

Supplementary File to the article:

Genomic evidence for intraspecific hybridization in a clonal and extremely halotolerant yeast

by

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Figure S1. Mapping of sequencing reads from strains C (upward pointing histograms) and D (downward pointing histograms) to assembled diploid *H. werneckii* genomes. The mapping depth was calculated as a moving median in a 1000 nt window for all contigs larger than 2500 nt.

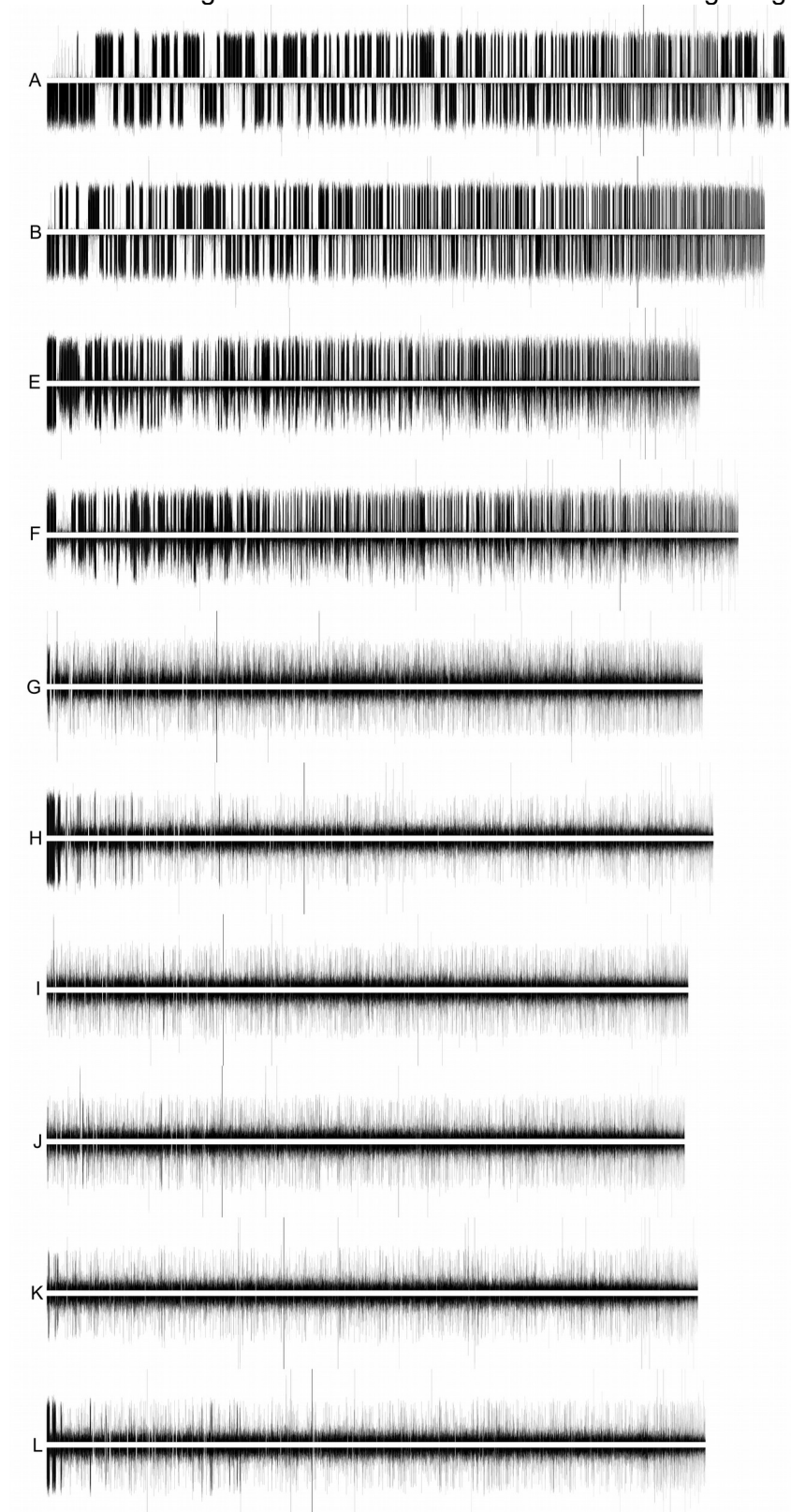
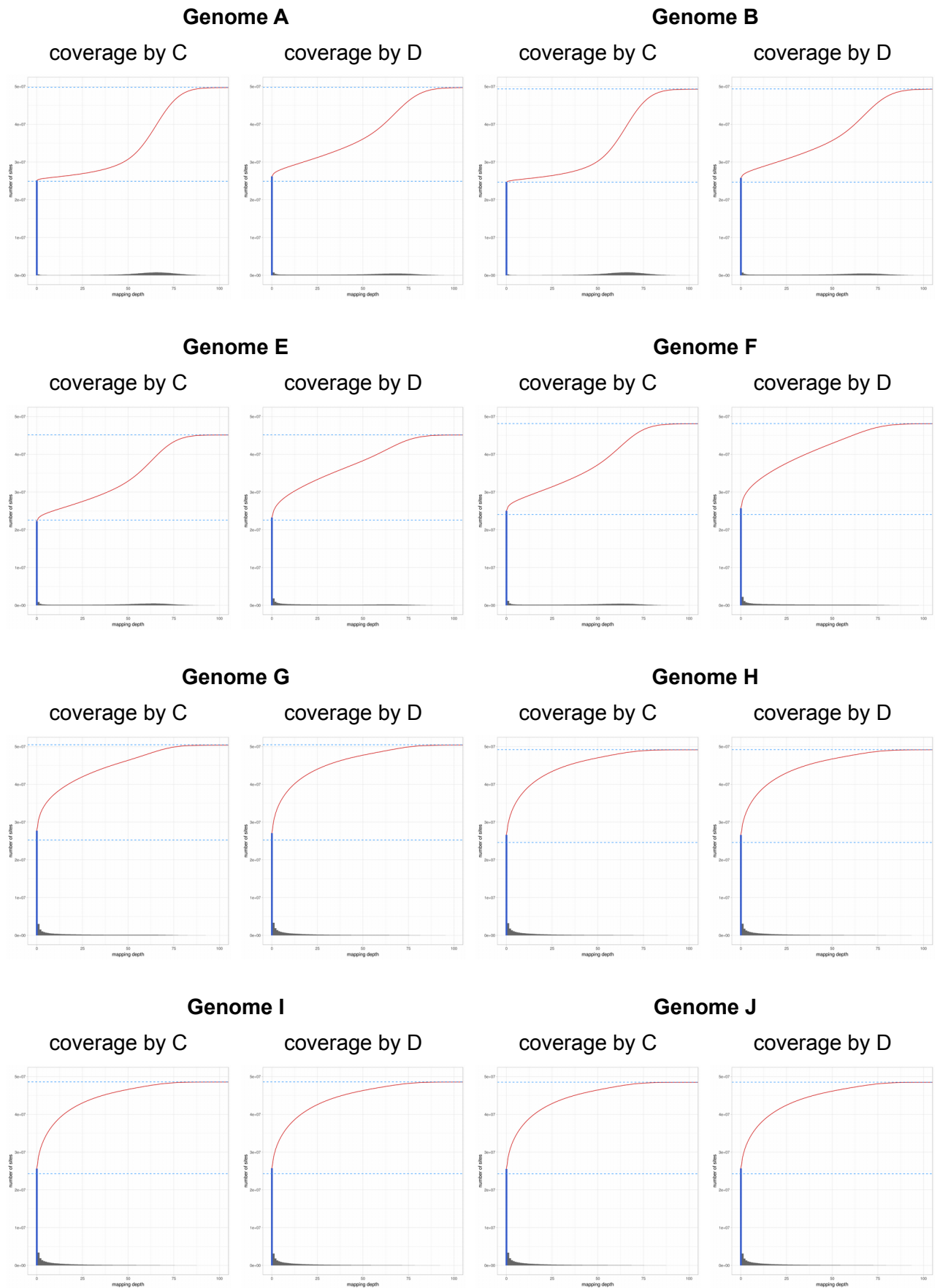
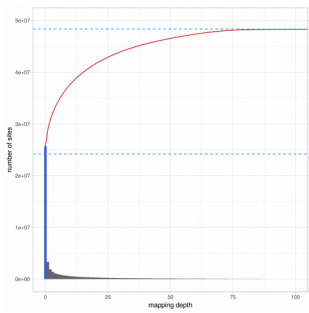


