

**CD45RO (UCHL1), IHC-Paraffin Staining Report**

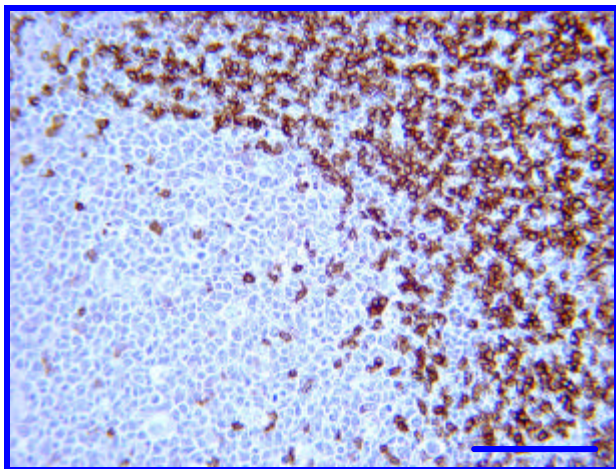
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|--------------------------------------|--|
| <b>Target Antigen:</b>               | Human CD45RO antigen   |
| <b>Catalog or ID #:</b>              | IHC2082  |
| <b>Host and Isotype:</b>             | Mouse IgG2a, Kappa.  |
| <b>Clone</b>                         | UCHL1  |
| <b>Purity and Method:</b>            | Protein "A" Affinity Chromatography  |
| <b>Concentration and Quantity:</b>   | Prediluted   |
| <b>Storage Conditions:</b>           | 2-4.0°C refrigerated   |
| <b>Species Reactivity:</b>           | Labels most thymocytes, a subpopulation of resting T cells within both the CD4 and CD8 subsets, and mature active T cells. Cells of myelomonocytic series are also UCHL1-immunoreactive.   |
| <b>IHC-Paraffin Fixation Tested:</b> | 10% neutral Buffered Formalin  |
| <b>IHC-Paraffin Titer:</b>           | N/A  |
| <b>IHC-Paraffin Pretreatment(s):</b> | Heat Induced Epitope Retrieval (HIER). Recommend No Epitope retrieval or TE Buffer, pH 9.0. Diffusive background is increased with Citrate Buffer, pH 6.0 and EDTA, pH 8.0   |
| <b>IHC-Paraffin Protocol:</b>        | <b>IHC-Select, HRP-DAB</b> (Brief Outline):<br>Blocking Reagent for 5 min.<br>Primary Antibody (or Negative Control Reagent) for 10 min.<br>Biotinylated Secondary Antibody for 10 min.<br>Streptavidin-HRP for 10 min.<br>DAB for 10 min.<br>Hematoxylin for 1 min. |
| <b>Positive Tissue(s):</b>           | Tonsil and lymph node. Reactive lymph nodes exhibit immunoreactivity primarily in the paracortex and absence of labeling in germinal center.   |
| <b>Subcellular Location(s):</b>      | Plasma Membrane staining   |
| <b>Negative Tissue(s):</b>           | Non-lymphatic tissue   |
| <b>Comments and References:</b>      | Staining is as expected since it is plasma membrane-expressed on the vast majority of cells on the Mantle Zone with a low density in the Germinal Center. No signal is detected on mature erythroid cells and vascular endothelial cells.                            |



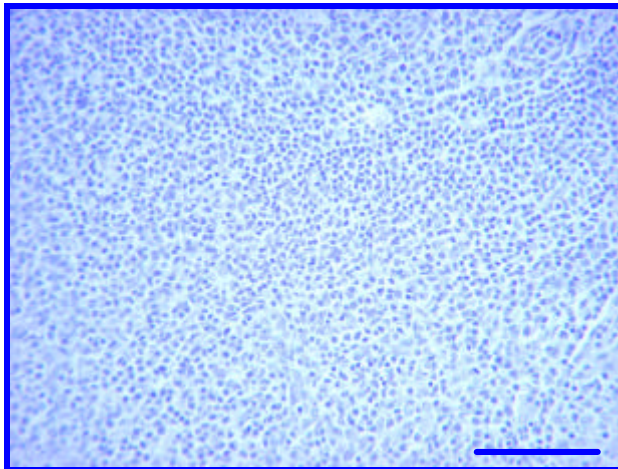
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**IHC – Paraffin Staining Examples:**

