Supplemental Note S2: Effects of altering substitution rates and generation times for simulations

Although our calculations of generation numbers and substitution rates for each branch of the evolutionary tree were based on estimates of divergence times and divergence times from recent literature, these values nevertheless are subject to uncertainty. For example, while we estimated a substitution rate of 1.20 x 10⁻⁸ substitutions per nucleotide per generation for Branches 3 and 4 based on observed divergence of 0.84% between human and chimpanzee and assuming 700,000 generations, the observed divergence between human and chimpanzee could instead have resulted from a higher substitution rate combined with a lower number of generations, or vice versa.

To test the effect of this uncertainty on our simulations, we considered two fairly extreme cases: 1) The true substitution rates in all lineages were twice as high as we estimated, while the true number of generations was halved; 2) The true substitution rates in all lineages were half as high as we estimated, while the true number of generations was doubled. We then repeated our simulations for both of these cases ("high substitution" and "low substitution") and compared our results to the results obtained using our original calculations. We find that these alterations make no difference to our inference of the magnitude of GC bias at 0.70 (difference between observed and simulated results not significant, p>0.05).