Figure S2. Number of sites with specific coverage by reads from haploid *H. werneckii* genomes C and D in diploid *H. werneckii* genomes. The histograms show the frequency of sites with coverage from 0 to 100. The number of sites with zero coverage from either genome is represented by a blue column. The red curves show the cumulative frequency of sites with specific coverage. Dashed blue lines mark the diploid (upper line) and haploid (lower line) genome sizes.

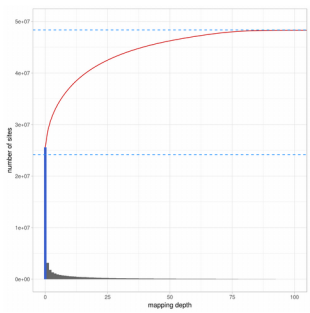


Genome K

coverage by C

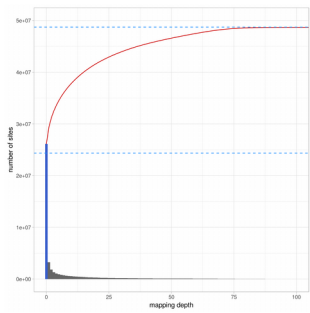


coverage by D



Genome L

coverage by C



coverage by D

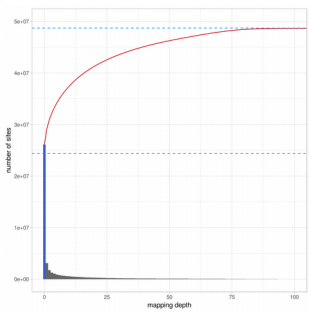
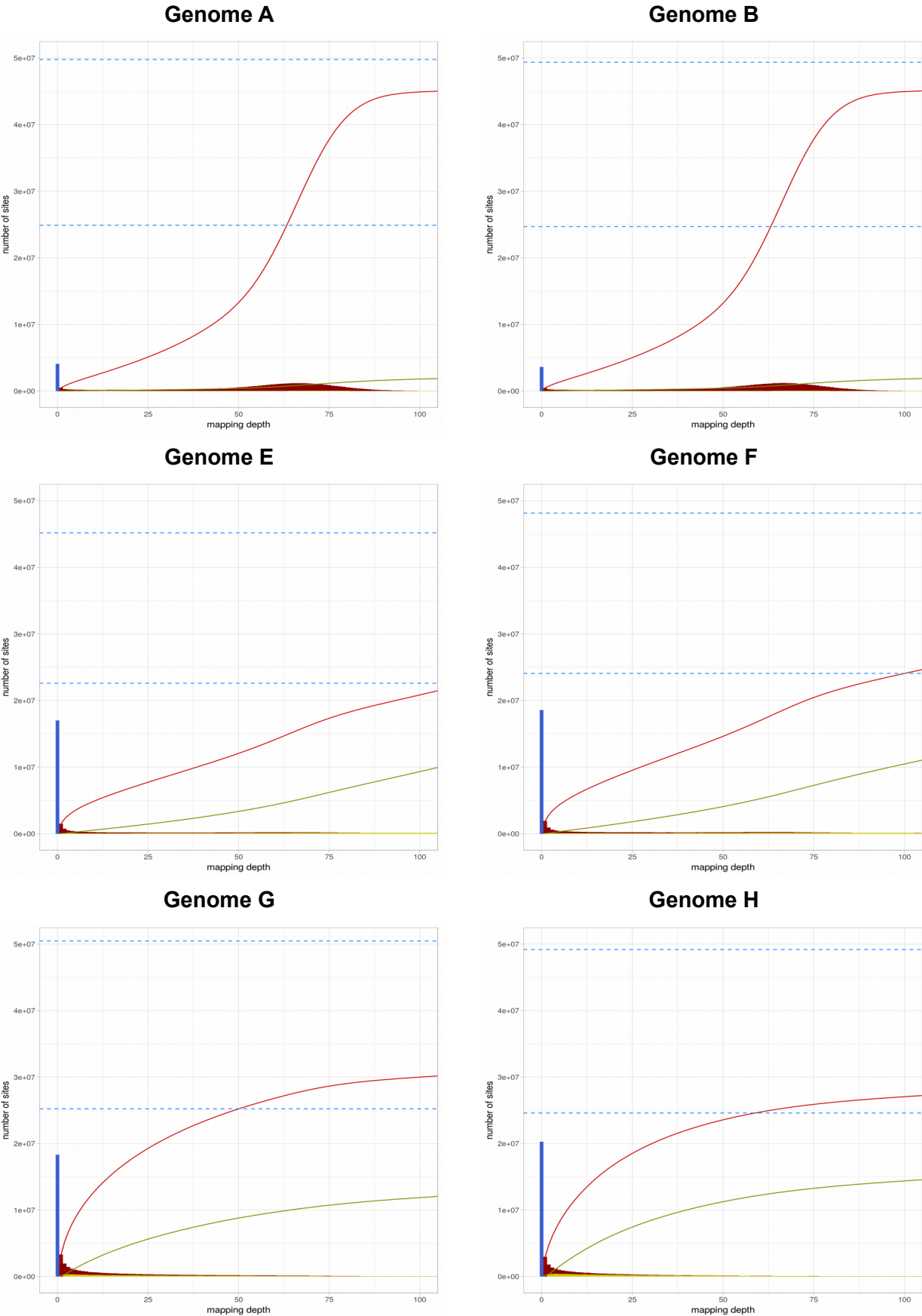
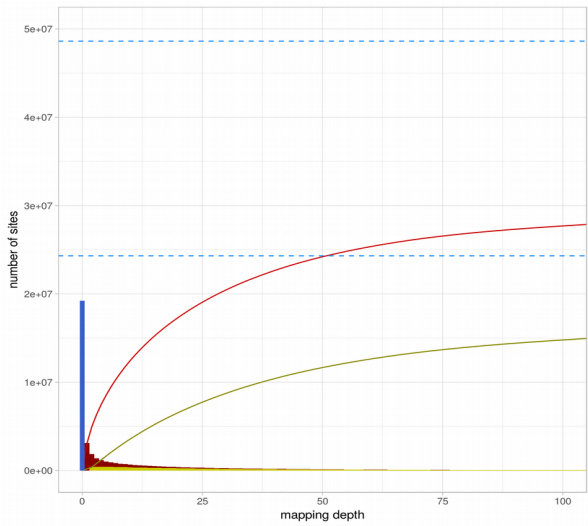