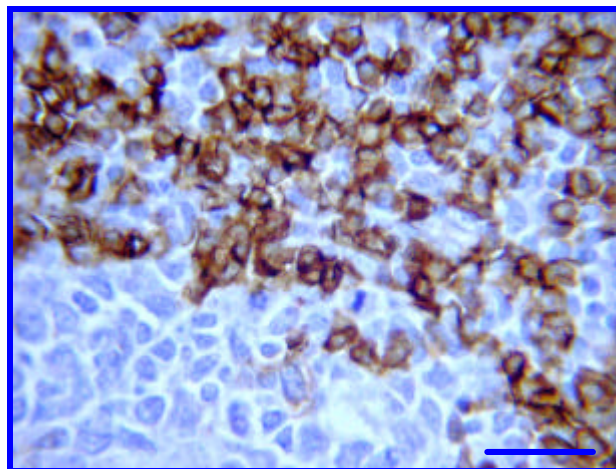
**Pictures of Optimal Staining Without Epitope Retrieval: Tonsil**



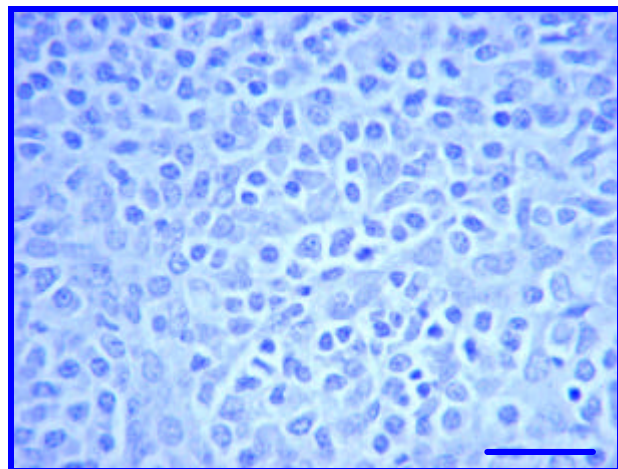
CD45 (IHC2082, Clone: UCHL1) staining of Human Tonsil. Tissue pretreated without Epitope retrieval. Prediluted antibody, IHC-Select Detection with HRP-DAB. Lymphocytes in the Mantle Zone stained strongly (brown). Bar = 100µm.



Negative Control staining of Human Tonsil. Tissue pretreated with Citrate Buffer, pH 6.0. Negative Control Reagent, IHC-Select Detection with HRP-DAB. No background staining is detected. Bar = 100µm.



CD45 (IHC2082, Clone: UCHL1) staining of Human Tonsil. Tissue pretreated without Epitope retrieval. Prediluted antibody, IHC-Select Detection with HRP-DAB. Lymphocytes in the Mantle Zone stained strongly (brown). Bar = 25µm.



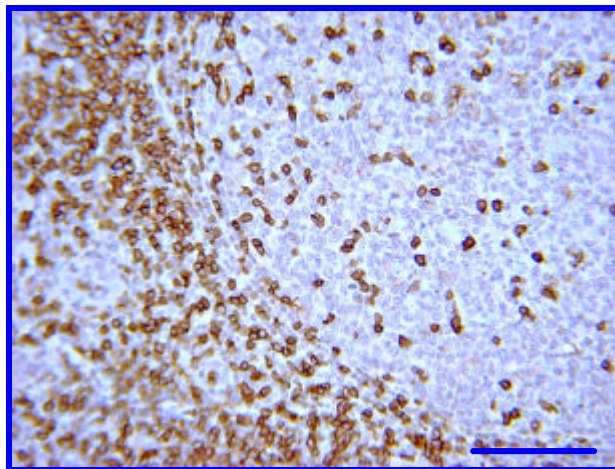
Negative Control staining of Human Tonsil. Tissue pretreated with Citrate Buffer, pH 6.0. Negative Control Reagent, IHC-Select Detection with HRP-DAB. No background staining is detected. Bar = 25µm.



