

Complete Microbial Synthesis of Crocetin and Crocins from Glycerol in *Escherichia coli*

Jun Ho Lee^{1,+}, Seong-Rae Lee^{1,+}, Sang Yup Lee², and Pyung Cheon Lee^{1,*}

Table S1. Primers and oligonucleotide used in this study

Table S2. The sequence of *E. coli*-codon optimized genes

Figure S1. Schematic description of zeaxanthin-producing ZEA-1 strain and effect of culture temperature on growth and zeaxanthin production of ZEA-1 strain

Figure S2. Phylogenetic relationships of crALDHs of *C. sativus* (Cs) and *Synechocystis* spp (Syn), inferred using the neighbor-joining method

Figure S3. Phylogenetic tree of UGTs from various organisms

Table S1. Primers and oligonucleotide used in this study

Primer for cloning	Sequence (5'-3')	Final plasmid
pTrc_CrtY_EcoRI_F	cgGAATTCaaggagatataccatgccgcggtatgatctga	pTrc99A_CrtYIB
pTrc_CrtB_Sall_R	acgcGTCGACcgctcgcgagaacgcatg	
pKK_CsCCD2_EcoRI_F	cgGAATTCatggcgaacaagaagagg	pKK_CsCCD2
pKK_CsCCD2_HindIII_R	cccAAGCTTttaggtctccgcttgatgc	
ald_Syn6803_XbaI_F	gcTCTAGAaggaggattacaaaatgaatactgctaaaactgttgt	pUCM_Syn-aldH6803
ald_Syn6803_EcoRI_R	cgGAATTCctaggagaacaactttttgatta	
aldH_7942_XbaI_F	gctCTAGAaggaggattacaaaatgactgctgctgttctcc	pUCM_Syn-aldH7942
aldH_7942_EcoRI_R	cgGAATTCctagagcttgccggaagag	
pBBR-pUC-sub-USER-3-F	agacagUctcgaggctgacggtat	pBBRM_Syn-aldH6803
pBBR-pUC-sub-USER-1-R	atgcaacUagccatcacaaacggcat	
pSTVM2-pUC-sub-USER-1-F	agttgcaUcccgaactggaagcg	
pSTVM2-pUC-sub-USER-3-R	actgtcUatgcggtgtgaaataccg	
sub-Sall_F	acgcGTCGACccgactggaagcg	pBBRM_Syn-aldH7942
aldH_7942_EcoRI_R	cgGAATTCctagagcttgccggaagag	
pSTVM2-pUC-sub-USER-1-R	atgcaacUcgtaggacag	pSTVM_CsCCD2-Syn-aldH6803(M),
pSTVM2-pUC-sub-USER-3-F	agacagUcataagtgcgg	
pSTVM2-pUC-sub-USER-1-F	agttgcaUcccgaactggaagcg	pSTVM_CsCCD2-Syn-aldH7942(M)
pSTVM2-pUC-sub-USER-3-R	actgtcUatgcggtgtgaaataccg	
ccd2_backbone_F	aatcagcUagcttctgttttggcggatga	pSTVM_CsCCD2-Syn-aldH6803(P),
ccd2_backbone_R	acgctctUaggtctccgcttgatgct	
aldH_F	aagagcgUctagaaggaggattacaaaa	pSTVM_CsCCD2-Syn-aldH7942(P)
aldH_R	agctgatUgtactgagagtgcacat	
pET_GjUGT1_EcoRI_F	cgGAATTCatggttcagcagcgtcac	pET21a(+)_GjUGT1
pET_GjUGT1_XhoI_R	ccgCTCGAGgttgctctccgcttgatag	
pET_GT1-316_BamHI_F	cgGGATCCatggtctccgagagtctag	pET21a(+)_GT1-316
pET_GT1-316_Sall_R	acgcGTCGACtgccgatgatccgactaac	
pET_NtUGT_BamHI_F	cgGGATCCatggttcaacctcatgtctct	pET21a(+)_NtUGT
pET_NtUGT_HindIII_R	cccAAGCTTacaaccttttctacttcttgc	
pET_FaGT2_BamHI_F	cgGGATCCatgggtagtgtgctt	pET21a(+)_FaGT2

