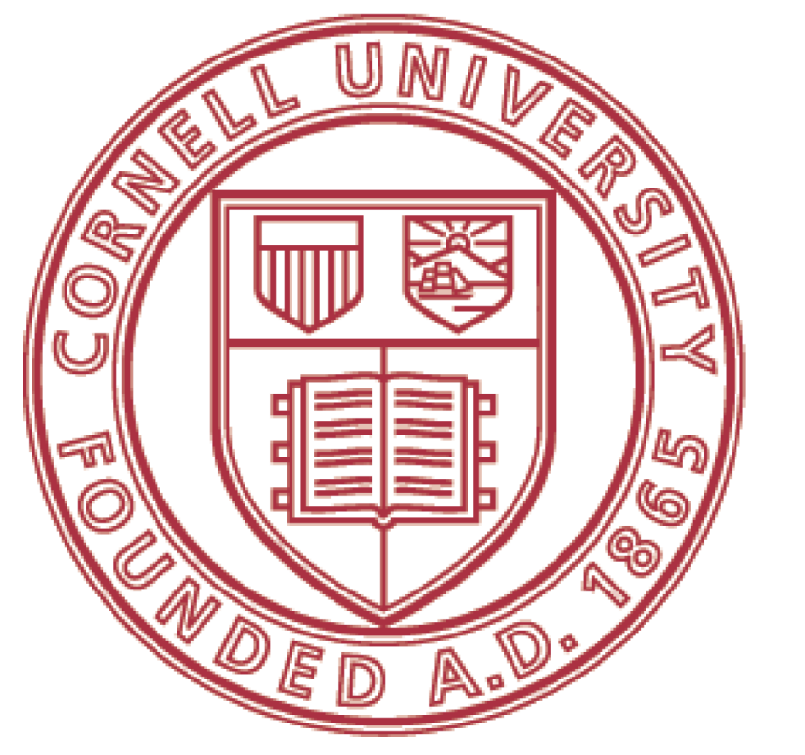


Evolutionary Changes in Gene Expression and Active Regulatory Element Usage Quantified in Primate CD4⁺ T-Cells.