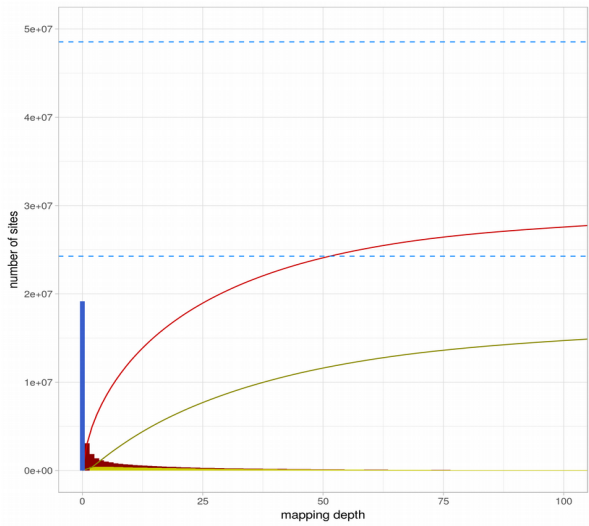
Figure S3. Number of sites with summarized coverage by reads from haploid *H. werneckii* genomes C and D in diploid *H. werneckii* genomes. The red histograms show the frequency of sites with summarized coverage by reads from both C and/or D. The yellow histograms show only sites covered by reads from *both* C *and* D. The numbers of sites with zero coverage from either genome are represented by blue columns. Red and yellow curves show the cumulative frequencies corresponding to red and yellow histograms, respectively. Dashed blue lines mark the diploid (upper line) and haploid (lower line) genome sizes.