pET_FaGT2_HindIII_R	cccAAGCTTtgactctactaactcgactt	
pET_StUGT_EcoRI_F	ccgGAATTCatggttcaacctcatgtgctt	pET21a(+)_StUGT
pET_StUGT_XhoI_R	cgcCTCGAGacaagattttccacttcctg	
pET_CaUGT3_BamHI_F	cgGGATCCatggccaccgaacagca	pET21a(+)_CaUGT3
pET_CaUGT3_SalI_R	acgcGTCGACtacgcacaattgcttcagc	
pET_NsUGT_BamHI_F	cgGGATCCatggacacagaacacagca	pET21a(+)_NsUGT
pET_NsUGT_SalI_R	acgcGTCGACcaacatcatgttattacttttgc	
pUCrop_NtUGT_XbaI_F	gcTCTAGAaggaggattacaaaatggttcaacctcatgtcc	pUCrop_NtUGT
pUCrop_NtUGT_XmaI_R	tcccCCCGGGttaacaaccttttctacttcttg	
Nt_RBS_CaUGT3_XmaI_F	tcccCCCGGGgaggaggacagctaaatggccaccgaacagc	pUCrop_
Nt_CaUGT3_NotI_R	aaggaaaaaGCGGCCGCttatacgacaattgcttcagc	NtUGT_CaUGT3
Primers used for genome editing	Sequence (5'-3')	Donor DNA
pfkA-UP-F	ggatgtaattatccatcagg	Donor DNA CrtE (pfkAΔ::PLac-CrtE)
pfkA-UP-R	cccgcctttccagtcgggaagactacctctgaactttgg	
pLac-CrtE-F	tttcccgcactggaaagcg	
pLac-CrtE-R	atgcggtgtgaataaccg	
pfkA-DOWN-F	tgcggtatttcacaccgcataattaccgaacatagtgtgat	
pfkA-DOWN-R	atcgacgctatcgaaaaca	
ushA-UP-F	ggcagaaaacaaaaaacagg	Donor DNA CrtE (ushAΔ::PLac-CrtE)
ushA -UP-R	cccgcctttccagtcgggaactggtttcagcgatttcag	
pLac-CrtE-F	tttcccgcactggaaagcg	
pLac-CrtE-R	atgcggtgtgaataaccg	
ushA -DOWN-F	tgcggtatttcacaccgcattggatgtgagtgttatgaac	
ushA -DOWN-R	cactggggcagaaaaaacg	
atpI-UP-F	ttgtgtgtaaatgcatatta	Donor DNA CrtYIB (atpIΔ::PTrc-CrtYIB)
atpI-UP-R	tagccaaaagggtgaataaat	
trc_YIB_F	atttattcaccttttggtagtcacaccgaaacgcgc	
trc_YIB_R	taaggcttagagtcaagcattgtagaacgcaaaaaggcc	
atpI-DOWN-F	atgcttgacttaagcct	
atpI-DOWN-R	gcaaatatcataaacaacg	
ldhA-UP-F	gataacggagatcggggaatg	Donor DNA
ldhA-UP-R	tttgtctataaacggcgagt	CrtYZ

trc_CrtY_OV_F	ctcgccgtttatagcacaattagcgcggaattgatctggt	(ldhAΔ::PTrc-CrtYZ)
trc_CrtY_OV_R	tcattgcatcgccctgttga	
CrtZ_OV_F	tcaacaggcgatgcaatgaaaggagatataccatgttggtgattggaatgc	
CrtZ_OV_R	aggtttcgcctttttccagattactcccgggtggcg	
ldhA-DOWN-F	tctggaaaaaggcgaaacct	
ldhA-DOWN-R	cttggctgtcagttcacca	

Primer for quantitative analysis of gene transcription levels	Sequence (5'-3')
CysG_RT_F	atcaggccgcgactattcag
CysG_RT_R	gttcagtttatcgcgaggc
CsCCD2_RT_F	gcgttgacttccgcgtatc
CsCCD2_RT_R	gaagatcgcttcgctaccgt
AldH7942_RT_F	gtctgcctcaacgacacgat
AldH7942_RT_R	gagcttgcggaagagggtga

* Underlined sequences: sequences of an enzyme digestion site *Capital letter 'U' indicate uracil base