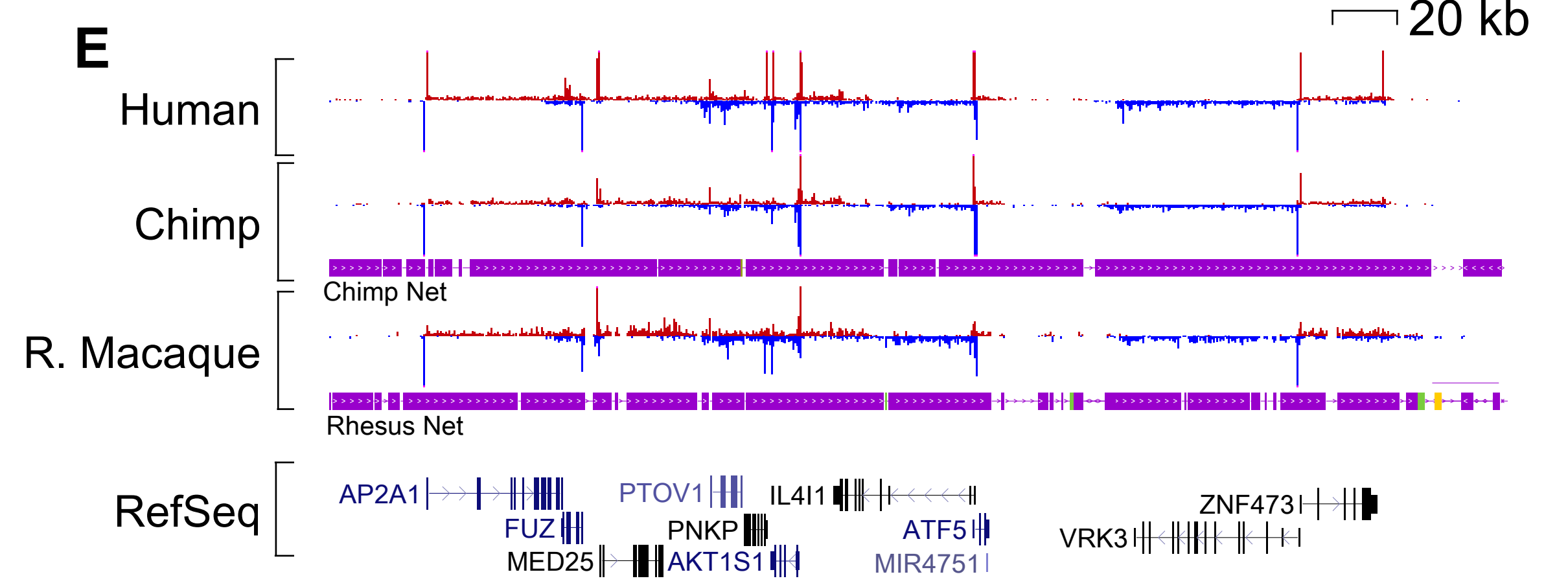
