

## Gene amplification as a mechanism of yeast adaptation to nonsense mutations in release factor genes

Evgeniia M. Maksiutenko<sup>1,2</sup>, Yury A. Barbitoff<sup>1,3</sup>, Andrew G. Matveenko<sup>1</sup>, Svetlana E. Moskalenko<sup>1,2</sup>, Galina A. Zhouravleva<sup>1,4</sup>

<sup>1</sup> - Dpt. of Genetics and Biotechnology, St. Petersburg State University, St. Petersburg, Russia

<sup>2</sup> - St. Petersburg Branch, Vavilov Institute of General Genetics of the Russian Academy of Sciences, St. Petersburg, Russia

<sup>3</sup> - Bioinformatics Institute, St. Petersburg, Russia

<sup>4</sup> - Laboratory of Amyloid Biology, St. Petersburg State University, St. Petersburg, Russia

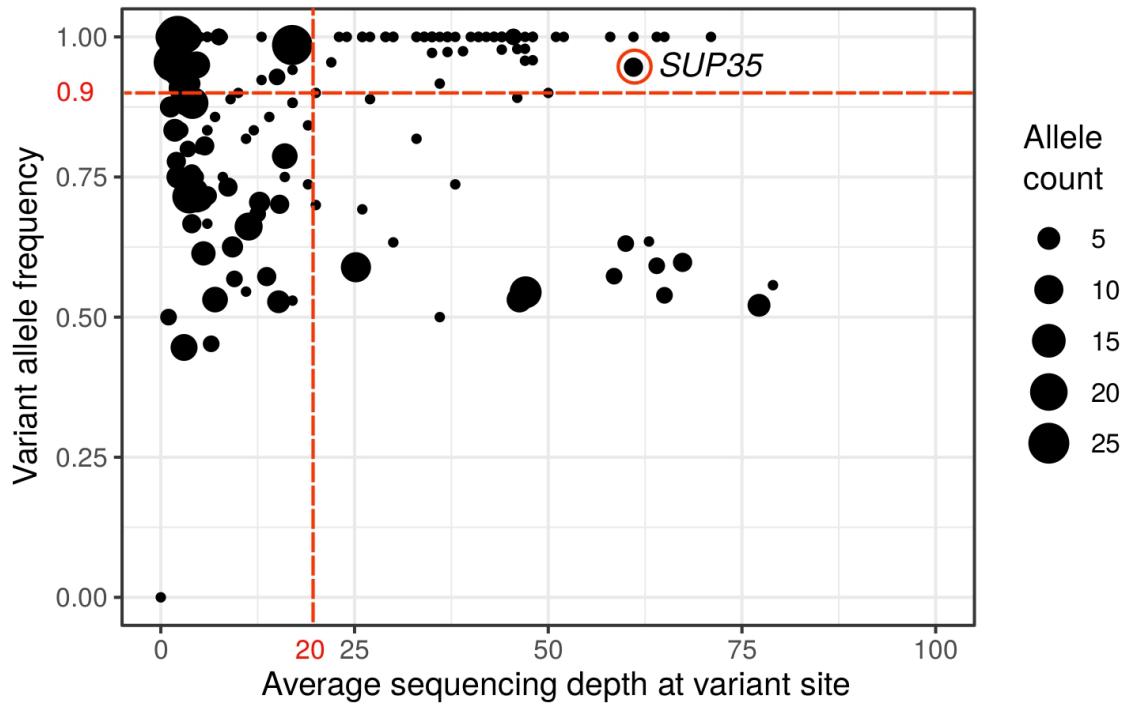
### Supplementary Tables

**Table S1.** Variants in the plasmids carrying mutant alleles *sup35-n* and *sup45-n*. Position and region annotations conform to the pRSU1 and pRS315-SUP45 maps (Supplementary File S1) in cases of *sup35* and *sup45* alleles, respectively.

