ATLAS ANTIBODIES

Histones

Eukaryotic cells use histone proteins to store DNA in the form of chromatin. Chromatin consists of tightly packed nucleosomes, where each nucleosome particle is built of an octamer with pairs of the four core histones H3, H4, H2A and H2B. Each histone octamer wraps approximately 147 bp of DNA in 1.7 turns around itself. The linker histone H1 links the nucleosomes together and plays a role in modifying chromatin density and folding.

Structure

The main structure of histone proteins is the histone fold domain (HFD), a shared conserved structure composed of three α-helices connected by two loops. Besides the HFD, the histone proteins have more unique "tail" sequences which extend outside the nucleosome particle where they are subjected to various post-translational modifications (PTMs). The PTMs on histones are important for the cells regulation of gene expression.

Histone Exchange

Another mechanism for modifying chromatin structure and gene expression is histone exchange. Each of the four core histones can be replaced either by a new core histone or by one of its variants. The histone variants range from slightly different, where the sequence is altered by just a few amino acids, to having whole domains adding to their structure. By replacing a core histone with a histone variant, specialized functionalities can be achieved.

Divergence

The histone variants have different epigenetic purposes and their occurrence can be tissue specific or linked to certain developing stages.

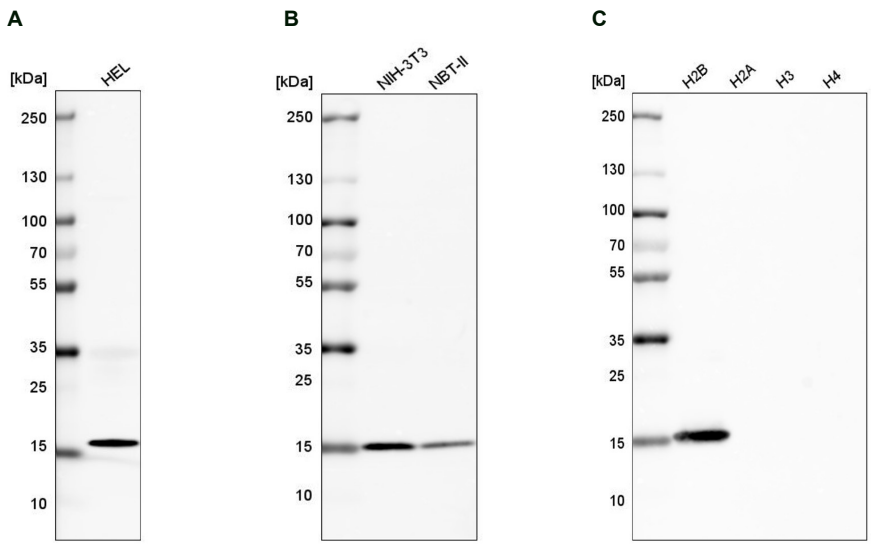
For example, the variants H3.3 and H2A.Z regulate transcription, whereas the sperm-specific variants TH2B and H2BFWT help condensate the DNA during spermatogenesis. H3 variant CENPA alters the orientation of the DNA wrapping around the nucleosome and is found in centromeric DNA.

The much larger histone H2A variants macroH2A.1 and macroH2A.2 are enriched in female inactivated X chromosomes (Xi) which suggests a role in transcriptional silencing.

PrecisA MONOCLONALS™
DESIGNED BY RESEARCHERS FOR RESEARCHERS

The PrecisA Monoclonal Histone Antibodies are developed by Atlas Antibodies, based on the knowledge from the Human Protein Atlas with careful antigen design and extended validation of antibody performance.

The antibodies specifically target regions of the histones that show low or no post-translational modifications producing a good visualization of the total amount of any histone variant in the tissue/cell line. The PrecisA Monoclonal Histone Antibodies are therefore great to combine with antibodies targeting specific modifications when performing epigenetic research.