Table S2. The sequence of *E. coli*-codon optimized genes

Gene	Sequence
<i>CsCCD2</i>	ATGGCGAACAAAGAAGAGGCGGAAAAACGCAAGAAAAAACCGAAGCCGCTGA AGGTGCTGATTACCAAAGTTGACCCGAAACCGCGCAAGGGTATGGCGAGCGTTG CGGTGGATCTGCTGGAAAAAGCGTTCGTTTATCTGCTGAGCGGCAACAGCGCGG CGGATCGTAGCAGCAGCAGCGGTCGTCGTCGTCGTAAGAGCACTACTATCTGA GCGGTAACACGCGCCGGTTGGTCACGAAACCCCGCCGAGCGATCACCTGCCGA TCCACGGTAGCCTGCCGGAGTGCCTGAACGGTGTGTTTCTGCGTGTGGGTCCGA ACCCGAAGTTCGCGCCGGTGGCGGGTTACAACCTGGGTTGACGGTGACGGCATGA TTCACGGTCTGCGTATCAAAGACGGTAAAGCGACCTATCTGAGCCGTTACATCA AGACCAGCCGTTTCAAGCAAGAGGAATACTTCGGTCGTGCGAAATTCATGAAGA TTGGCGATCTGCGTGGTCTGCTGGGCTTCTTTACCATTCTGATCCTGGTGCTGCG TACCACCCTGAAGGTGATCGATATTAGCTATGGTCGTGGCACC GGTAACACCGC GCTGGTGTATCACAACGGTCTGCTGCTGGCGCTGAGCGAGGAAGACAAGCCGTA TGTTGTGAAAGTTCTGGAGGATGGTGACCTGCAAACCCCTGGGTATCCTGGATTA CGACAAGAACTGAGCCACCCGTTACCCGCGCACCCGAAGATTGACCCGCTGAC CGACGAAATGTTTACCTTCGGCTATAGCATTAGCCCGCCGATCTGACCTATCGT GTGATTAGCAAAGACGGCGTTATGCAGGACCCGGTTCAGATCAGCATTACCAGC CCGACCATTATGCACGATTTTCGCGATTACCGAAAACCTATGCGATCTTCATGGATC TGCCGCTGTACTTTACGCCGGAAGAGATGGTGAAGGGTAAATTCGTGAGCAGCT TCCACCCGACCAAACGTGCGCGTATTGGTGTCTGCCGCGTTACGCGAAAGATG AACACCCGATTCGTTGGTTTGACCTGCCGAGCTGCTTCATGACCCACAACGCGA ACGCGTGGGAGGAAAACGACGAGGTTGTGCTGTTACCTGCCGTCTGGAGAGCC CGGATCTGGACATGCTGAGCGGCCCGCGGAAGAAGAGATTGGCAACAGCAAG AGCGAGCTGTATGAAATGCGTTTCAACCTGAAGACCGGTATCACCAGCCAGAAG CAGCTGAGCGTTCGAGCGTTGACTTTCCGCGTATCAACCAGAGCTACACCGGT CGTAAGCAACAGTATGTGTACTGCACCCTGGGTAACACCAAGATTAAAGGCATC GTGAAGTTCGATCTGCAAATTGAGCCGGAAGCGGGTAAAACCATGCTGGAAGTT GGTGGCAACGTTCAAGGCATTTTTGAACTGGGTCCGCGTCGTTACGGTAGCGAA GCGATCTTCGTTCCGTGCCAACCGGGTATTAAGCGATGAAGACGATGGTTAC CTGATTTTCTTGTGCACGACGAAAACAACGGTAAAAGCGAAGTTAACGTGATT GATGCGAAAACCATGAGCGCGGAGCCGGTTGCGGTGGTTGAGCTGCCGAGCCG TGTGCCGTATGGTTTCCACGCGCTGTTCTCTGAATGAAGAAGAGCTGCAAAAGCA TCAAGCGGAGACCTAA
<i>NtUGT</i>	ATGGTTCAACCTCATGTCTACTTGTACATTTCCAGCACAAGGACATATTAACC CCTGCTTGCAAGTTTGCCATGAGGCTGATCCGTATGGGAATCGAAGTAACGTTTG CTACCTCAGTGTTTGCTCACCGTAGAATGGCTAAGACCGCAACAAGCACCTGC CCAAGGGCCTAAACTTTGCCGCGTTCAGCGACGGATACGACGATGGGTTTAAGG CAGACGAGCATGATTCCAGCAGTACATGTCTGAAATCAAGTCTAGGGGGAGCG AGACGCTGAAGGATATAATTCTGAAGAGTTCTGATGAAGGGAGACCCGTCACGT CCCTTGTTGATAGCCTGCTTTTACCCTGGGCTGCAAATGTTGCGAGGGAATTTCA CATACCCTGTGCGTTGCTATGGATACAACCGGCAACTGTTTTGGATATTTATTAC TACTACTTTAATGGGAGTGAGGACGCCATAAAAGGATCTACCAACGATCCGAAT TGGTGATCCAGTTGCCAAATCTACCTCTATTGAAATCTCAAGACTTACCTTCCT TCCTACTGTCCTCAAATAACGATGAGAAATACAGTTTTTGCTTTGCCAACGTTCAA AGAACAGCTGGACACATTGGATGTGGAAGAAAACCCCAAGTTTTGGTCAATAC ATTCGATGCGCTAGAGCAGGAAGAGCTGAAGGCGATTGAGAGATATAATTTAAT CGGGATCGGACCTTTGATACCGAGCAGTTTCTTAGATGGAAAGGACCCGCTGGA TTCAAGTTTTGGTGGCGACTTATTTTCAAGAGTCCAACGATTATATTGAATGGCTA AACTCCAAAGATAACAGTAGCGTAATATATATCTCATTCTGGTTCACTGTTGAAC CTATCCAAAACAGAAAGGAGGAAATAGCGAAAGGGCTAATCGAGATAAAAAG GCCATTCTGTGGGTCATCAGAGACCAGGAAAATGGCAAAGGCGATGAAAAAG AAGAAGAAAAGTTGAGTTGTATGATGGAAGTAGAGAAGCAAGGTAAAATCGTA CCATGGTGCTCACAATAAGTCTTGACTACCCCTCCCTTGGTTGCTTCGTCT CTACTGCGGTTGGAATTCAACACTTGAGAGTTTGTCTACAGGCGTGCCAGTGG TTGCGTTCCCTCATTGGACCGATCAGGGAACCTAACGCGAAACTTATCGAAGACG

