

Supplementary information

Dynamic control of *ERG20* expression combined with minimized endogenous downstream metabolism contributes to the improvement of geraniol production in *Saccharomyces cerevisiae*

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Table S1. Primers used in this study.

Primer name	Sequences (5'→3')	Amplified genes
P _{ERG20} -UP-F	GACAATCATTACCACAAAGATGAACAC	ERG20 upstream arm
P _{ERG20} -UP-R	GCGTACGAAGCTTCACCGCTAGAACACCTC ACACTG	
P _{ERG20} -DN-F	ATGGCTTCAGAAAAAGAAATTAGG	ERG20 downstream arm
P _{ERG20} -DN-R	GTCGACTTGTCTTCAGGTGC	
kanMX-F	AGGTATTCTAAGCGGTGAAGCTTCGTACGCT GCAGG	KanMX expression cassette
kanMX-R	CGCATAGGCCACTAGTGGATCTGAT	
P _{BTSI} -F	GTGATATCAGATCCACTAGTGGCCTATGCGA CGATGTATAGCCGCCATCTC	BTSI promoter
P _{BTSI} -R	CTCTCTCTAATTCTTTCTGAAGCCATTG ATTTTCCAGACTCGTAAAC	
P _{CTR3} -F	GTGATATCAGATCCACTAGTGGCCTATGCGA AGATAATAGACAGTCATAGCATGA	CTR3 promoter
P _{CTR3} -R	CTCTCTCTAATTCTTTCTGAAGCCATAG CAGTGCTGCTACTGCCTC	
P _{HXT1} -F	GTGATATCAGATCCACTAGTGGCCTATGCGT GCAGGGTCTCATCTGGAATATAATTCC	HXT1 promoter
P _{HXT1} -R	CTCTCTCTAATTCTTTCTGAAGCCATGA TTTACGTATATCAACTAGTTGAC	
OYE2-UP-F	AAAACGGAGTAGAACATCGGTAAG	OYE2 upstream arm
OYE2-UP-R	CGTCTATATTAGCTTAATATGATG	
OYE2-DN-F	TAGTGTAAACCGTACTTGTAG	OYE2 downstream arm
OYE2-DN-R	ATAGGATGATGAATGACAGCAT	
ATF1-UP-F	ACTTTTGGACATTGAGCTAAG	ATF1 upstream arm
ATF1-UP-R	GAGAGCTGATAAATTGATGGT	
ATF1-DN-F	ATCTCACATGATGCTTGACTG	ATF1 downstream arm
ATF1-DN-R	CGACGATTCTGACCCTTCTA	
OYE2-kanMX-F	TAAATCATATTAAGCTAAATATAGACGTG AAGCTCGTACGCTGCAGG	OYE2 deletion fragment
OYE2-kanMX-R	AAATGGTGCACAAAGTACGGTTAACACTAC GCATAGGCCACTAGTGGATCT	
ATF1-kanMX-F	ATCACAAATACCATCAATTATCAGCTCTG AAGCTCGTACGCTGCAGG	ATF1 deletion fragment
ATF1-kanMX-R	GAATAATATCAGTCAGCATCATGTGAGATC GCATAGGCCACTAGTGGATCT	
LEU2-F	CAAAGGGACGTTCTCACCTCCTTGGAAAT GTGTTCCACTATCCTGTACATGAACGTGGGG	LEU2 expression cassette
LEU2-R	AATACTCAGGTATCGT CAAATATATTCCATGGCCTCTAGTTGGCAA CCCAAGACTCGGCATACCTCGACTACGTCGT TAAGGCCG	