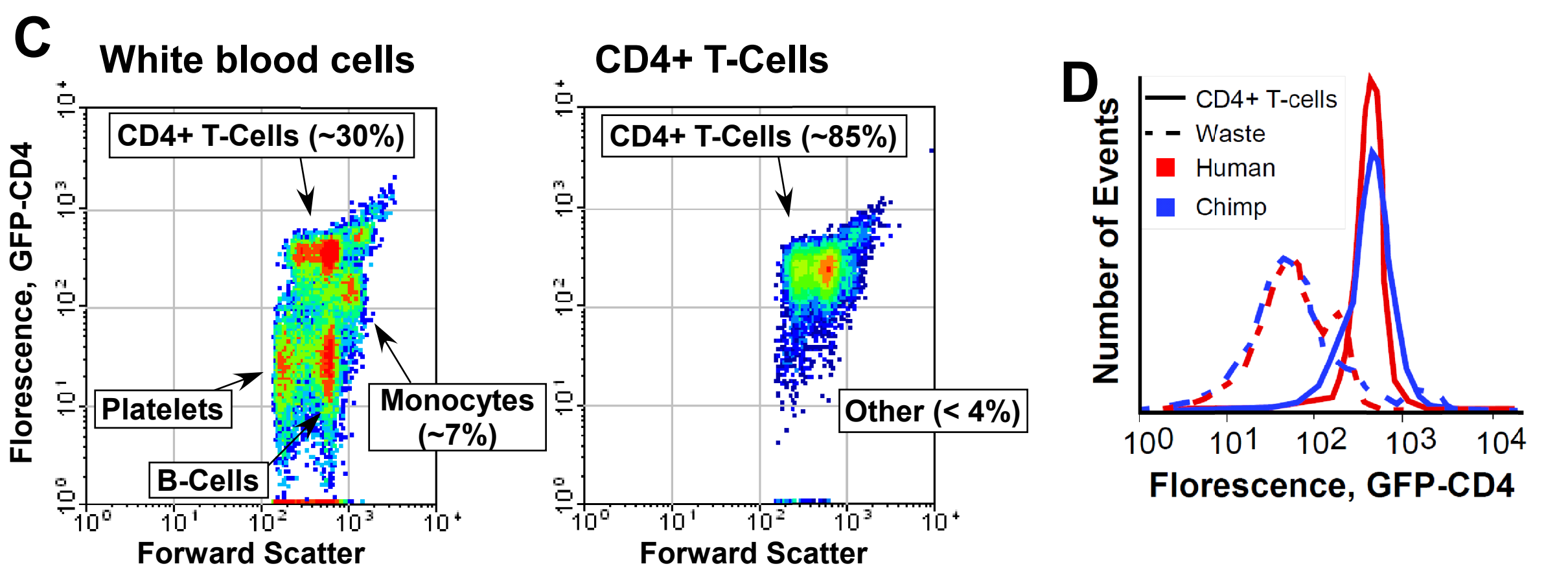
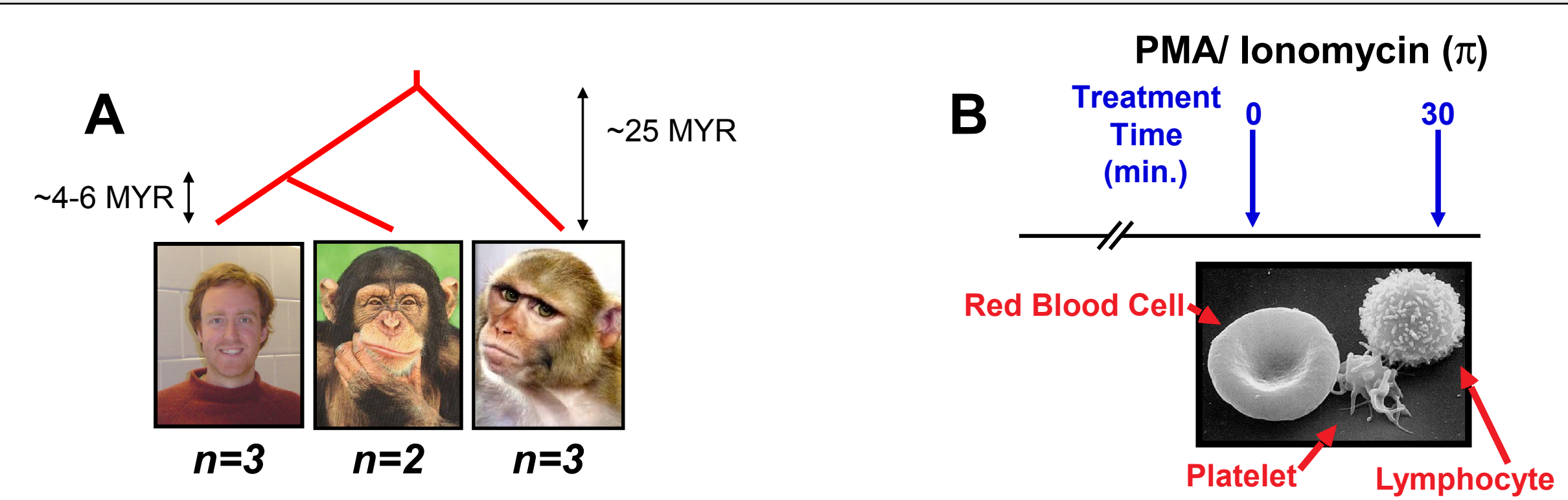