	TATGGAAGATCGGTGTGCGTTTGAAGAAAAATGAAGATGGAGTAGTTGAGAGC GAGGAGATAAAAAGGTGTATAGACATGGTTATGAACGGAGGTGAAAAGGGAGA AGAGATGCGTCGTAAACGCACAGAAGTGGAAGAAGTAGCTCGTGAAGCCGTTA AAGAGGGAGGAAGTTCTTATATGAACCTTAAAAGCCTTCGTGCAAGAAGTAGGA AAAGGTTGTAA
<i>StUGT</i>	ATGGTTCAACCTCATGTGCTTCTTGTAAACGTTCCCGGCACAAGGTCACATCAACC CCTCACTTCAGTTTGCCAAGAGATTAATTAAGATGGGCATCGAGGTTACCTTTAC TACAAGCGTCTTCGCCCACAGACGTATGGCGAAGACGGCTGCTTCCAACGCTCC CAAGGGCCTTAATCTGGCCGCTTCTCAGATGGTTTTGATGATGGCTTCAAATCT AATGTGCGATGATTCAAAGCGTTATATGTCTGAGATTAGATCTAGGGGCAGCCAG ACCCTGCGTGACATTATCTTAAAGAGTAGCGACGAAGGAAGGCCCGTAACAAGT CTAGTATATACCCTACTATTGCCTTGGGCTGCTGAAGTAGCAAGGGAGCTGCAT ATTCCTTCAGCTTTGTTATGGATTACGCCAGCGACAGTCCTAGACATATATTATT ATTACTTCAATGGCTACGAAGATGAGATGAAATGTAGCTCAAACGATCCTAATT GGTCAATACAATTACCTCGTTTGCCCTTACTTAAGTCCCAAGATTTACCCTCTTT CCTTGTGTCCTCTAGCTCCAAGGATGATAAATACAGCTTTGCTCTACCAACGTTT AAAGAACAACCTTGACACTCTTGACGGAGAGGAGAATCCCAAGGTCTTAGTGAAT ACGTTTGACGCCCTAGAATTAGAGCCGTTAAAAGCAATCGAGAAGTATAATCTT ATCGGGATTGGACCTCTAATTCCGTCTTCATTCTGGGCGGGAAGATTCACTAG AAAGTAGCTTCGGAGGCGATTTATTTAGAAAAGCGACGATGATTACATGGAAT GGCTAAACACTAAACCCAAATCTTCAATTGTGTACATTTCTTCGTTCACTACT TAACCTATCCAGAAATCAAAAAGAGGAAATCGCAAAAGGGTTAATTGAAATCA AGCGTCCTTTTCTGTGGGTCAATTAGGGATCAGGAAAATATAAAGGAGGTGGAGA AGGAGGAAGAGAAGTTAAGCTGCATGATGGAGCTGGAAAAACAAGGTAAAATT GTTCTTGGTGTAGCCAACTAGAGGTATTAACATCATCCCTCCCTGGGATGCTTCG TAAGCCATTGTGGATGGAACAGTACACTTGAAAGTTTATCTTCAGGCGTGCCCG TGGTCGCCTTCCCTCACTGGACAGACCAAGGTACTAACGCGAAATTAATAGAAG ATGTTTGAAGACAGGTGTACGTATGCGTGTTAGCGAGGATGGCGTCGTGCAAT CAGAGGAGATTAAGAGATGTATAGAGATAGTATGATGGATGGGGGCGAGAAAGGC GAGGAAATGAGAAAAAACGCTCAGAAATGGAAAGAACTAGCCAGGGAAGCGG TGAAGGAGGGGGGAGCAGTGAGGTCAACCTAAAGGCTTTTGTGCAGGAAGTG GGAAAATCTTGTTAA
<i>FaGT2</i>	ATGGGTAGTGAGTCTTTGGTTCACGTCTTTCTTGTGTCCTTTATTGGCCAAGGAC ACGTCAACCCGCTTCTGAGGCTGGGGAAAAGACTGGCGGCTAAAGGGCTGCTTG TAACCTTTTGCACGGCGGAGTGTGTGCGGAAAGAAATGAGGAAATCTAATGGG ATAACCGATGAGCCTAAACCAGTAGGAGACGGATTTATACGTTTGAATTCTTC AAAGATAGGTGGGCAGAAGACGAACCCATGCGTCAGGACCTGGATCTATATTTA CCCCAGTTGGAGCTAGTTGGAAAAGAAGTGATACCTGAGATGATAAAGAAGAA CGCAGAACAAAGGAGACCAGTATCATGCCTAATCAATAATCCGTTTATTCCGTG GGTCTGTGACGTCGCCGAATCTCTGGTTTACCATCCGCAATGCTGTGGGTTCAA TCTGCCGCCTGTCTAGCTGCTTACTATCACTACTACCATGGACTAGTCCCATTCC CATCTGAGTCTGACATGTTTTGTGATGTCCAAATACCCAGCATGCCATTGTAAA ATATGACGAGGTTCCAGCTTTTTGTATCCAACGAGCCCATATCCATTTTTGAGA CGTGCGATCTTGGGTCAATACGGGAACCTAGAGAAGCCGTTTTGTATCCTTATG GATACGTTTCAAGAAGCTAGAGTCTGAAATAATTGAATATATGGCCAGATTATGT CCCATAAAGGCCGTCGGGCCATTGTTCAAGAATCCCAAGGCTCAAAACGCGGTT AGAGGCGACTTTATGGAAGCCGATGACTCAATCATCGGATGGTTGGACACAAAA CCCAAGAGTTCAGTTGTTTACATTAGCTTCGGGAGCGTTGTATACTTAAAGCAG GAGCAAGTGGATGAAATTGCTCACGGACTTTTATCATCCGGCGTGCTTTTATAT GGGTGATGAAACCTCCTCATCCGATAGCGGATTTGAATTGTTGGTATTACCAG AAGGTTTCCTAGAAAAGGCGGGAGACCGTGGAAGGTTGTGCAATGGAGTCCA CAAGAAAAGATATTAGAACACCCTTCAACTGCATGTTTTGTGACACACTGCGGG TGGAATAGTACGATGGAAAGTTTAAACCAGCGGGATGCCCGTGGTCGCGTTTCCA CAGTGGGGGGATCAGGTAAGTACGCGAAGTACCTAGTGGATGAATTTAAGGT AGGAGTTAGGATGTGCAGAGGCGAGGCCGAGGACAGGGTGATTCTAGAGACG AGGTTGAAAAGTGTCTTTTGAAGCTACGTCTGGTTCAAAAGCTGCAGAGATGA AACAAAATGCGCTTAAGTGGAAAGCTGCTGCAGAGGCTGCGTTCTCCGAAGGA GGATCTTCAGATAGGAATCTGCAAGCCTTCGTGACGAAGTCAGACGTATTAGC GCCAGCTTAAATCTAAGTCCTCTGCTGTCGGCTATGTCAAGAGTAAGATCAAC

