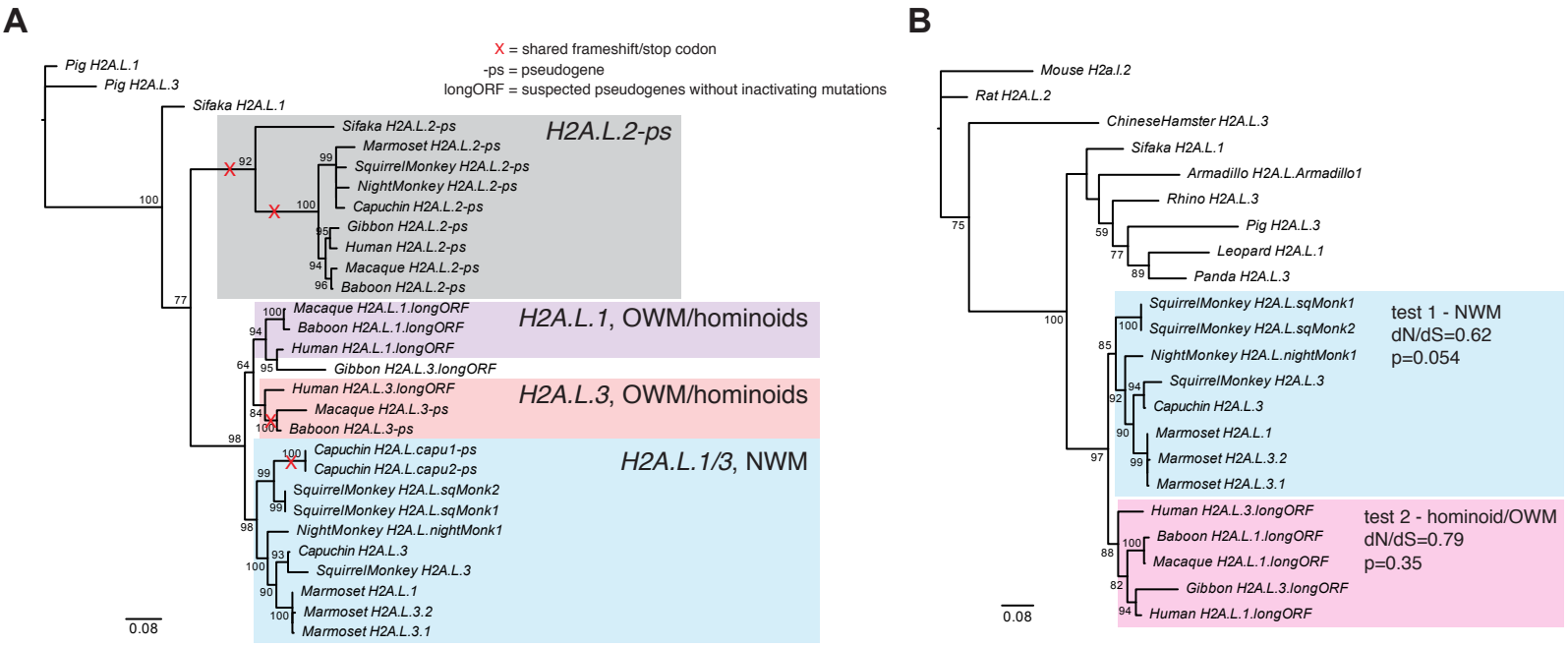

Supplemental Fig S4: Evolutionary analysis of primate *H2A.L* sequences.
Analysis of selective pressures on primate *H2A.L* sequences suggests that hominoids and Old World monkeys lack functional H2A.L sequences, but that New World monkeys likely retain functional H2A.L proteins. (A) Nucleotide phylogeny of primate *H2A.L* nucleotide sequences and pig outgroups, showing all bootstrap values >50%. The *H2A.L.2* locus acquired a stop codon in the common ancestor of all primates, and has evolved on an independent trajectory since that time. The status of the ORFs in the *H2A.L.1* and *H2A.L.3* loci is less clear-cut. Although a few sequences do exhibit inactivating mutations, others appear intact, with some encoding an unusually long C-terminal extension to the protein (labeled "longORF"). (B) Nucleotide phylogeny of selected primate and outgroup *H2A.L* genes, excluding those with obvious inactivating mutations, and showing all bootstrap values >50%. We used PAML's codeml algorithm to estimate dN/dS on selected sets of branches and to test the hypothesis that those branches are evolving non-neutrally. While New World monkey *H2A.L* genes are somewhat likely evolve under purifying selection (dN/dS=0.62, p-value for non-neutral evolution=0.054); Old World monkey/hominoid *H2A.L* sequences evolve at a rate that cannot be distinguished from neutral evolution (dN/dS=0.79, p=0.35).

Supplemental Fig S4
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