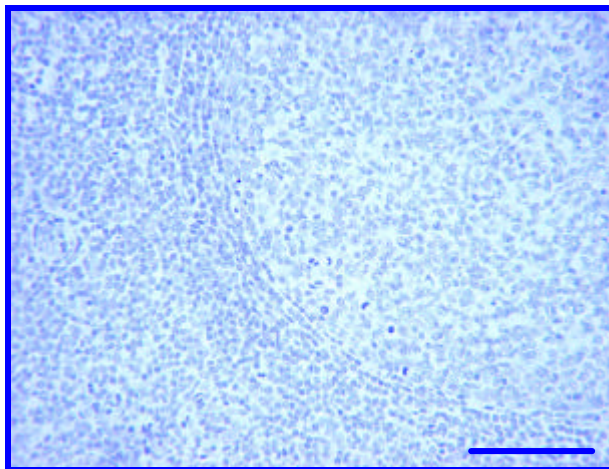
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**IHC – Paraffin Staining Examples:**

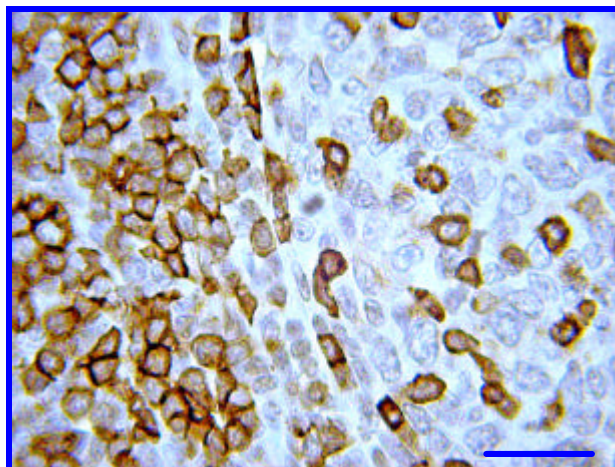
**Pictures of Optimal Staining With TE Buffer Epitope Retrieval: Tonsil**



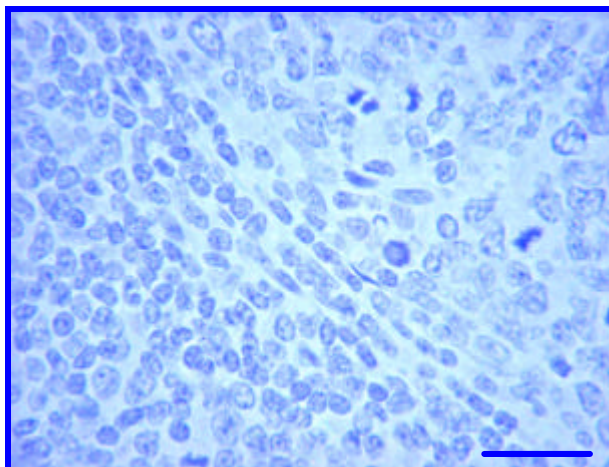
CD45 (IHC2082, Clone: UCHL1) staining of Human Tonsil. Tissue pretreated with Te Buffer, pH 9.0. Prediluted antibody, IHC-Select Detection with HRP-DAB. Lymphocytes in the Mantle Zone stained strongly (brown). Bar = 100µm.



Negative Control staining of Human Tonsil. Tissue pretreated with TE Buffer, pH 9.0. Negative Control Reagent, IHC-Select Detection with HRP-DAB. No background staining is detected. Bar = 100µm.



CD45 (IHC2082, Clone: UCHL1) staining of Human Tonsil. Tissue pretreated with Te Buffer, pH 9.0. Prediluted antibody, IHC-Select Detection with HRP-DAB. Lymphocytes in the Mantle Zone stained strongly (brown). Bar = 25µm.



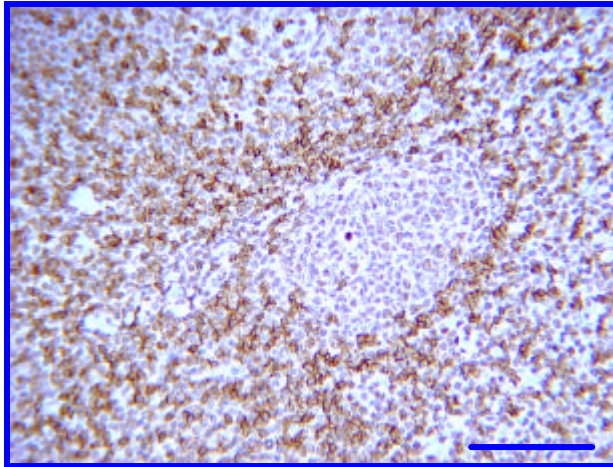
Negative Control staining of Human Tonsil. Tissue pretreated with TE Buffer, pH 9.0. Negative Control Reagent, IHC-Select Detection with HRP-DAB. No background staining is detected. Bar = 25µm.