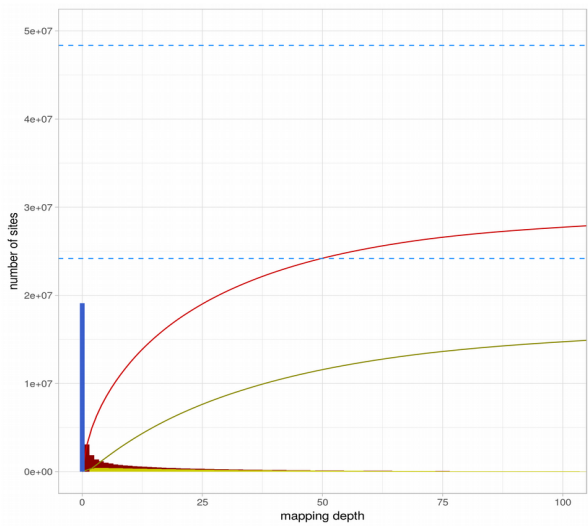
Genome I



Genome J



Genome K



Genome L

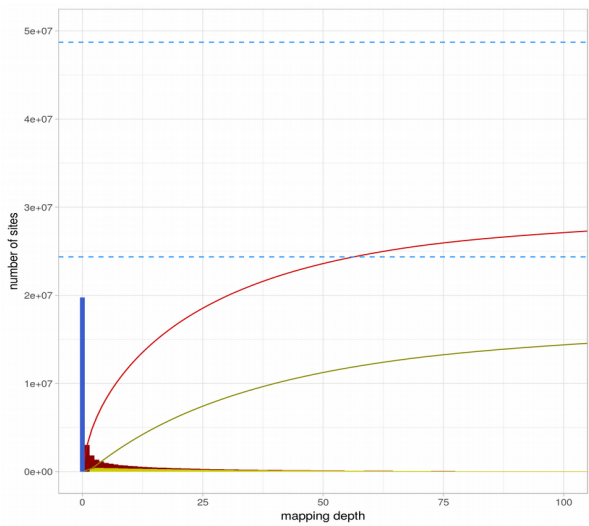


Figure S4. Alignment of assembled genomes of strains C and D to the reference genome A.
Only contigs longer than 25 kbp and best aligning regions are shown.