	GGAGTGGTGAATATGTCGATAGTAAGTTAAACGGGAAAGCCGCCCGGTGGA GGAGGCGAACACTCGTACAAATGGCATCGCAAAGGTAGAACAACCTAAGGCAG CAAACGGCAAGGTGGAATAGCTGAATTAACCTCCGATTAATGGCAAGGTTGAG ATTGCCGAAGTGAAGCCAATAAACGGGAAAGTCGAGTTAGTAGAGTCATAA
<i>GTI-316</i>	ATGGTCTCCGAGAGTCTAGGTACCTATTCTAGTATCTTTTCCGGGGCAGGGGC ATGTAAACCCCTTTATTAAGATTAGGAAAAATCCTTGCTAGTAAAGGTTTCCTAGT TACCTTTTCCACCACCGAGACAACAGGGGAGCAGATGCGTAAGGCAAGTGACAT CATAGATAAACTTACACCATTCCGGTGACGGCTTCATTTCGTTTTGAATTCATTGCC GACGGTTGGGAGGAGGATGAACCCCGTAGGCAAGACCTTGATCAATATCTACTA CAACTTGAATTAGTGGGCAAGCAGGTAATACCGCAAATGATTAAGAAGAATGC CGAACAGGGTAGACCCGTGTCCTGCCTGATCAACAACCCCTTCATCCCATGGGT CACAGATGTCGCTACGACTCTAGGCCCTTCCTCAGCTATGCTGTGGGTACAAAGT TGTGCTTGTGTTTGCAGCTACTATCACTACTACCATGGCACAGTCCCTTTTCCCG ATGAGGAGCACCCCGAAATTGATGTACAGCTACCGTGCGATGCCTTTGCTTAAAGT ATGATGAGGTCCCTAGTTATCTTTACCCTACGACGCCGTACCCCTTTTAAAGCG TGCAATATTGGGTCAGTATAAAAACCTGGACAAGCCGTTTTGCATCTTAATGGA GACTTTTGAAGAGCTTGAACCGGAACCTTATCAAGCATATGAGTGAAATTTTCC AATTAAGGCGGTTGGACCGCTATTCCGTAACACAAAAGCGCCCAAAACAACCTGT GCACGGCGACTTCTTAAAGCAGATGATTGTATAGAATGGTTGGATACCAAAACC TCCATCTTCTGTCGTTTACGTATCATTCGGCTCTGTTGTGCAGCTAAAAACAGGAC CAATGGAACGAGATAGCTTACGGTTTACTTAACTCTGGGGTTTCCTTTTTATTGG TGATGAAGCCCGCTCATAAAGACGCTGGGCATGACCTTCTAGTACTTCCTGACG GCTTCTTAGAGAAAGCTGGCGACAGAGGGAAGGTGGTCCAGTGGTCACCTCAA GAAAAGGTACTAGGTCACCCGAGCGTAGCCTGCTTTGTAACCTACTGTGGATGG AACAGCACGATGGAAGCTCTGACCTCTGGAATGCCCGTGGTCGCTTTTCCCCAG TGGGGTGATCAGGTTACAAATGCGAAATACTTGGTAGACATACTAAAGGTAGGA GTCAGGATGTGTAGGGGAGAGGCCGAAAATAAGCTTATTACAAGAGATGAAAT TGAAAAATGCCTATTAGAAGCTACGGTAGGACCTAAAGCTGTTGAGATGAAGCA AAACGCCATGAAATGGAAGGAAGCTGCTGAAGCCGCGGTGCGCGAAGGTGGCT CAAGTGACCAAAACATACGTTACTTTACTGATGACATCGTGAAAGCAAATGAGT CCGAGATTGCTAGAAAGTGTATCGGCTCTAACGAATTCCCGGTGTCAGTCGTTG TAAATCTAATGAGAAAGTGGATGAGTTAGTCGGATCATCCGCATAA