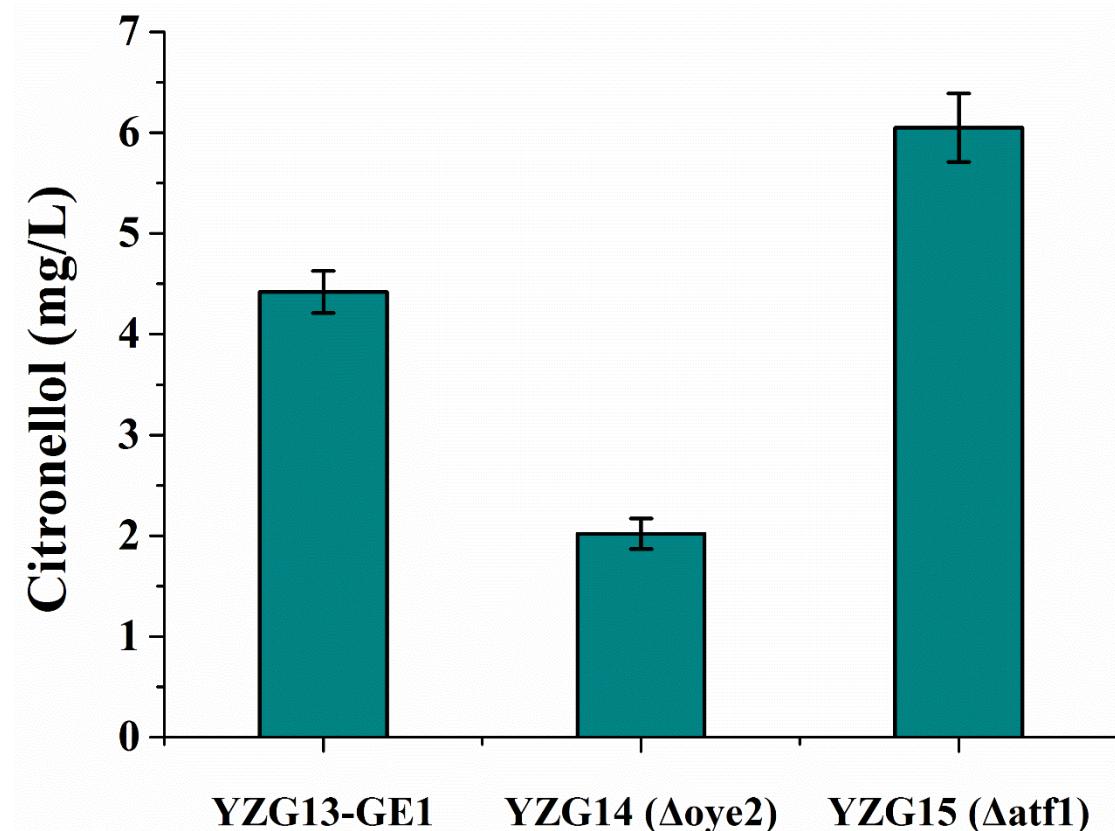


Figure S1. The conversion of geraniol to citronellol in control strain and deletion strains.

The data shown are representative of duplicate experiments, and the error bars represent the standard deviation.