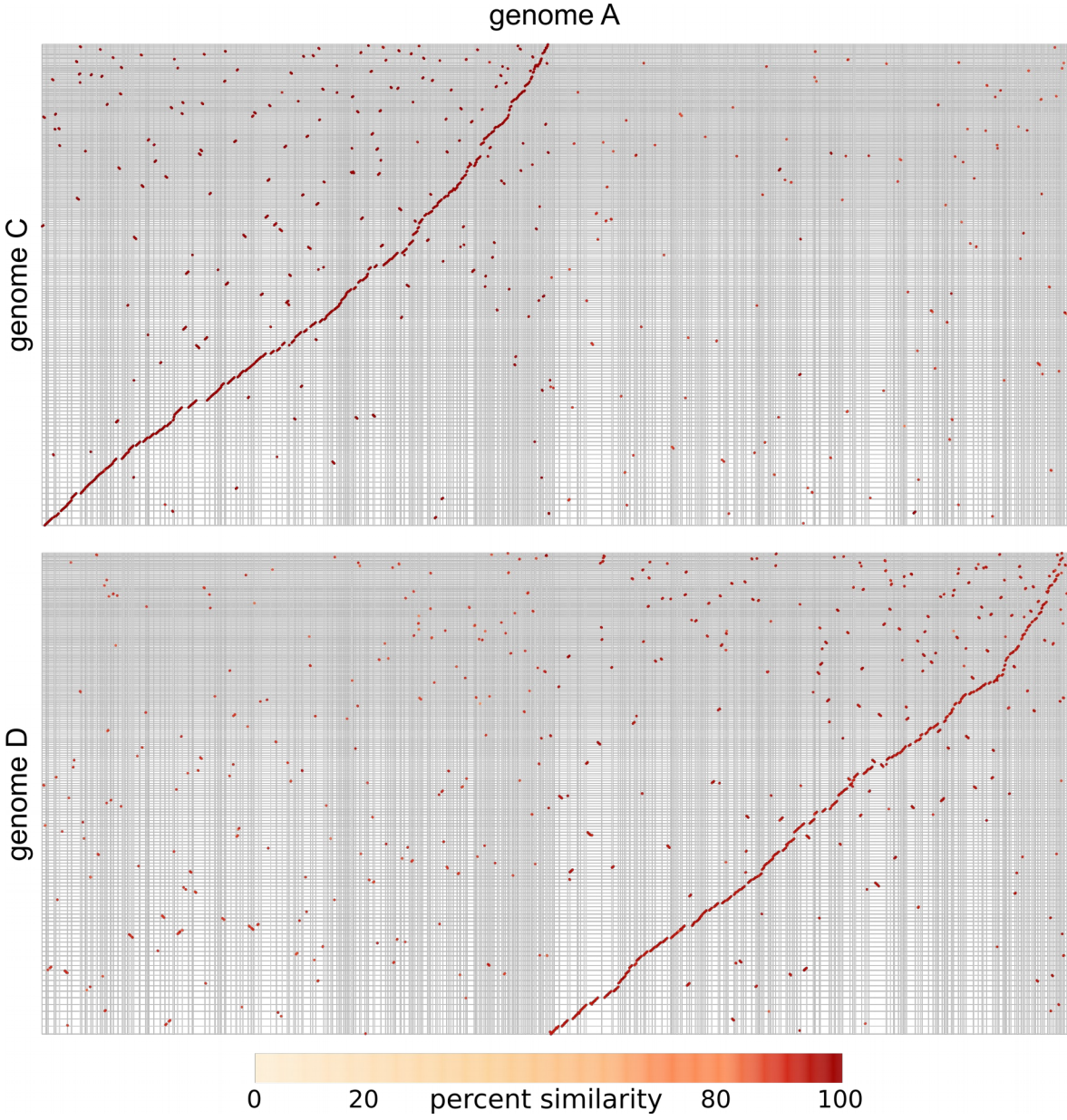


Figure S5. Phylogeny of taxonomic markers. Maximum likelihood phylogenies based on genes encoding RNA polymerase II and beta tubulin in twelve strains of *H. werneckii*. The consensus phylogeny based on 1273 phylogenies of core genes (left; as shown in Fig. 4) is provided for comparison.

