

Supporting information

Supplementary Table 1. Strains and plasmids

Strain	Relevant genotype	Source
<i>E. coli</i>		
DH5α	<i>F- λ- endA1 glnV44 thi-1 recA1 relA1 gyrA96 deoR nupG</i> <i>Φ80dlacZΔM15 Δ(lacZYA-argF) U169, hsdR17(rK- mK+)</i>	TaKaRa (Dalian China)
<i>S. cerevisiae</i>		
BY4741	<i>MATa his3Δ1 leu2Δ0 met15Δ0 ura3Δ0</i>	ATCC
BY4742	<i>MATa his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i>	ATCC
JX1	BY4741/pYX212-GGPPS-P2A-CARRP	This study
JX2	BY4741/pYX212-CARB-P2A-CARRP	This study
JX3	BY4741/pYX212-CARRP-P2A-CARB	This study
JX4	BY4741/pYX212-GGPPS-P2A-CARB-P2A-CARRP	This study
JX5	BY4741/pYX212-CARB-P2A-GGPPS-P2A-CARRP	This study
JX6	BY4741/pYX212-CARRP-P2A-GGPPS-P2A-CARB	This study
JX7	BY4741/pYX212-GGPPS-P2A-CARRP-P2A-CARB	This study
JX8	BY4741/pYX212-CARB-P2A-CARRP-P2A-GGPPS	This study
JX9	BY4741/pYX212-CARRP-P2A-CARB-P2A-GGPPS	This study
Plasmid	Description	Source
pUC18	Amp ^R	TaKaRa
pYX212	Expression vector, <i>URA3</i> selection marker for <i>S. cerevisiae</i> , Amp ^R	Zhou <i>et al.</i> 2012
pJX1	pYX212-GGPPS-P2A-CARRP	This study
pJX2	pYX212-CARB-P2A-CARRP	This study
pJX3	pYX212-CARRP-P2A-CARB	This study
pJX4	pYX212-GGPPS-P2A-CARB-P2A-CARRP	This study
pJX5	pYX212-CARB-P2A-GGPPS-P2A-CARRP	This study
pJX6	pYX212-CARRP-P2A-CARB-P2A-GGPPS	This study
pJX7	pYX212-GGPPS-P2A-CARRP-P2A-CARB	This study
pJX8	pYX212-CARB-P2A-CARRP-P2A-GGPPS	This study
pJX9	pYX212-CARRP-P2A-CARB-P2A-GGPPS	This study
pJX11	pUC18-GGPPS-P2A-CARB-P2A-CARRP	This study
pJX12	pUC18-CARB-P2A-GGPPS-P2A-CARRP	This study
pJX13	pUC18-GGPPS-P2A-CARB-P2A-CARRP	This study

Supplementary Table 2. Primers used in this study

Primer name	Sequence (5'–3')
ADH2-EcoRI-GGPPS-F	ctgacagctgccacgtctccgacagtcacagaattcatgctcacctcgtccaagtcgac
GGPPS-P2A-BamHI-R	catggatccagggccagggttctcttcgacgtcgccagcctgcttgaggagcgagaagttggtag cgcccagaccgtgatggatgatgggtggttcgtt
BamHI-CarB-F	cctggatccatgtcggatcagaagaagcacatc
CarB-P2A-KpnI-R	catggtaccagggccagggttctcttcgacgtcgccagcctgcttgaggagcgagaagttggtag cgcccagaccgtgatggatgatgggtggtgatcg
KpnI-CarRP-F	cctggtaccatgtcgattttgacctacctcgag
CarRP-P2A-HindIII-R	cataagcttagggccagggttctcttcgacgtcgccagcctgcttgaggagcgagaagttggtag cgcccagaccgtgatggatgatgggtggtgac
ADH2-EcoRI-CarB-F	ctgacagctgccacgtctccgacagtcacagaattcatgtcggatcagaagaagcacatc
P2A-BamHI-R	catggatccagggccagggttctcttcgacgtcgccagcctgcttgaggagcgagaagttggtag cgcccagaccgtgatggatgatgggtggtg
BamHI-GGPPS-F	cctggatccatgtcacctcgtccaagtcgac
P2A-KpnI-R	catggtaccagggccagggttctcttcgacgtc
ADH2-EcoRI-CarRP-F	ctgacagctgccacgtctccgacagtcacagaattcatgtcgattttgacctacctcgag
KpnI-CarB-F	cctggtaccatgtcggatcagaagaagcacatc
P2A-HindIII-Hyg-R	ctcgaccgagctcgccgtgagctccggcataagcttagggccagg
CarB-P2A-HindIII-R	cataagcttagggccagggttctcttcgacgtcgccagcctgcttgaggagcgagaagttggtag cgcccagaccgtgatggatgatgggtggtgatcg
GGPPS-dB-CarRP-F	ggacgtcctctccgtcaacgaa
GGPPS-dB-CarRP-R	acaagtggaactcgaggtaggtcaaaatcgacatggatccagggccagggttctcttc
CarB-dS-CarRP-F	cagaactcgaacgacatccgcac
CarRP-dS-CarB-F	gatcaaatcgttctctcgtcgac
CarRP-dS-CarB-R	caatcaccacgatgtgcttctctgatccgacatggatccagggccagggttctcttc
CarRP1373-F	agatgggcttggtcctcc
RP-CarB-pYX-R	taagcttggtggccctagatccatggctagtatggatgatgggtggtgatcggtatgcgtt
RP-GGPPS-pYX-R	taagcttggtggccctagatccatggctagtatggatgatgggtggtggttcgttgacggagag
CarB1279-F	cgagcacgaggaggtcaa

Supplementary Table 3. Gradient method for carotenoid analysis

Time (min)	Buffer A%	Buffer B%
0	90	10
10	83	17
20	75	25
25	75	25
27	50	50
35	30	70
40	30	70
42	25	75
45	90	10
55	90	10

buffer A [methanol/H₂O (92:8, v/v) and 10 mM ammonium acetate], buffer B (methyl *tert*-butyl ether).

Referce

1. Zhou Y, Gao W, Rong Q, *et al.* 2012. Modular pathway engineering of diterpenoid synthases and the mevalonic acid pathway for miltiradiene production. *J Am Chem Soc* **134**: 3234-3241.