Allele	Type	Substitution	Position	Region	Number of clones
<i>sup35-n</i>	Insertion	T > TCTAG	5110	MCS	34
<i>sup35-21</i>	SNP	C > T	2964	<i>SUP35</i> ORF	9
<i>sup35-74</i>	SNP	C > T	2088	<i>SUP35</i> ORF	8
<i>sup35-74</i>	SNP	A > G	2391	<i>SUP35</i> ORF	8
<i>sup35-74</i>	SNP	A > G	2594	<i>SUP35</i> ORF	8
<i>sup35-74</i>	SNP	T > C	2800	<i>SUP35</i> ORF	8
<i>sup35-74</i>	SNP	T > C	10205	Bacterial ori	8
<i>sup35-218</i>	SNP	G > T	2241	<i>SUP35</i> ORF	9
<i>sup35-218</i>	Deletion	TTA > T	3849	Unannotated region	2
<i>sup35-218</i>	Deletion	GA > G	8463	ARSH4	9
<i>sup35-218</i>	SNP	A > C	10117	Bacterial ori	7

<i>sup35-240</i>	SNP	C > T	1866	<i>SUP35</i> ORF	8
<i>sup35-240</i>	Deletion	TTA > T	3849	Unannotated region	1
<i>sup45-n</i>	SNP	A > G	1715	<i>SUP45</i> ORF	55
<i>sup45-n</i>	SNP	G > T	2165	<i>SUP45</i> ORF	55
<i>sup45-n</i>	SNP	T > C	2333	<i>SUP45</i> ORF	55
<i>sup45-n</i>	SNP	T > A	2348	<i>SUP45</i> ORF	55
<i>sup45-n</i>	SNP	G > A	2573	<i>SUP45</i> ORF	55
<i>sup45-n</i>	SNP	T > C	2654	<i>SUP45</i> ORF	55
<i>sup45-101</i>	SNP	A > T	1651	<i>SUP45</i> ORF	2
<i>sup45-101</i>	SNP	G > T	2442	<i>SUP45</i> ORF	11
<i>sup45-102</i>	SNP	T > A	1805	<i>SUP45</i> ORF	11
<i>sup45-104</i>	SNP	T > A	2494	<i>SUP45</i> ORF	11
<i>sup45-105</i>	SNP	A > T	891	<i>SUP45</i> promoter	1
<i>sup45-105</i>	SNP	G > T	2799	<i>SUP45</i> ORF	11
<i>sup45-105</i>	SNP	G > A	3380	Unannotated region	1
<i>sup45-105</i>	SNP	G > A	6151	Unannotated region	1
<i>sup45-105</i>	Deletion	AT > A	6352	<i>LEU2</i> terminator	1
<i>sup45-107</i>	SNP	T > G	2596	<i>SUP45</i> ORF	11