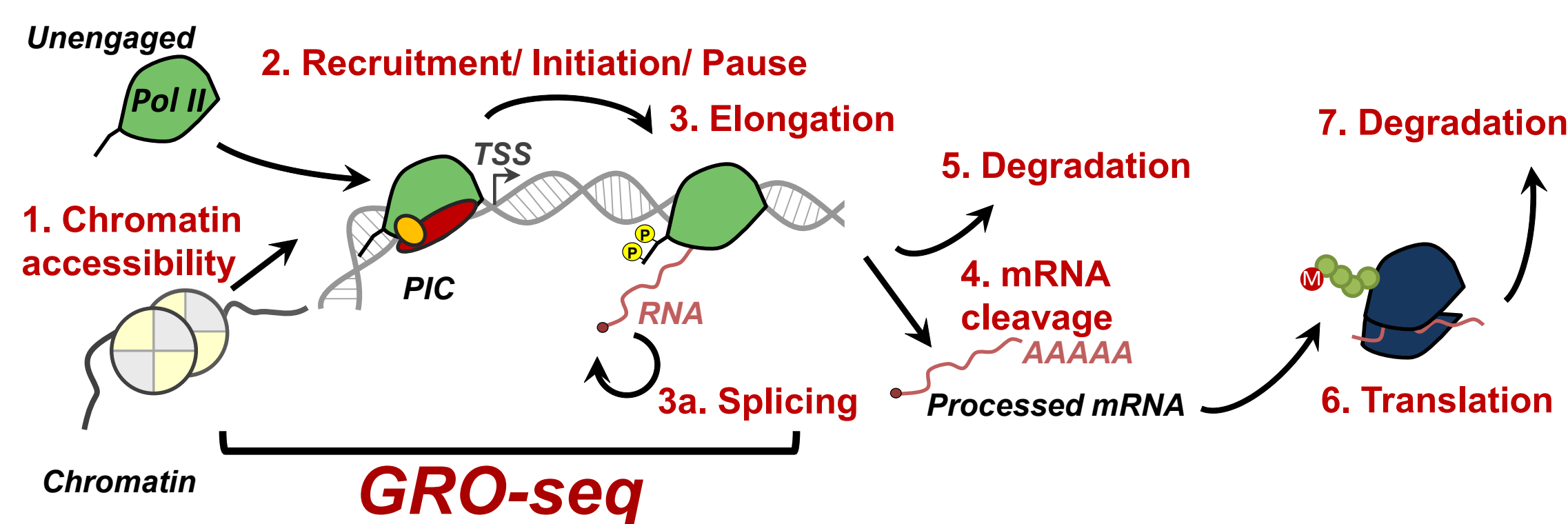


