cells in breast, proliferating basal cells of cervix,	Nuclear
p63/CK5/CK14	Stains basal cells of normal and benign prostate glands, and myoepithelial cells of breast.	p63 nuclear DAB, CK5/CK14 cytoplasmic DAB.
P120 Catenin	Distinguishing between ductal and lobular breast carcinomas.	Ductal-membranous Lobular- cytoplasmic (without strong membranous staining)
p504S, Alpha-Methylacyl-CoA Racemace (AMACR) *	Prostate adenocarcinoma and atypical adenomatous hyperplasia	Granular/Cytoplasmic Best if used in conjunction with high molecular weight cytokeratin. Order DAB or RED.
PAX-5	B-cell, lymphoma/leukemia typing	Nuclear
PAX-8	Ovarian serous ca, renal cell ca, and thyroid tumors	Nuclear
PDL-1	NSCLC, GEJ adenoca, urothelial carcinoma, & other tumors/immune cells	Cell membrane of tumor cells & cell membrane and/or cytoplasm of immune cells
PIN4 *	See separate markers	p63 nuclear DAB, CK5/CK14 cytoplasmic DAB, & P504S RED.
PMS2 *	Screening for Lynch Syndrome	Nuclear – stains normal cells and non-mutant cells.
PRAME	Distinguishing melanomas from benign nevi. About 80-90% of melanomas will stain positive with PRAME	Nuclear
Progesterone Receptor, PRA	Breast prognostic marker	Nuclear, cytoplasminc is considered non-specific. Follow CAP & ASCO guidelines.
Prostate Cocktail	Prostate carcinoma	Combo of PAP and PSA Antibodies

Antibody	Common Applications	Staining Characteristics
Prostate Specific Antigen PSA	Prostate carcinoma	Cytoplasmic. Staining is predominantly intracytoplasmic and secretions are also frequently stained positively.
Prostatic Acid Phosphastase (PAP)	Prostate carcinoma	Cytoplasmic
S100	Melanoma, neural marker	Cytoplasmic. Order DAB or RED.
SATB2	Identifies lower gastrointestinal tract epithelium	Nuclear
Smooth Muscle Myosin Heavy Chain (SMMHC)	Myoepithelial marker, smooth muscle differentiation	Cytoplasmic
SOX-10	Melanocytic and breast ductal epithelium	Nuclear
SOX-11	Identify Cyclin D1- negative mantle cell lymphoma	Positive: Nuclear Staining of at least moderate intensity in > or = 30% of neoplastic cells
STAT6	Solitary fibrous tumors	Nuclear
Synaptophysin	Neuroendocrine differentiation	Cytoplasmic pattern, occasionally revealing a punctuate or granular pattern
TAG-72	Adenocarcinoma vs. mesothelioma	Cell surface and cytoplasmic
Terminal Deoxynucleotidyl Transferase (TdT)	Elevated levels have been reported in tumor cells of lymphoblastic lymphomas (MLLB), lymphoid blast crisis of chronic	Nuclear
Thyroglobulin	Thyroid carcinomas	Staining is confined to the lumen of thyroid follicles and the apical surface of thyrocytes. In carcinomas it may also
Tryptase	Mast cells	Granular cytoplasmic staining pattern, corresponding to the secretory granules of mast cells
TTF-1, Thyroid Transcription Factor 1	Lung & thyroid marker, also some neuorendocrine	Nuclear
Tyrosinase	Melanocytic lesions	Cytoplasmic. Order DAB or RED.
Vimentin	Metastatic carcinoma of unknown primary, sarcomas	Cytoplasmic
Wilms' Tumor (WT1)	Wilms' Tumor, serous carcinoma, & other tumors	Nuclear