<i>GjUGT1</i>	ATGGTTCAGCAGCGTCACGTTTTGTTGATTACCTATCCAGCACAGGGTCACATTA ACCCAGCGTTGCAGTTCGCACAGAGATTGTTGCGTATGGGTATTCAAGTTACCCT GGCGACCAGCGGTGACGCTCTGAGCCGTATGAAAAAGAGCAGCGGTAGCACCC CGAAAGGTCTGACCTTTGCAACCTTCAGCGATGGTTACGATGACGGTTTTTCGTCC GAAAGGTGTTGACCATAACGAATATATGAGCAGCCTGGCGAAGCAAGGTAGCA ATACCTGCGTAATGTGATCAACACCAGCGCTGATCAGGGTTGCCCGGTTACCT GTTTGGTGTATACCTTGTGCTGCCATGGGCTGCTACCGTTGCACGTGAATGCCA CATTCCGAGCGCGCTGCTGTGGATCCAACCGGTTGCTGTGATGGATATCTACTAT TACTATTTCCGTGGTTATGAAGATGACGTTAAGAATAACAGCAATGACCCGACC TGGAGCATCCAGTTTCCGGGTCTGCCGAGCATGAAAGCTAAGGATCTGCCGAGC TTCATTCTGCCGAGCAGCGACAACATCTACAGCTTTGCACTGCCGACCTTCAA AAGCAACTGGAACCCCTGGATGAAGAAGAACGTCCGAAAGTTCTGGTGAATAC CTTTGACGCGCTGGAACCGCAGGCACTGAAGGCGATTGAAAGCTATAACCTGAT TGCAATTGGTCCACTGACCCCGAGCGCTTCCTGGATGGTAAAGACCCGAGCGA AACCAGCTTTAGCGGTGACCTGTTTCAGAAAAGCAAGGACTACAAGGAATGGCT GAATAGCCGTCCGGCTGGTAGCGTTGTGTATGTTAGCTTTGGTAGCCTGCTGACC CTGCCGAAACAACAGATGGAAGAAATTGCACGTGGTCTGCTGAAGAGCGGTGCG TCCGTTTCTGTGGGTTATTTCGTGCGAAAGAAAACGGTGAAGAAGAAAAGGAAG AAGATCGTCTGATTGTCATGGAAGAACTGGAAGAACAGGGTATGATTGTTCCGT GGTGTAGCCAGATCGAAGTGCTGACCCATCCGAGCCTGGGTTGCTTTGTTACCC ACTGTGGTTGGAATAGCACCCCTGGAACCCCTGGTTTGCAGGTGTGCCGTTGTGG CTTTCCCGCATTGGACCGATCAAGGTACCAATGCAAACTGATCGAAGACGTTT GGGAAACCGGTGTGCGTGTGTGCCGAACGAAGATGGTACCGTTGAAAGCGAC GAAATTAACGTTGTATCGAAACCGTTATGGATGACGGTGAAAAAGGTGTGGA ACTGAAGCGTAACGCGAAAAAGTGAAGGAAGTGGCTCGTGAAAGCAATGCAGG AAGATGGTAGCAGCGACAAAAACCTGAAAGCGTTCGTGGAGGATGCGGGCAAG GGctatCAAGCGGAGAGCAACTAA