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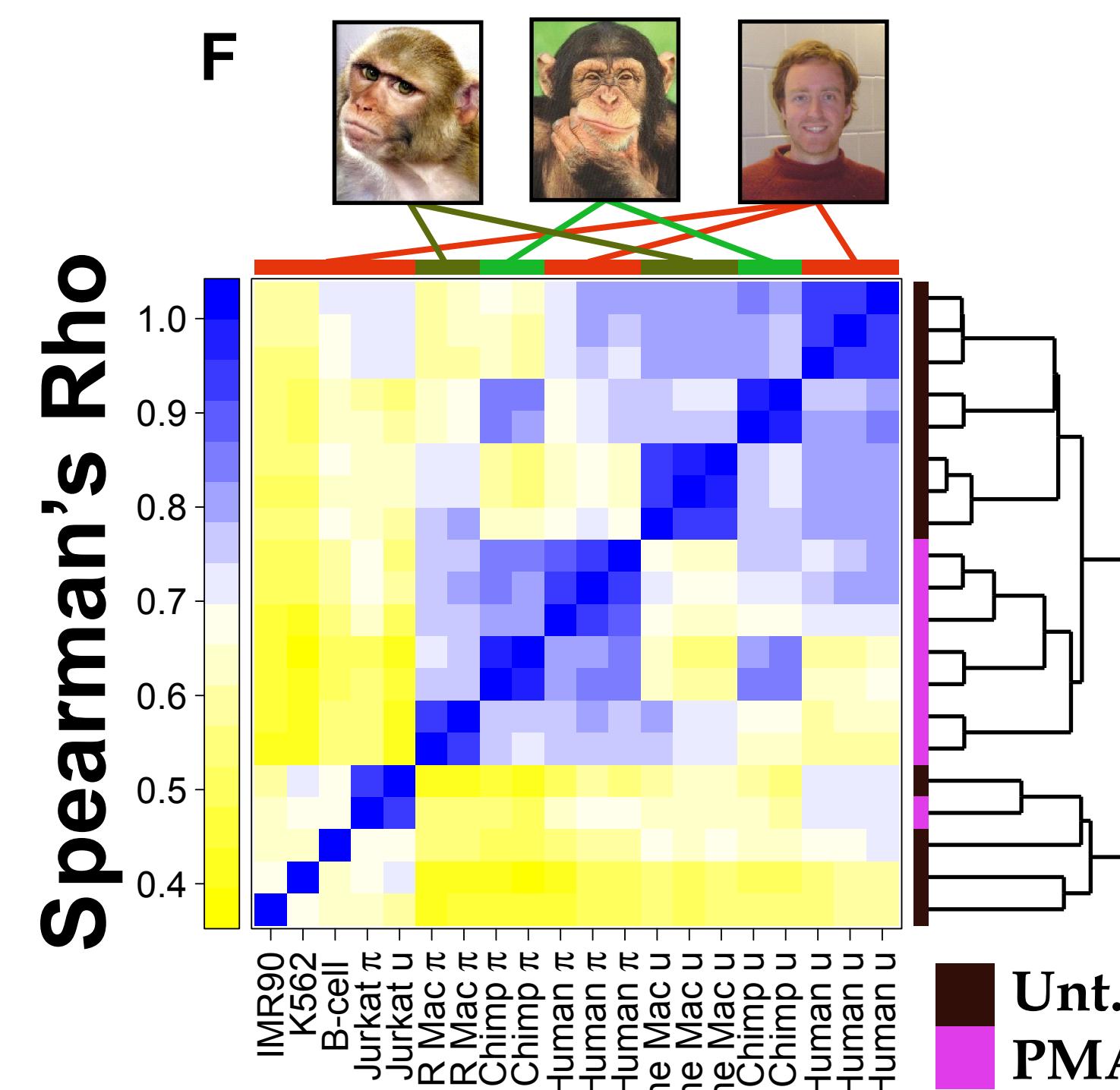
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Background:

