



**Figure S5** Proportion of overlaps caused by the elongation of gene 2. Data shown corresponds to the 30% GC content scenario. The simulations were run separately with three criteria: preference for gene 2 contraction (criterion “Con”); gene 2 elongation/contraction equally probable (criterion “Both”); preference for gene 2 elongation (criterion “Elong”). “Prop = F”, start codons were chosen at random, “Prop = T”, start codons chosen according to empirical codon usage in prokaryotic genomes (80% ATG, 17% GTG and 3% TTG). Each scenario was replicated  $10^6$  times. In all simulated scenarios, the formation of overlapping regions originated by the elongation of the 3'-end of gene were significantly more frequent than those originated by the elongation of gene 2 ( $p$ -values  $\leq 0.001$ ).