SpUGT

ATGGGAACTCAAGTTACAGAACATGGAACATCTAATCTAAGGGTAGTCATGTTCCCTTGGCTGGCATAACGGTCACATCTCACCATTCTTTATGTCGCAAAAAAAGTGGCAGACAGGGGTTTCTGATTTACTTATGTAGTACTCCCATTAATCTTAAGTCCACAATAAAGAAAAATACCAGAAAAATACGCCGACTCCATCCATTTAATCGAGCTTCAATTCCAGAAATTGCCAGAGTTGCCTCCTCATTATCATAACACGAACGGCCTTCCACCTCATCTAAATCACACACTACAGAAGGCCTTGAAGATGTCAAAGCCCAATCTATCCAAAATACTAAAAAACCTTAAGCCAGATTTAATGATCTACGACGTACTACAAACAGTGGGCTGAGAGGGTAGCGAACGAACAGTCCATTCCGGCTGTAAGGTTATTAACCTTCGGTGCCGCAGTTTTTTCATATTTTTGCAATTTGGTTAAGAAACCCGGAGTCGAGTTTCCATTCCCCGACATTTATTTGAGAAAAATTGAGCAGGTCAAAGTGTGAAATGTTGGAGAAATCTGCCAAAGACCAAGATCCTGACGATGAAGAAAGGTTAGTAGATGAGTACAAACAAATTGCTTTAATTTGTACCAGTAGAACTATTGAAGCCAAATACATCGACTTCTATTGGAGCTGAGTAACCTAAAGGTCGTTCCAGTCGGTCTCCCGTCCAGGACTTGATCACCAATGATGCCGATGATATGGAATTAATCGACTGGCTTGGCTCAAAGGATGAAAATTCCACCGTTTTTCGTAAGTTTCGGGAGTGAGTATTTTTTAAGCAAAGAAGATATGGAAGAGGTCGCGCTAGGCCTTGAAGTGAAGCAACGTAACTTTGTATGGGTCGCTAGGTTCCCCAAGGGAGAAGAACAATACTAGAAAGATGCCTTACCGAAGGGGTTCTGGAGAGAATAGGGGAACGTGGGAGAGTCTGGATAAGTTTGCACCGCAGCTGAGGATATTAAATCACACTTCCACTGGTGGATTTATATCACATTGTGGTTGGAACAGCGTGATGGAATCTATACACTTCGGAGTTCCGATTGTGGCGATGCCAATGCATTTGGACCAGCCAATGAACGCGAGGCTTATCGTTGAGCTTGGAGTAGCGGTGGAGATTGTTAGGGACGATGATGGCAAAATTCACAGGGAAGAAATTGCGAAGACTCTTAAGGACGTGATAACAGAGCGTATTGGAGAGAATCTAAGGGCTAAGATGCGTGATATTTCAATGAACCTAAACAGTATTAACGCGAGAGGAGATGGACGCAGCTGCTCATGAACCTATACAGTTCTGTAAGATCAACACCAATTAA

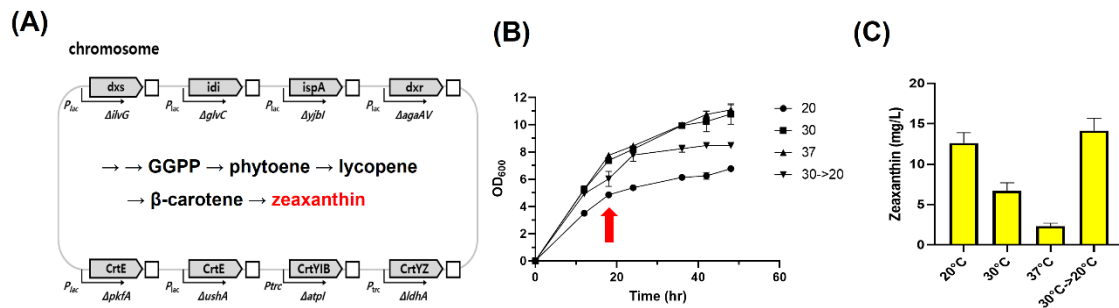
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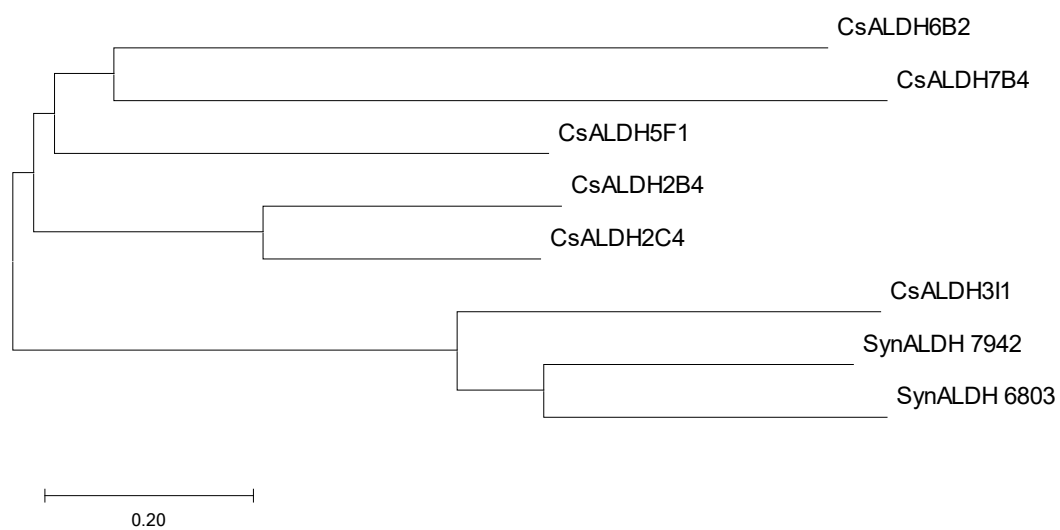
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Fig. S1