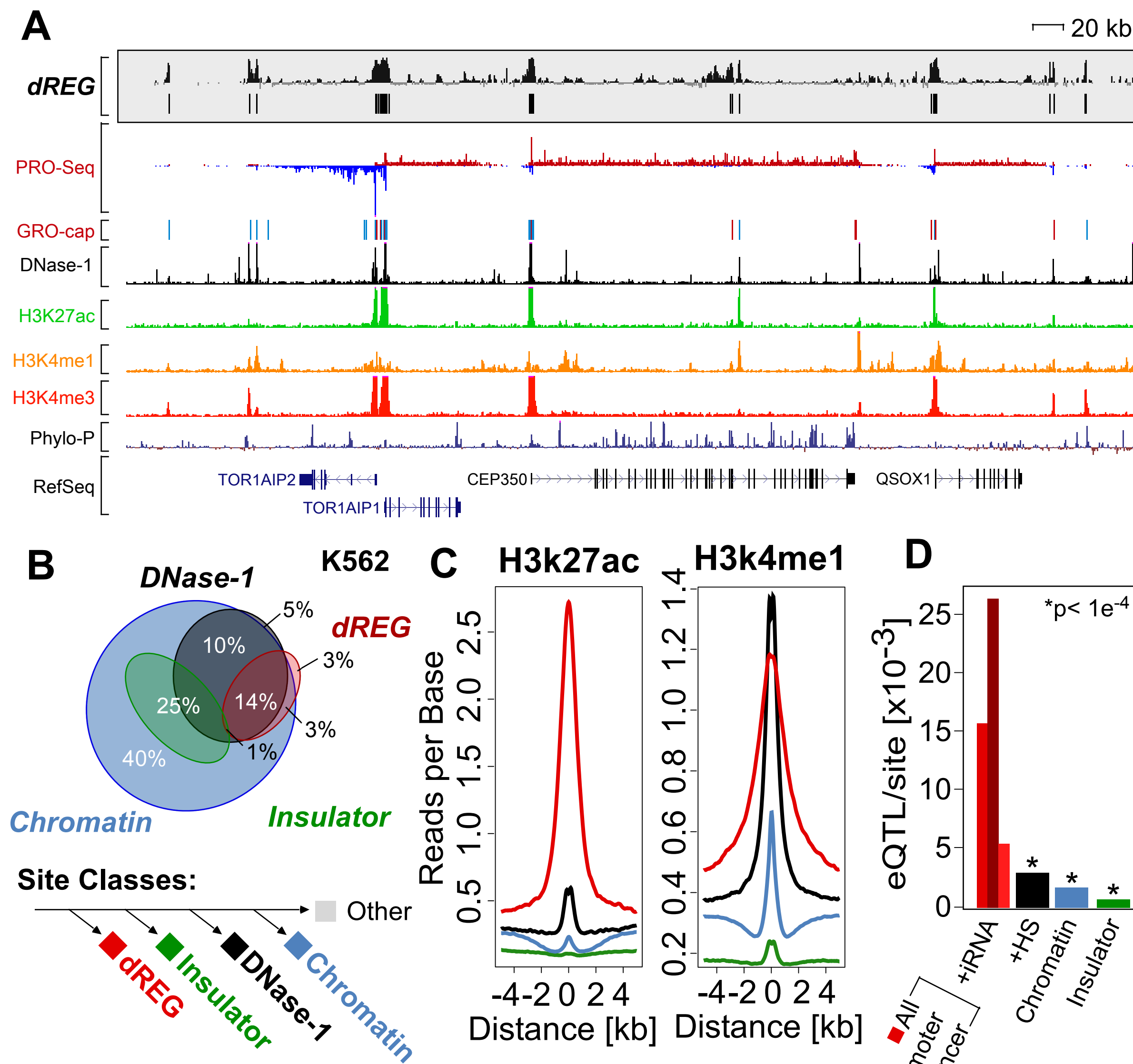
The human adaptive immune system incorporates many recent evolutionary innovations that play a central role in human biology and disease. To determine the genetic basis for differences in adaptive immunity, we used Global Run-On and Sequencing (GRO-seq) to map of the location of RNA polymerases genome-wide in primary CD4⁺ T-cells isolated from humans, chimpanzees, and rhesus macaques.



