

The rate of spontaneous mutations in yeast deficient for MutS β function

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Supplementary Tables

Supplementary Table 1

Mutation accumulation and corresponding SRA identifiers

Supplementary Table 2

SNMs identified in MA strains outside of SSR loci; note that the two mutations in strain *A4* on chromosome XI are within 19 bp of each other, and therefore counted as part of a single locus.

Supplementary Table 3

% genome that is above 10x coverage in *both* the MA strain listed and its ancestor, and which is not part of a repeat (telomeric, centromeric, or LTR regions)

Supplementary Table 4

Insertions/deletions identified in MA strains; ancestral sequence listed on top, MA line sequence on the bottom

Supplementary Table 5

Mutations in SSR loci; ancestral sequence listed on top, MA line sequence on the bottom. A single SSR locus is found in two strains. One mutation (Chrom VII:675527-675544) is likely to be a false positive: it is borderline significant, and the same mutation is called in many other strains at ΔGL below the ΔGL quantile threshold we implemented.

Supplementary Table 6

Primers used for amplifying the *MSH3* knockout cassette and genotyping. For genotyping primers, the F primer of the pair was typically used for Sanger sequencing the PCR product.

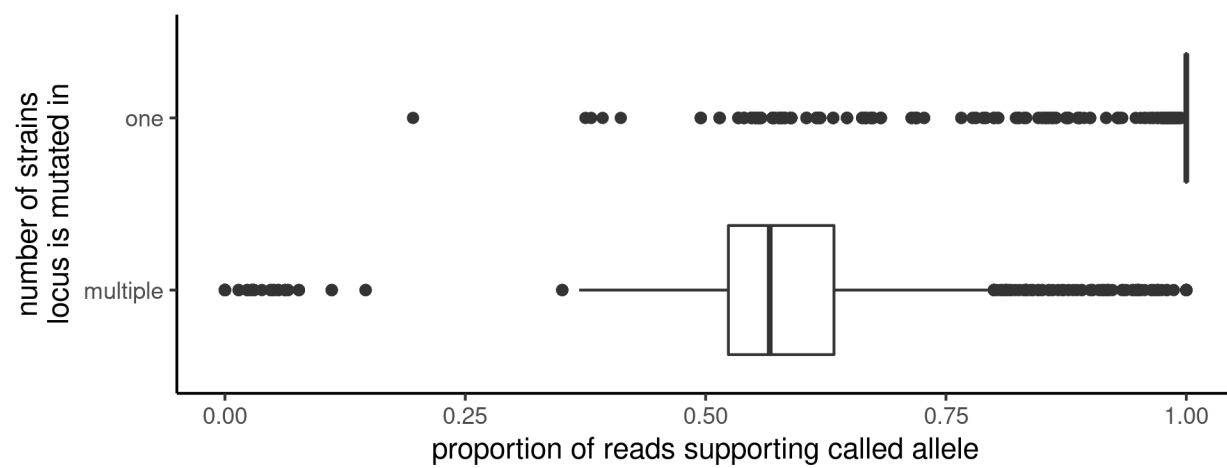
Supplementary Table 7

List of SSR loci used in this study

Supplementary Table 8

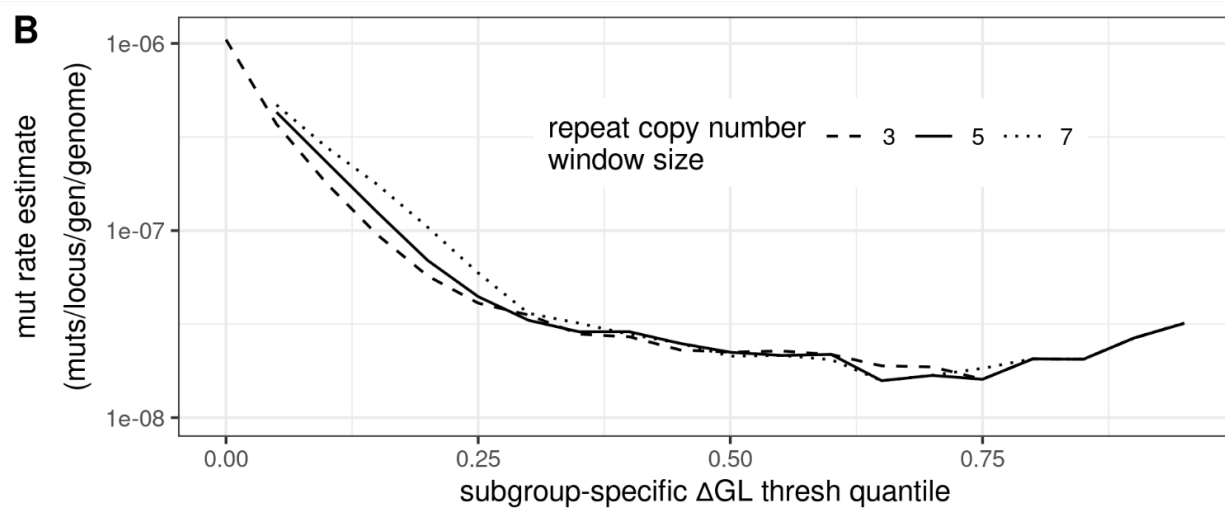
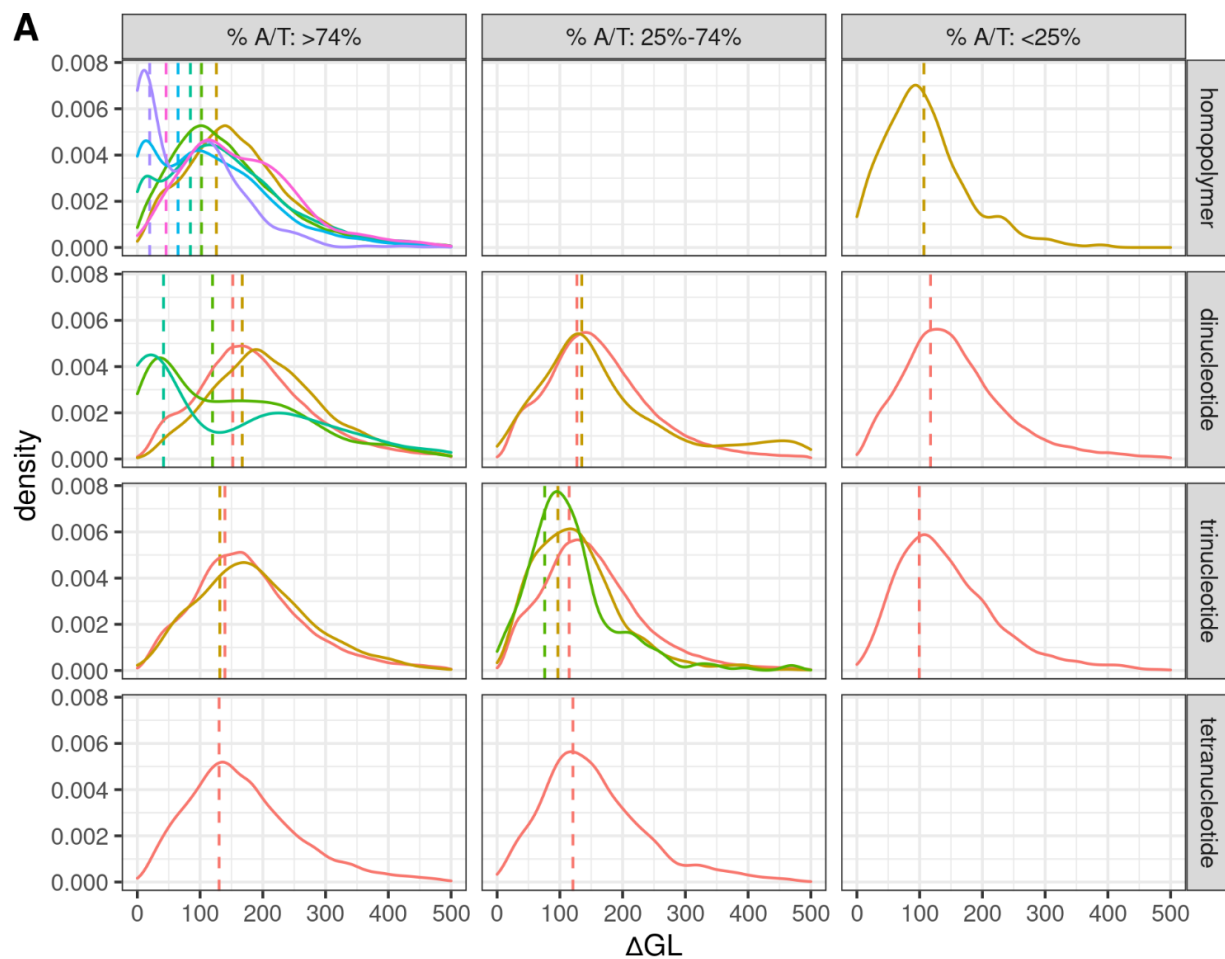
List of genomic regions excluded in this study due to being part of LTR, telomere, centromere, or ribosomal array (not including the 100-bp buffer used in calling mutations)