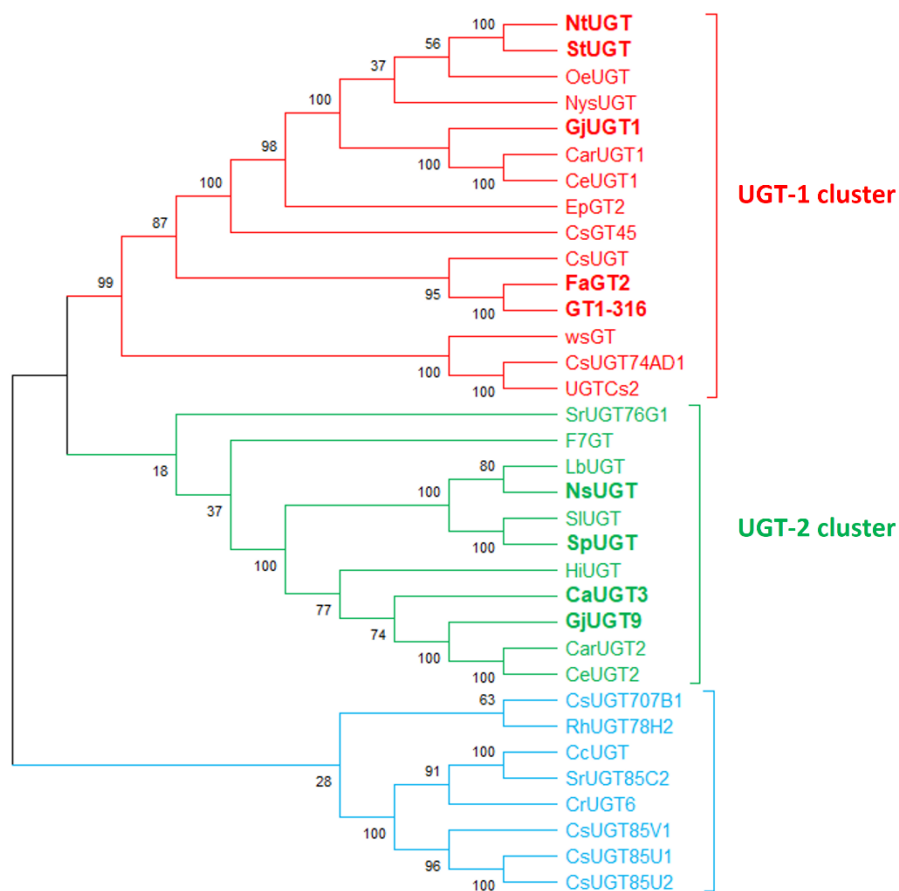
Schematic description of zeaxanthin-producing ZEA-1 strain and effect of culture temperature on growth and zeaxanthin production of ZEA-1 strain. (A) Schematic description of construction of the zeaxanthin-overproducing strain (ZEA-1) by modularly expressing five zeaxanthin biosynthetic pathway genes on the genome of the IPP- overproducing *E. coli* strain. (B) Cell growth of ZEA-1 was monitored at four culture temperatures. An arrow indicates the time of temperature shifting 30 to 20°C. (C) Quantification of zeaxanthin production in the ZEA-1 strains grown at four different temperatures. All experiments were done in biological triplicates and error bars represent mean $\pm$ SD.

Fig. S2



Phylogenetic relationships of crALDHs of *C. sativus* (Cs) and *Synechocystis* spp (Syn), inferred using the neighbor-joining method.

Fig. S3



Phylogenetic tree of UGTs from various organisms. The UGTs in red and in green were clustered as first-step UGT-1 (crocin glycosyltransferase) and second-step UGT-2 (crocin-1/2 glycosyltransferase). Five UGTs in boldface were selected and utilized for the expression and functionality study.