GRO-seq in CD4⁺ T-cells: (A-B) Experimental design. (C) Flow cytometry analysis of the cell population from human blood samples. (D) Human CD4 microbeads isolate CD4⁺ cell populations in human and chimpanzee. A rhesus-specific reagent was used in rhesus macaque. (E) Browser shot comparing patterns of gene expression among primate species. (F) Spearman's Rank Correlation between each sample in treated and untreated CD4⁺ T-cells.



Comparing 'active' regulatory sites.

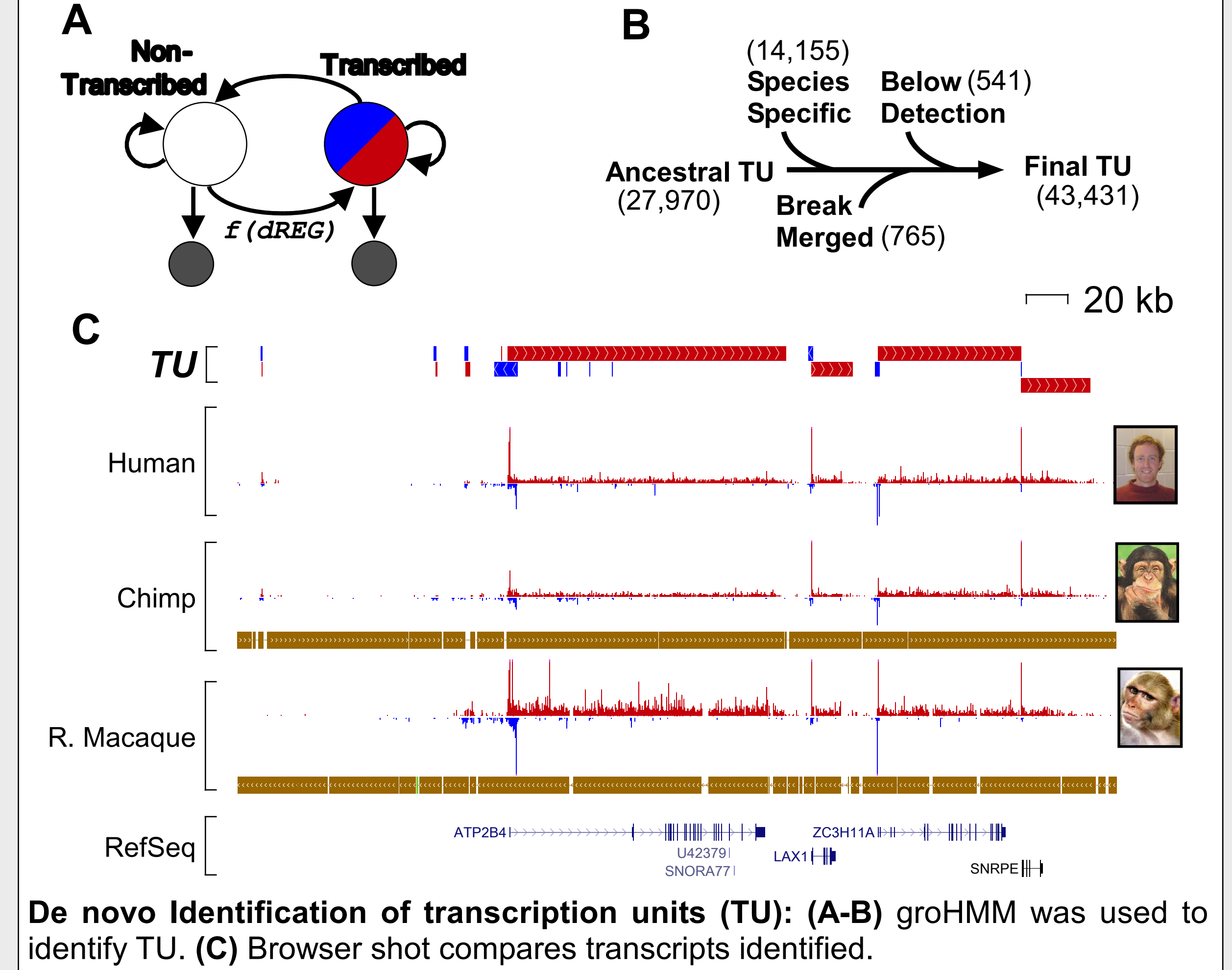


Identifying active promoters and enhancers using dREG: (A) Browser shot compares raw dREG scores and peaks to other assays. (B) dREG identifies a subset of DNase-1 accessible and chromatin modified sites. (C-D) Comparison of histone ChIP-seq and eQTL densities in regulatory element classes introduced in B.