Supplementary Figures



Supplementary Figure 1

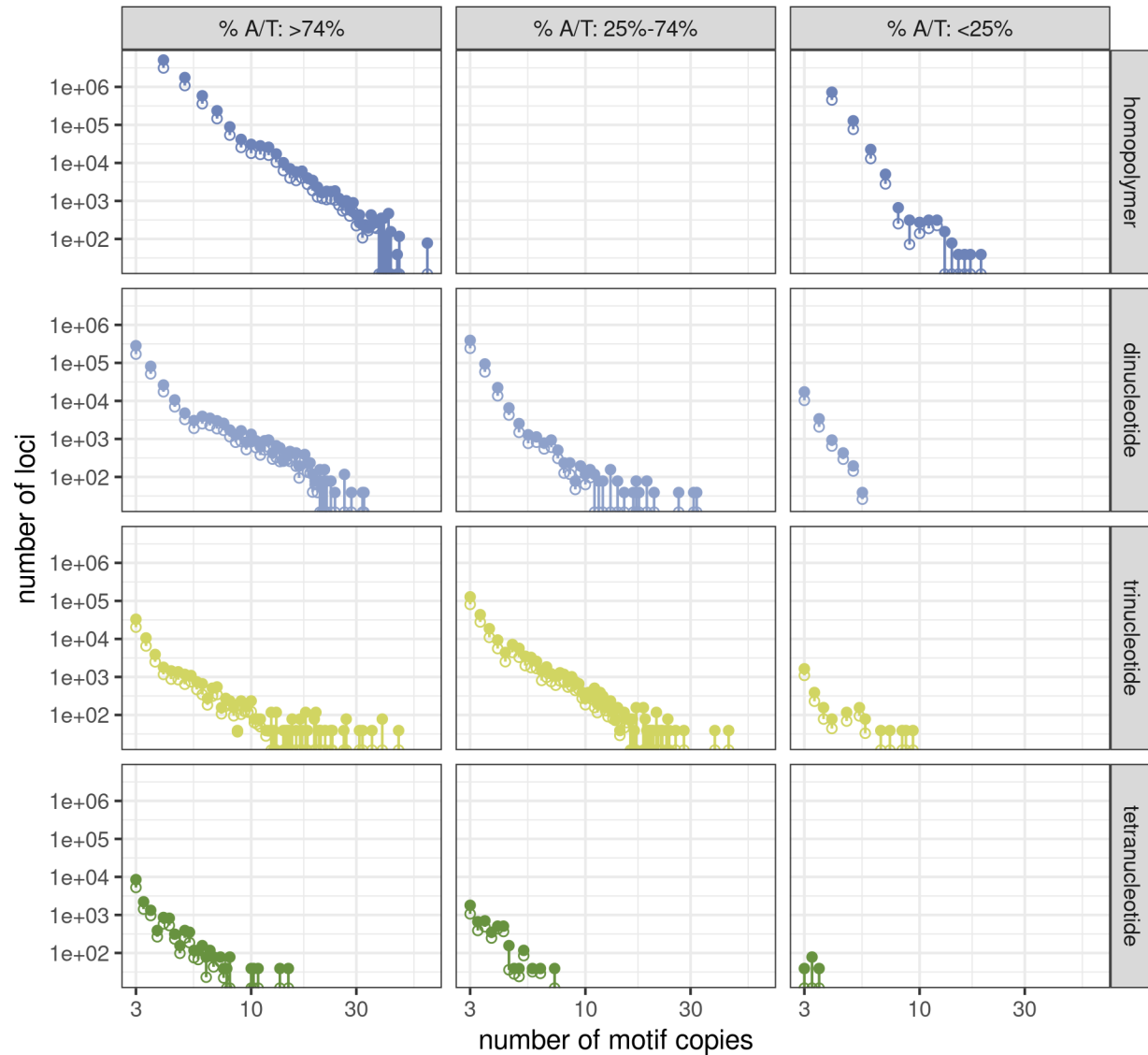
Allele calls for loci mutated in multiple strains have a low proportion of reads supporting them



Supplementary Figure 2: Filtering of SSR locus calls

(A) Empirical distributions of ΔG values across SSR locus properties. For representation clarity, thresholds for only a subset of motif copy numbers are shown. Dotted line in each plot represents the 35th-percentile cutoff ΔG value for each category; distributions include ΔG values for all SSR calls within the repeat copy number window specified in parentheses. Locus call ΔG values vary across locus properties, but setting a within-group ΔG percentile-based threshold ensures that mutations in loci that have generally lower call confidence can still be identified.

(B) Choice of grouping window size for repeat copy numbers during quantile-based ΔG filtration does not substantially affect the estimated mutation rate. Solid line corresponds to **Figure 2C**.



Supplementary Figure 3: Filtering of SSR locus calls

Removing rare groups of loci in filtration does not significantly bias the types of loci genotyped by motif complexity or A/T proportion. Solid circles represent the total number of potential calls in the dataset (locus number x number of MA strains) corresponding to a motif complexity, A/T proportion, and copy number; empty circles represent the total number of calls made after filtration. With the exception of long SSR loci (>35 base pairs), which are rare in the yeast genome, a consistent proportion of potential calls in each category passed filtration.