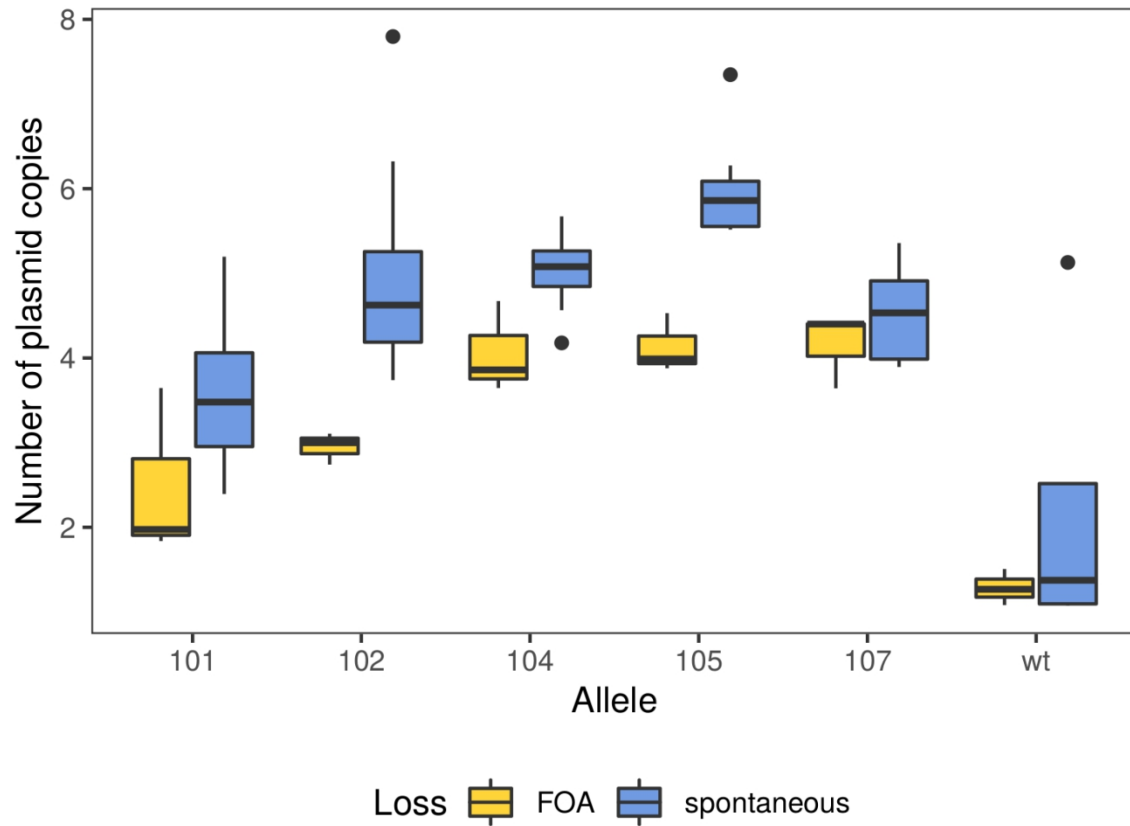
## Supplementary Figures



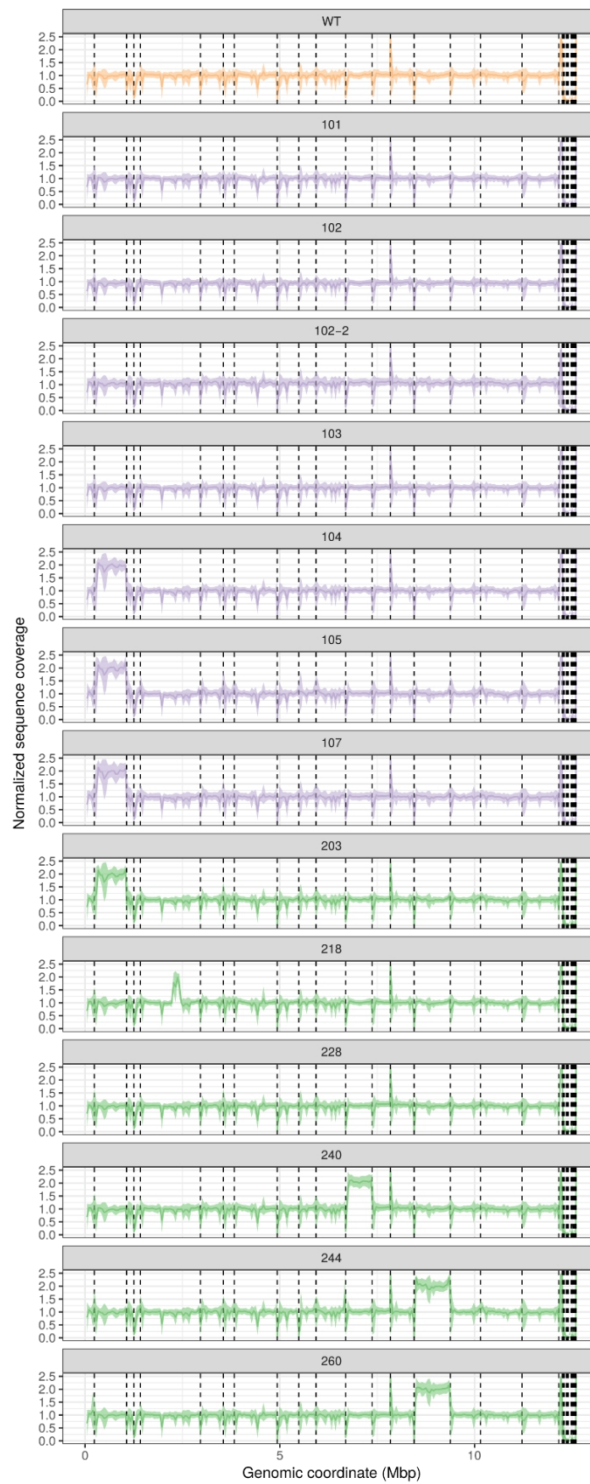
**Figure S1.** A scatter plot representing the results of variant calling against the reference genome sequence of the U-1A-D1628 strain. Each dot represent a variant site, size of the dot is proportional to the variant allele count in the set of 8