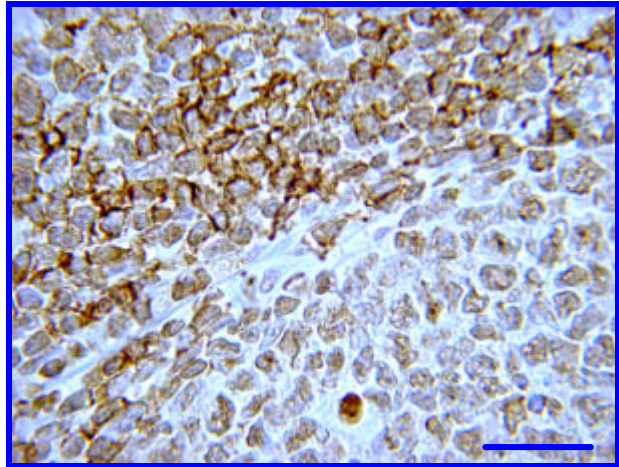
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**IHC – Paraffin Staining Examples:**

**Pictures of Over Staining With Citrate Buffer Epitope Retrieval: Tonsil**

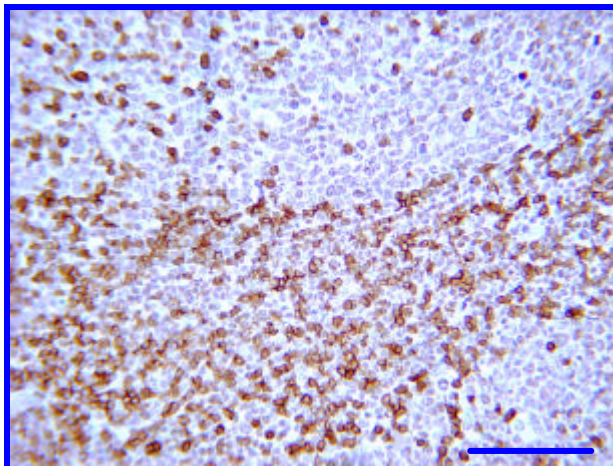


CD45 (IHC2082, Clone: UCHL1) staining of Human Tonsil. Tissue pretreated with Citrate Buffer, pH 6.0. Prediluted antibody, IHC-Select Detection with HRP-DAB. Lymphocytes in the Mantle Zone stained strongly (brown) with diffusive background. Bar = 100µm.

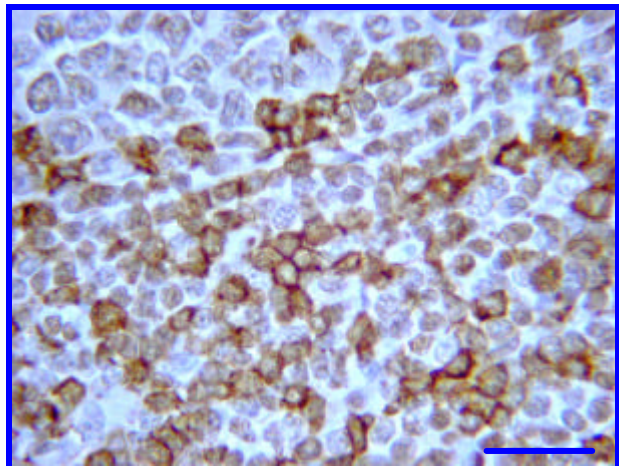


CD45 (IHC2082, Clone: UCHL1) staining of Human Tonsil. Tissue pretreated with Citrate Buffer, pH 6.0. Prediluted antibody, IHC-Select Detection with HRP-DAB. Lymphocytes in the Mantle Zone stained strongly (brown) with diffusive background. Bar = 25µm.

**Pictures of Over Staining With EDTA Epitope Retrieval: Tonsil**



CD45 (IHC2082, Clone: UCHL1) staining of Human Tonsil. Tissue pretreated with EDTA, pH 8.0. Prediluted antibody, IHC-Select Detection with HRP-DAB. Lymphocytes in the Mantle Zone stained strongly (brown) with diffusive background. Bar = 100µm.



CD45 (IHC2082, Clone: UCHL1) staining of Human Tonsil. Tissue pretreated with EDTA, pH 8.0. Prediluted antibody, IHC-Select Detection with HRP-DAB. Lymphocytes in the Mantle Zone stained strongly (brown) with diffusive background. Bar = 25µm.