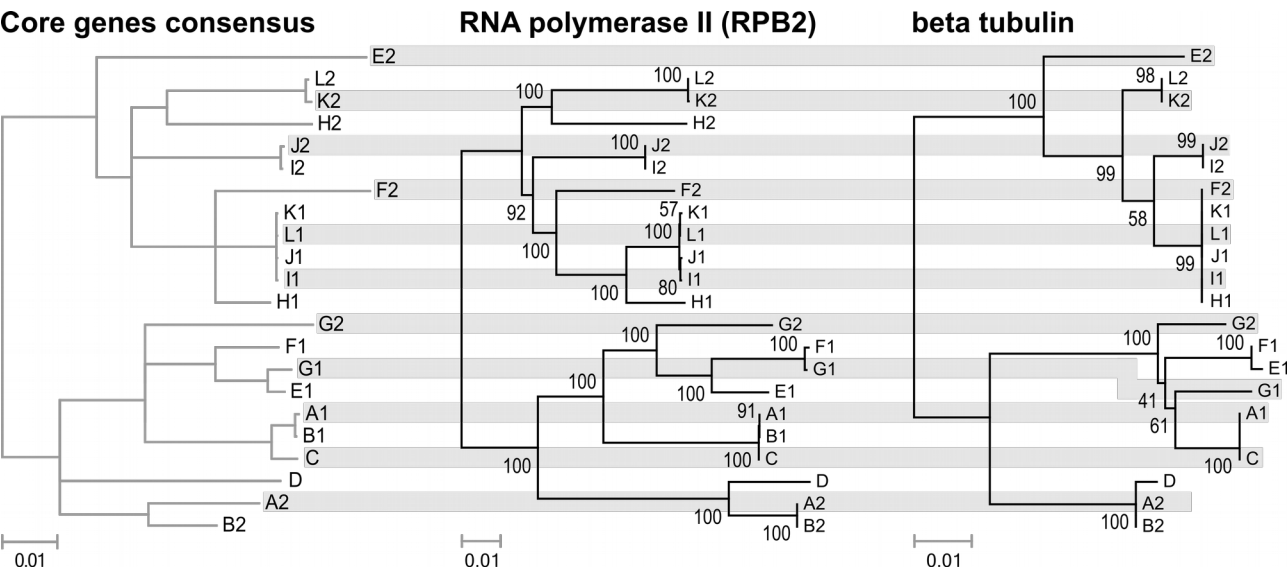


Figure S6. Test of sexuality/clonality. The index of association $\bar{r}_d$ was calculated on ten random sampled data subsets (the figure is representative for all of them), each containing 1000 SNPs. 999 permutations were used to estimate the p-value for the rejection of the null hypothesis (that the loci are not linked and the population is sexual). The observed index of association is marked with the vertical dashed blue line, while the expected indices for non-linked (permuted) data are shown as a gray histogram. Data corrected for clonality produced the same result.

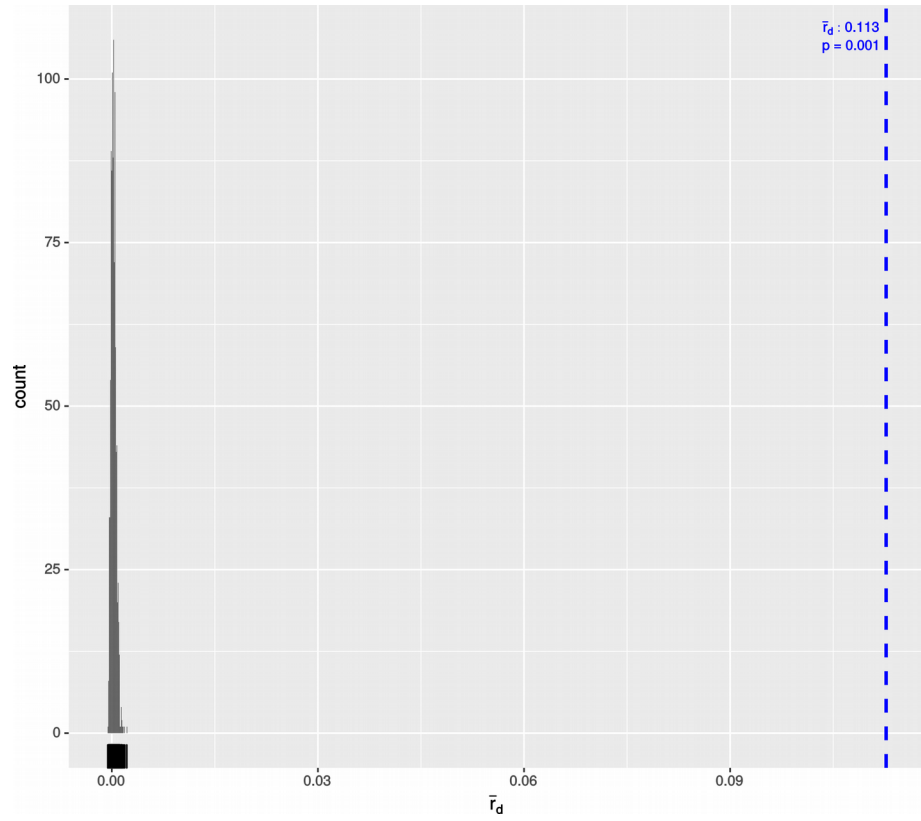


Figure S7. Filtering of SNP data by depth of coverage. To avoid the underestimation of heterozygosity due to mapping of sequencing reads from only one of the two subgenomes within a diploid genome, all variants with low coverage were removed in the filtering step of the variant calling pipeline. SNPs between the blue and red vertical lines were included in the final dataset.

