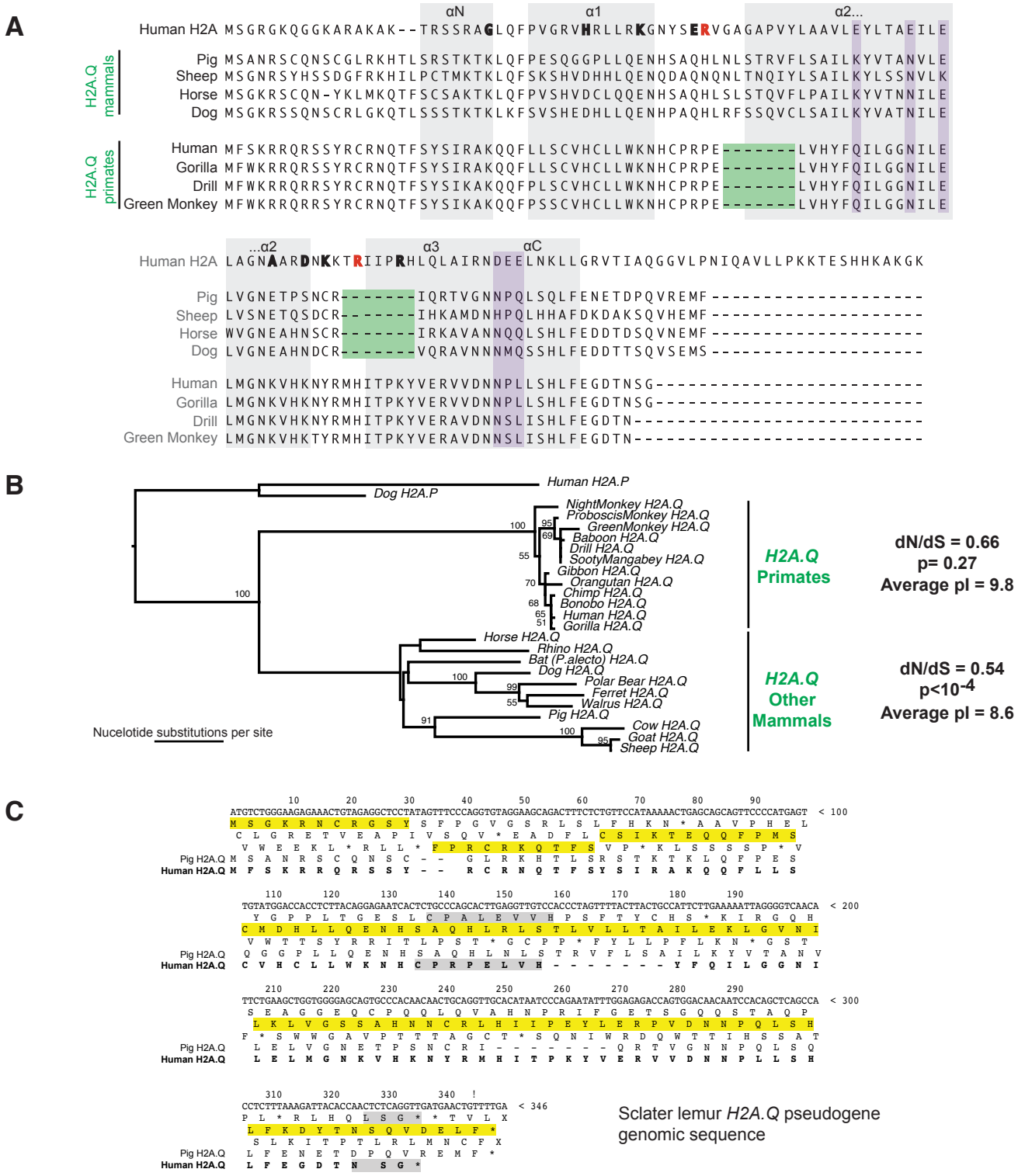


**Supplemental Fig S5: Evolutionary analysis of *H2A.Q* sequences demonstrate that primates encode *H2A.Q* pseudogenes.** (A) An alignment of representative primate and non-primate H2A.Q proteins, along with canonical H2A. Charge-altering residues that distinguish short histone variants from canonical H2A (see Figure 2A) are highlighted in bold black and the two arginine residues that make minor groove contacts are highlighted in bold red; the residues comprising the acidic patch are shown using purple shading. (B) Maximum likelihood nucleotide phylogeny of mammalian *H2A.Q* genes. Human and pig *H2A.P* are used as outgroup sequences. Non-primate *H2A.Q* genes have dN/dS values more consistent with a protein-coding open reading frame, whereas we cannot reject neutral evolution for primate *H2A.Q* sequences. (C) Alignment of Sclater's lemur (an ancestrally-branching primate) *H2A.Q* pseudogene sequence with human and pig *H2A.Q* genes shows that the ancestral H2A.Q histone fold domain had no deletions, and highlight frameshifts that occurred in simian primates, including in human *H2A.Q*. We show the DNA sequence of the *H2A.Q* pseudogene in Sclater's lemur (top), with a translation in all three frames immediately below, along with predicted protein sequences from pig and human H2A.Qs. The reading frame that aligns to intact *H2A.Q* genes, like pig *H2A.Q*, is highlighted in yellow, and putative frameshifts in the human sequence are highlighted in grey.

**Supplemental Fig S5**
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