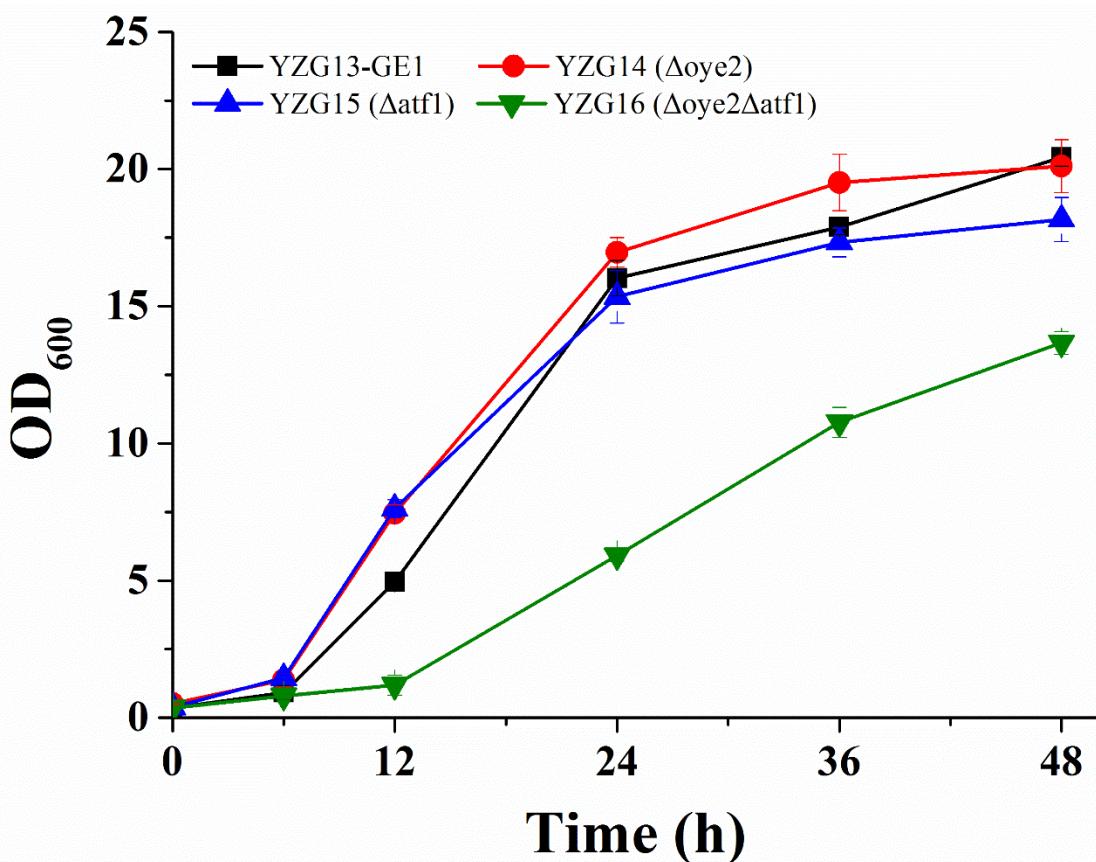


Figure S2. The effect of *OYE2* or/and *ATF1* deletion on cell growth in batch fermentation. All strains harbored pZGV6-GE1 and pZMVA4 plasmids for geraniol production. The data shown are representative of duplicate experiments, and the error bars represent the standard deviation.