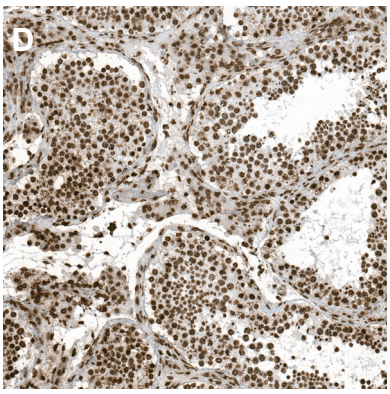


Figure 1.
Extensive validation of an antibody's specificity and selectivity.
A-C: Western Blot (WB) analysis of the Anti-H2B antibody AMAb91337. Note the presence of a specific band of expected size in both human HEL cell line lysate (**A**) and in mouse NIH-3T3 and rat NBT-II cell line lysates (**B**). The antibody selectivity is further confirmed by WB with recombinant core histone variants, where the antibody only binds to the H2B variant (**C**).
D: IHC staining of human testis with the Anti-H2B monoclonal antibody AMAb91337 shows strong nuclear immunoreactivity in cells in seminiferous ducts as expected..

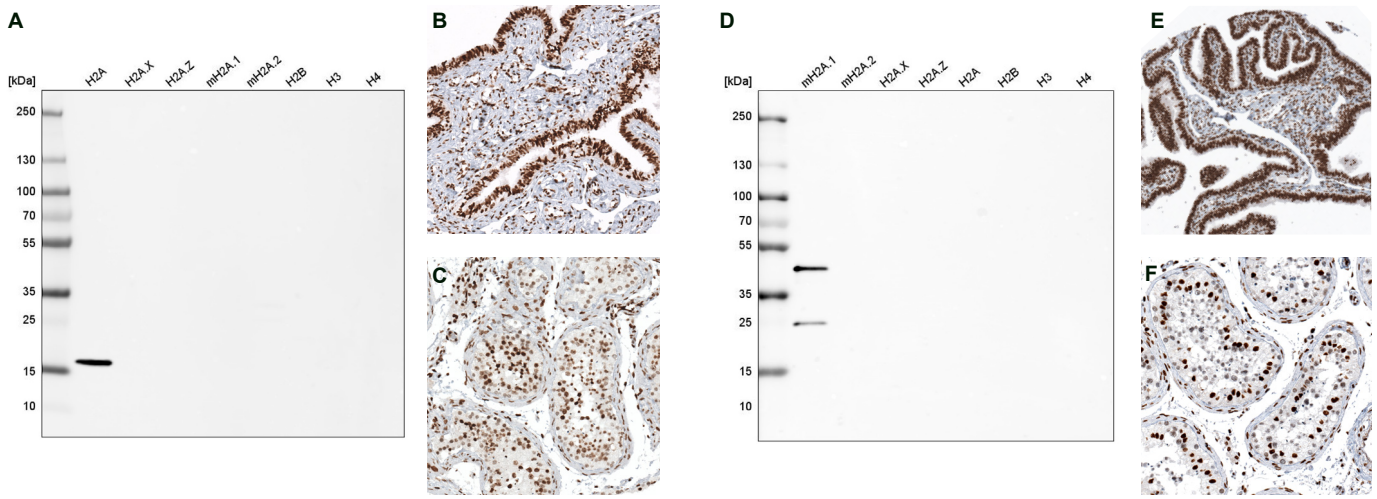


Figure 2.

Comparison of histone variant-specific antibodies.

A, D: The specificity of the antibodies for the respective subunit shown by Western Blot with recombinantly expressed histones. Note the presence of WB-signal only in the lane with the specific histone variant present, including the H2A (AMAb91335, **A**) and the macroH2A.1 (AMAb91347, **D**).

B, C, E, F: IHC staining of human tissues with the Anti-H2A antibody AMAb91335 and the Anti-macroH2A.1 antibody AMAb91347. Note that both antibodies show strong nuclear immunoreactivity in the fallopian tube (**B** and **E**). The nuclear positivity observed in testis differs: while the H2A expression is detected in nearly all cells in seminiferous tubules (AMAb91335, **C**), the macroH2A.1 is detected only in a subset of cells (AMAb91347, **F**).