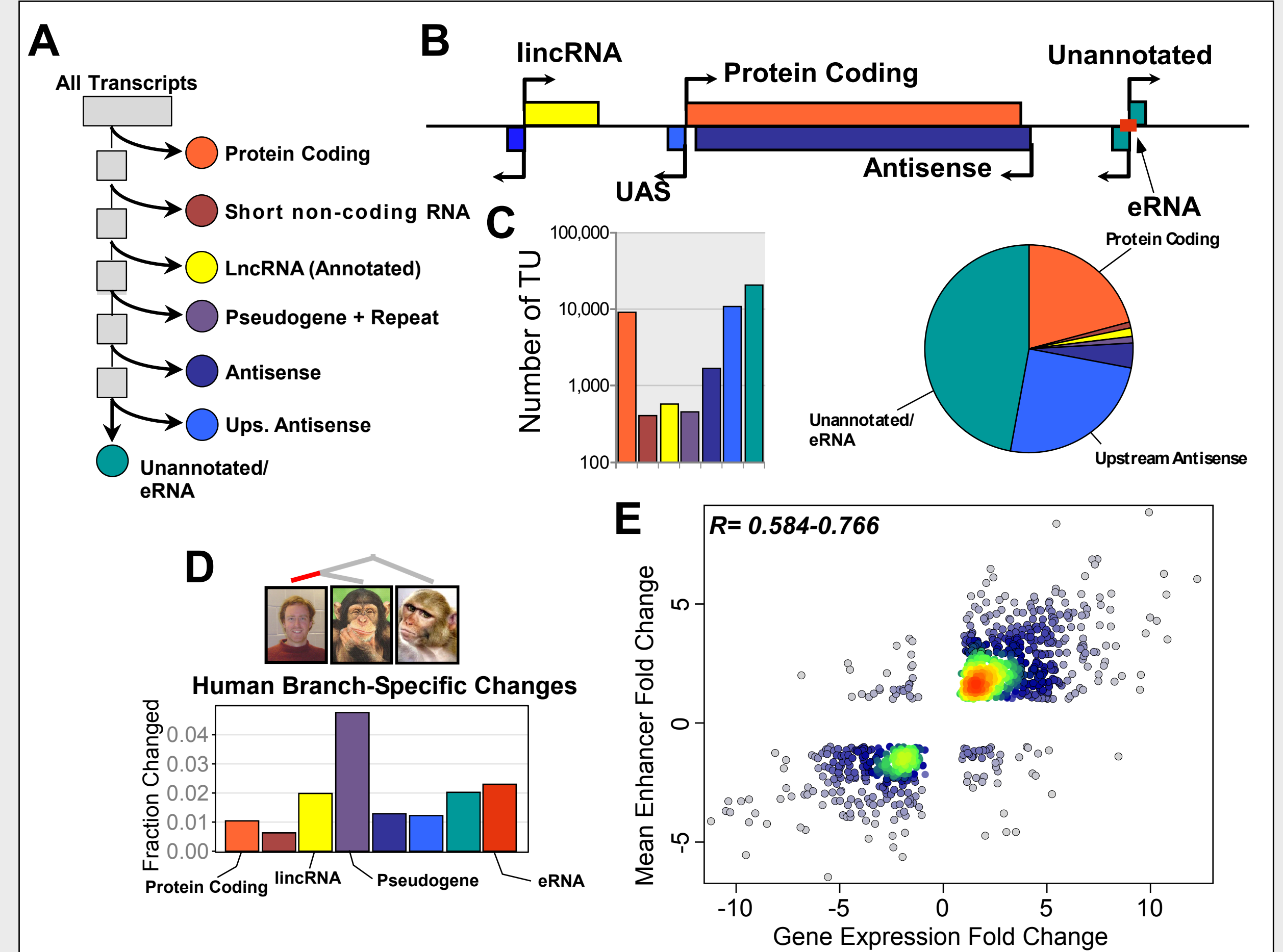


Comparing active promoters and enhancers across primate species: (A) dREG was used to identify regulatory sites across primate species. (B) Comparison of dREG scores in the specified samples. (C-E) INSIGHT was used to assess modes of natural selecting acting in dREG sites with human specific expression patterns.

Comparing transcription units.



De novo Identification of transcription units (TU): (A-B) groHMM was used to identify TU. (C) Browser shot compares transcripts identified.



Classification of transcription units (TU) active in primate CD4⁺ T-cells: (A-B) TU were sorted in seven non-overlapping classes. (C) Number of TU identified in each class. (D) The fraction of TU corresponding to each class observed to undergo human-specific changes in expression level. (E) Correlation between cross-species changes in gene body and distal enhancer transcription level range between 0.584 (upstream only) and 0.766, explaining 34-59% of the variance in expression level.

Conclusions.

- Changes in regulatory element activity common in primate species. Human specific changes associated with both positive and weak purifying selection.
- Changes in enhancer activity (excluding the promoter) explain almost 60% of the variance in transcription level.
- Different classes of functional element change expression at markedly different evolutionary rates.