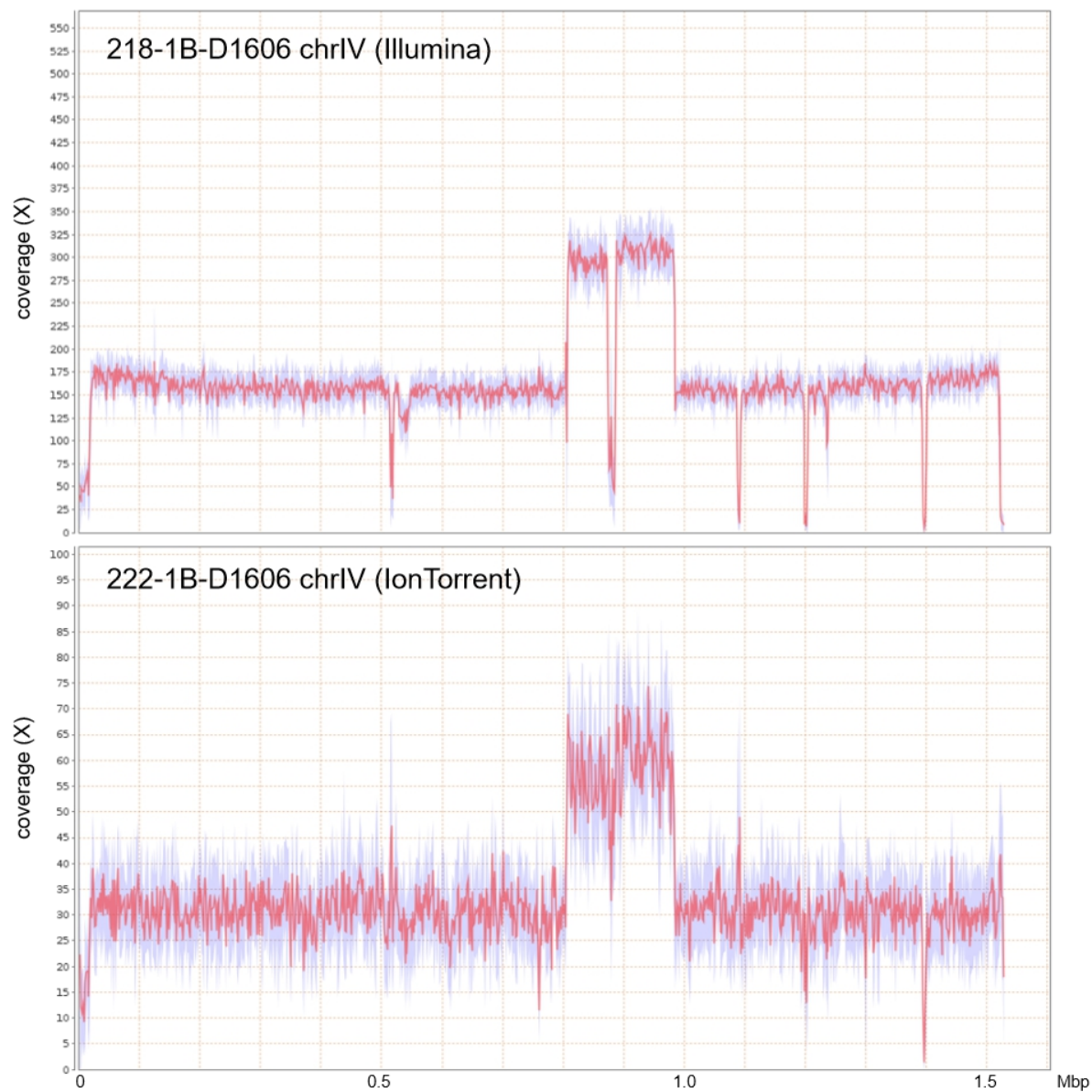
**Figure S2.** Coverage distribution in mutant and wild-type strains across plasmid sequences.



**Figure S3.** Plasmid copy number estimation in strains that were selected using 5-FOA (yellow) or spontaneously lost the plasmid containing the wild-type *SUP45* allele (blue).



**Figure S4.** Normalized sequence coverage profiles for all 1B-D1606 strain derivatives analysed. Color corresponds to the mutant gene (orange - wild-type, purple - *sup45-n*, green - *sup35-n*) Dashed lines correspond to contig boundaries. Two clones from two different sources with confirmed *sup45-102* allele (denoted as “102” and “102-2”) were analyzed.



**Figure S5.** Comparison of the chromosome IV coverage profiles for the 218-1B-D1606 and 222-1B-D1606 sequences aligned onto the 74-D694 genome (Barbitoff et al., 2021).

**Supplementary File S1.** A ZIP archive containing plasmid sequences in the SnapGene format.