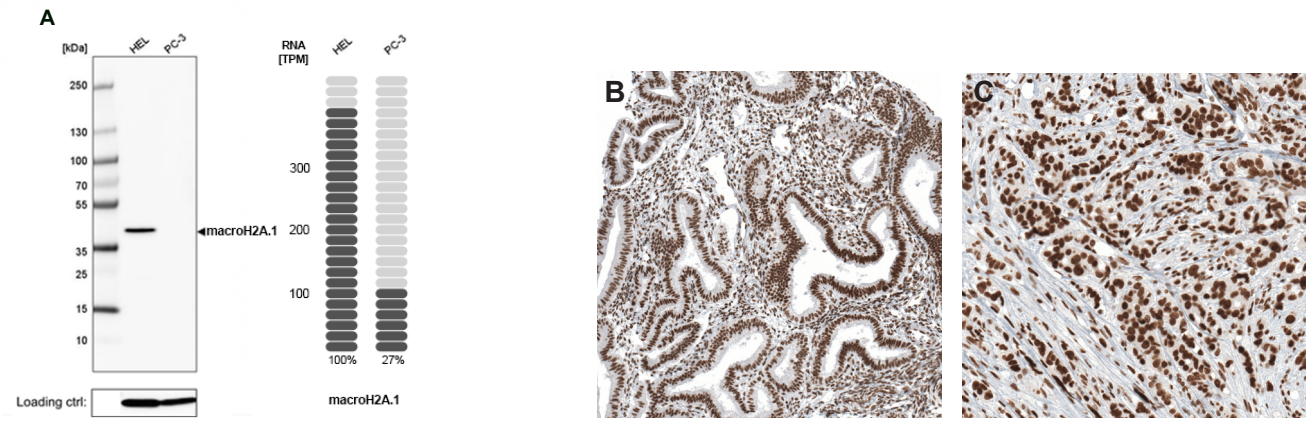


Figure 3.

Ortogonal validation of antibody specificity.

A: The specificity of the Anti-macroH2A.1 antibody (AMAb91347) was validated in WB experiments using a highly expressing cell line HEL and a lower expressing cell line PC-3. Note the band of correct size in the HEL cell line lysate, and the absence of signal in the PC-3 lysate. RNA expression (in TPM) for the respective cell lines is shown to the right.

B,C: Immunohistochemical (IHC) staining of human tissues with Anti-macroH2A.1 antibody AMAb91347 shows strong nuclear immunoreactivity in female tissues, including glandular and stromal cells in endometrium (**B**) and in tumor cells in breast cancer (**C**) as expected.

Table 1.

Description of the PrecisA Monoclonal Histone Antibodies by Atlas Antibodies.

Target gene description	Product Name	Product Number	Validated Applications	Isotype
Histone H3	Anti-HIST1H3A	AMAb91331	IHC, WB, ICC-IF	IgG2a
Histone H3	Anti-HIST1H3A	AMAb91332	IHC	IgG2b
Histone H2A	Anti-HIST1H2AG	AMAb91335	IHC, WB	IgG1
Histone H2A	Anti-HIST1H2AG	AMAb91336	IHC	IgG1
Histone H2B	Anti-HIST1H2BC	AMAb91337	IHC, WB	IgG2b
Histone H2B	Anti-HIST1H2BC	AMAb91338	IHC, WB	IgG1
Histone H2A.X	Anti-H2AFX	AMAb91346	IHC	IgG2a
Histone macroH2A.1	Anti-H2AFY	AMAb91347	IHC, WB, ICC-IF	IgG1
Histone macroH2A.1	Anti-H2AFY	AMAb91348	IHC	IgG1
Histone macroH2A.2	Anti-H2AFY2	AMAb91351	IHC, ICC-IF	IgG1

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