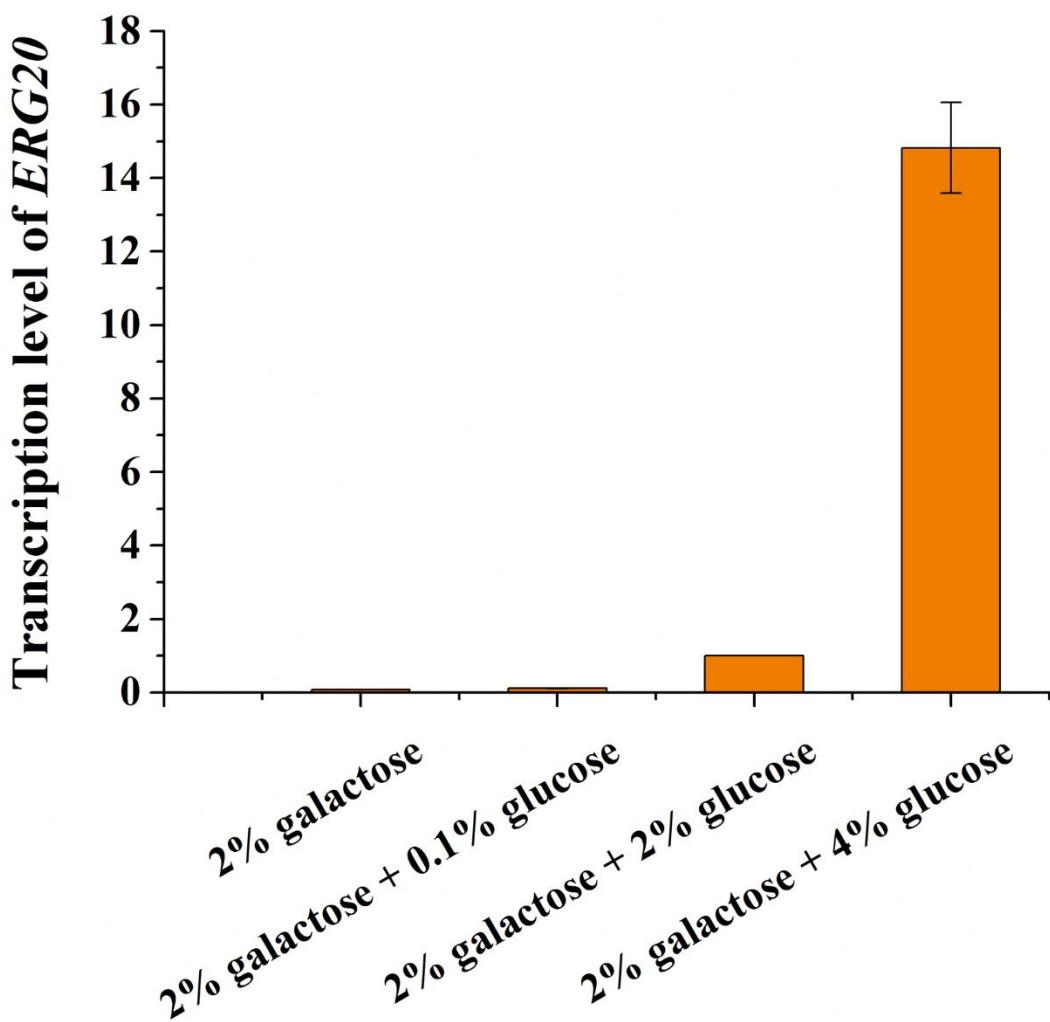


Figure S3. Transcription level of *ERG20* controlled by different promoters in SD-URA-HIS medium with different concentrations glucose. The cells were collected when OD₆₀₀ reached 0.6 to extract mRNA for *ERG20* mRNA and transcription determination. The data shown are representative of triplicate experiments, and the error bars represent the standard deviation.

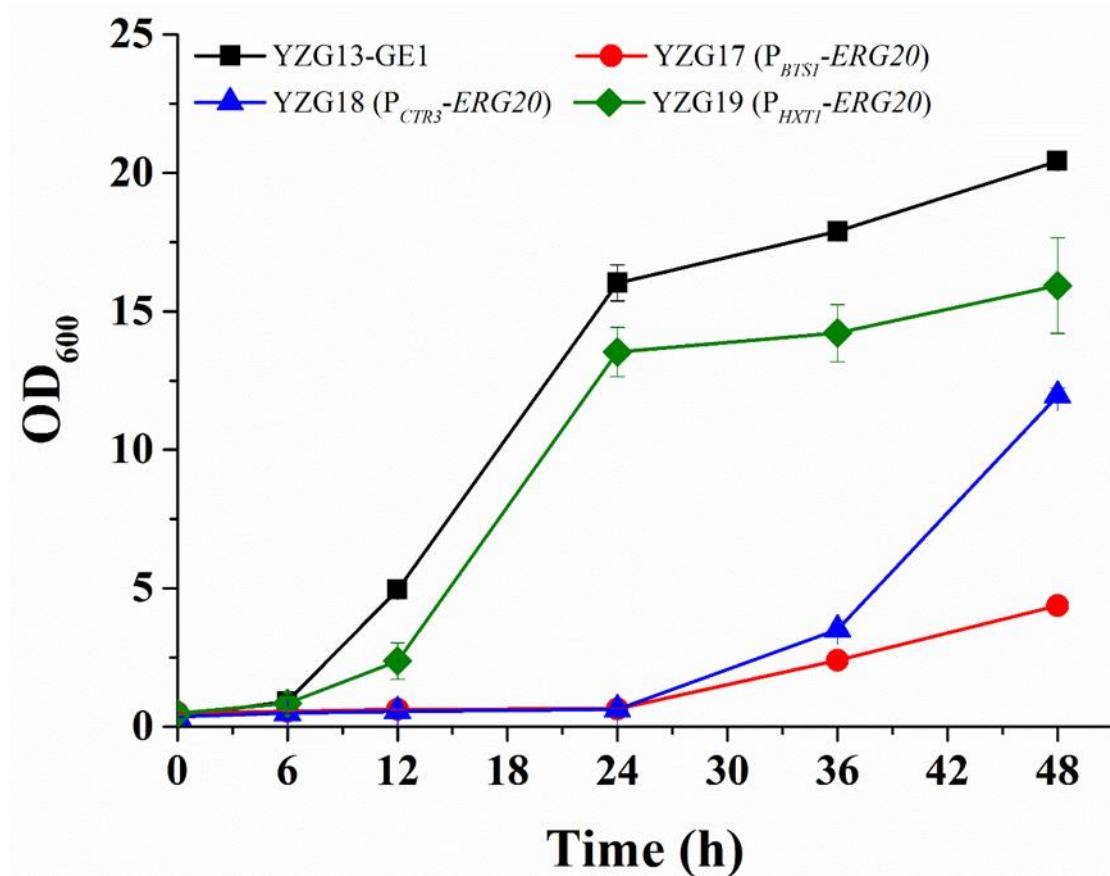


Figure S4. The effect of *ERG20* expression controlled by different promoters on cell growth in batch fermentation. All strains harbored pZGV6-GE1 and pZMVA4 plasmids for geraniol production. The data shown are representative of duplicate experiments, and the error bars represent the standard deviation.