**Table S1. List of Primers**

|  |  |
| --- | --- |
| *AFR3* + p*GAL1* | CCTCTATACTTTAACGTCAAGGAGAAAAAAATGTCTGCCATCGCCATCGA |
| *AFR3* + *CYC1* tt | TGAATGTAAGCGTGACATAACTAATTACATGATTTACTTGCGCAACGTGAGAG |
| *AFR3* F | ATGTCTGCCATCGCCATCGA |
| *AFR3* R | TTACTTGCGCAACGTGAGAG |
| qPCR *AFR3* F | GCCCATAGCACTAGGAGTCG |
| qPCR *AFR3* R | ATGTCATCCTGCGAATAGCC |
| Sc *ACT1* F | ATGGATTCTGAGGTTGCTGC |
| Sc *ACT1* R | GGTGTCTTGGTCTACCGACG |
| Cn *ACT1* F | CCCACACTGTCCCCATTTAC |
| Cn *ACT1* R | AACCACGCTCCATGAGAATC |
| qPCR *AFR1* F | CTTTCCGAGCTGGTGAACTC |
| qPCR *AFR1* R | CACCTTCGATCACACCAATG |
| qPCR *AFR2* F | GGTTCCGACTACATGGCTGT |
| qPCR *AFR2* R | GAGTTCACCAGCTCGGAAAG |
| qPCR *MDR1* F | CTCTTGATCACATCGCGAAA |
| qPCR *MDR1* R | ACCGACAATCTTGCTCTGCT |

Graphical user interface

Description automatically generated

**Figure S1. Confirmation of *Saccharomyces cerevisiae* Transformants.** (**A**) Plasmids were extracted from ADΔ cells and PCR was performed for the Afr3 cassette. PCR samples were run in a 0.8 % agarose gel and positive colonies were identified by the correct size of amplification (2 kb); (**B**) Expression of *AFR3* in the transformed ADΔ colonies were performed by RNA extraction, followed by cDNA construction. *S. cerevisiae ACT1* gene was used as a house keeping control for *AFR3* amplification.

Chart, box and whisker chart

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**Figure S2. Pump Compensation between Afr1 and Afr3.** Cells from H99, *Δafr1,* and *Δafr3* strains had their RNA extracted, followed by cDNA conversion, and expression analysis for *AFR1* and *AFR3*. *ACT1* gene was used as a house keeping control for amplification.

Chart, line chart

Description automatically generated

**Figure S3. Growth Curves.** Cells from H99, *Δafr1*, *Δafr2,* and *Δafr3* strains were monitored in a Spectrophotometer for 72 h, at *37* °Cwith shaking.