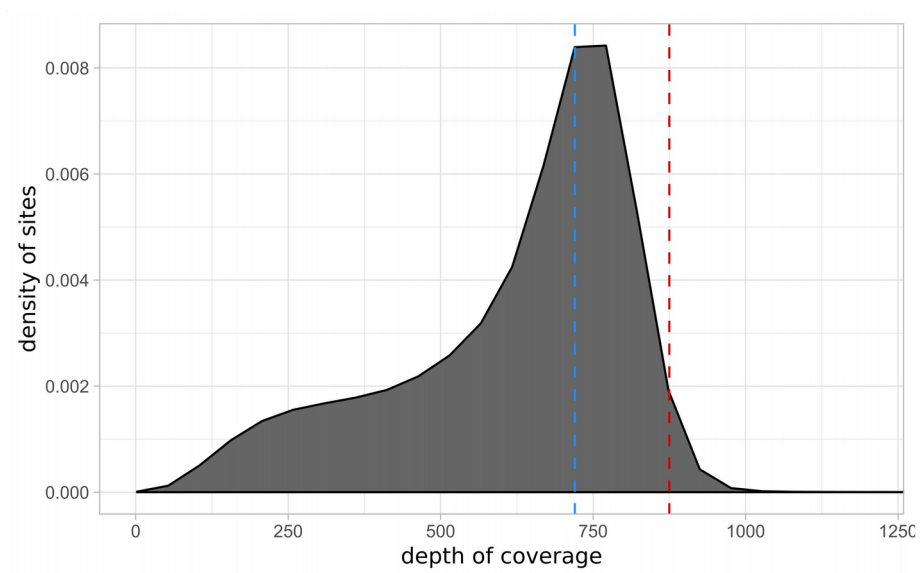


Table S1. Numbers of identified Benchmarking Universal Single-Copy Orthologs in the assembled genomes and predicted proteomes of *H. werneckii*, demonstrating high levels of completeness of the genomes and proteomes and a large proportion of duplicated genes/proteins in diploid strains.

Genome	B	C	D	E	F	G	H	I	J	K	L
Complete BUSCOs	278	282	283	282	282	255	269	267	268	269	274
Complete and single-copy BUSCOs	12	282	283	69	43	76	72	66	87	62	62
Complete and duplicated BUSCOs	266	0	0	213	239	179	197	201	181	207	212
Fragmented BUSCOs	7	2	3	6	4	28	17	20	16	16	11
Missing BUSCOs	5	6	4	2	4	7	4	3	6	5	5
Total BUSCO groups searched	290	290	290	290	290	290	290	290	290	290	290

Predicted proteome	B	C	D	E	F	G	H	I	J	K	L
Complete BUSCOs	278	278	281	277	275	258	263	263	264	265	269
Complete and single-copy BUSCOs	21	278	281	68	45	81	66	69	88	61	60
Complete and duplicated BUSCOs	257	0	0	209	230	177	197	194	176	204	209
Fragmented BUSCOs	9	6	6	9	10	25	21	19	21	21	17
Missing BUSCOs	3	6	3	4	5	7	6	8	5	4	4
Total BUSCO groups searched	290	290	290	290	290	290	290	290	290	290	290

Table S2. Percent of reads mapped to the reference *H. werneckii* genome (genome A) with the bwa aligner including and excluding reads aligning to more than one locus and using different minimum thresholds (T) for the alignment score.

strain	default bwa parameters	uniquely aligning reads with different alignment score thresholds (T)			
		T=30	T=60	T=90	T=95
B	99%	92%	92%	92%	92%
C	98%	87%	87%	87%	87%
D	94%	72%	71%	71%	71%
E	86%	47%	46%	45%	45%
F	85%	45%	44%	43%	43%
G	88%	54%	53%	52%	52%
H	79%	35%	34%	33%	33%
I	81%	36%	34%	33%	33%
J	82%	35%	34%	33%	33%
K	81%	36%	34%	33%	33%
L	78%	34%	33%	32%	32%

Table S3. Proportion of genomes that could be uniquely aligned to homologous regions within the same genome of diploid *H. werneckii* strains and the average nucleotide identity within the alignments.

Genome	A	B	E	F	G	H	I	J	K	L
% genome aligning within the genome	82.45	81.42	67.30	71.43	93.66	86.19	87.37	87.74	87.09	84.57
% identity of aligned regions	89.38	89.04	88.31	88.40	91.92	91.58	91.87	91.86	91.66	91.64