

Supporting Information

Engineered Production of Short-Chain Acyl-Coenzyme A Esters in *Saccharomyces cerevisiae*

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Plasmid construction

Construction of pathway plasmids is described in detail. Plasmids pNK18-22, pNK3, pNK13 are part of the pNK series. To construct plasmid pNK23 encoding the AIB pathway, expression units and the backbone were amplified from pNK3, pNK13, pNK19, pNK20 and assembled using yeast homologous recombination. Plasmid pNK26 encoding the PCC pathway, was assembled by amplifying the promoters from pYC06, the genes *accA1* and *pccB1* from gblocks and the backbone from pNK19. For the 3HP pathway (pNK30), single genes were amplified from a previously constructed plasmid pNC015³² and cloned into the pNK2.0 library, creating plasmids pNK27-pNK29. Together with the *mcrCa* gene amplified from pYC06, these expression units were then isolated by PCR and assembled into pNK30 by yeast homologous recombination. To construct pNK36, *prpE* was amplified and cloned into pNK21 using Gibson assembly. Plasmid pNK37, encoding the PrpE-PCC1 pathway, was then assembled using the *prpE* expression unit from pNK36 and the *accA1/pccB1* divergent construct from pNK26. To construct pNK42 the *ccr* gene was cloned into pNK19 by Gibson assembly, then isolated with its promoter and terminator by PCR and assembled together with the amplified pNK30 (except the *acr* expression unit) using Gibson assembly. Plasmid pNK44, encoding the BUT pathway, was constructed by isolating all required gene cassettes from plasmids pWL002 and pWL005 and combining them into the pRS425 backbone by Gibson assembly. The plasmid pNK54 represents an extended version of pNK37, with the additional subunit of the Pcc complex (encoded by *pccE*) and the biotin ligase BirA from *E. coli*. The different parts were amplified from pNK30, pNK37, pNK23, a synthetic DNA gblock and the genome of *E. coli*. A fraction of these parts was first

assembled into pNK54.1 and then extended with the remaining parts to pNK54, both times using Gibson assembly.

Table S1

Genes used in this study with their source organism and codon optimization

Gene	Sequence	Source organism	Codon optimization
<i>aibA</i>	ATGAAGACAGCTAGATGGTGCTCATTGGA AGAACGCCGTCGCCTCTATCCCTGATGGCG CATCATTGGCAACGGGTGGTTCATGTTG GGTAGAGCGCCAATGGCACTAGTAATGGA GCTGATAGCTCAGGGTAAGAGGGACTTAG GATTGATATCATTGCCAAACCCTCTTCCCG CCGAATTCTAGTAGCCGGAGGATGTCTG GCTAGACTAGAAATCGCTTCGGTGCATTA TCCTTGCAGGGTAGAGTTAGACCAATGCC ATGTCTAAAGAGAGCTATGGAACAGGGGA CTTAGCTTGGAGGGAACATGATGGTTATA GGGTGGTTCAGCGTTGAGAGCAGCTTCA ATGGGGTTGCCTTCATCCCTGCTCCTGAT GCTGACGTATCAGGTCTGGCAAGAACAGA ACCCCCCTCCTACTGTCGAAGACCCATTAC GGGATTGAGAGTAGCTGTTGAACCCGCTT	<i>M. xanthus</i>	<i>S. cerevisiae</i>

	TTTACCCGGATGTTGCGTTGCTTCACGCAA GAGCGGCAGACGAGAGAGGTAATCTATAC ATGGAAGATCCCACGACAGATTGCTGGT TGCAGGTGCTGCTAAAAGAGTTAGCCA CTGTGGAAGAACGTGTGGCCAAGCTACCA AGGGCTACTCTACCAGGTTTCAAGTTGAC AGAATAGTGTGGCACCGGGTGGAGCTT ACCGACTGGTTGTGCTGGTTATATCCACA TGACGACGAAATGTTGGCAAGATAATTGTC CCTTGCAGAGACGGGTAGGGAAGCGGAG TTCCTTGAGACACTACTGACGAGGAGAGC TGCCTAG		
<i>aibB</i>	ATGAGTGCTACTTGGACATAACTCCAGCG GAGACAGTCGTATCATTGTTAGCAAGACAA ATTGATGATGGTGGTAGTAGCTACAGG GGTTGCTCACCTTGGCAATATTAGCCAT CGCCGTTGCAAGGGCTACCCATGCACCTG ACTTAACTTACTTAGCCTGTGTGGGCTCCT TAGATCCGGAGATACCGACTTACTTCCAT CTTCCGAAGATTGGGTTACTGGATGGTC GTTCTGCTGAAATAACCATACCGATCTAT TTGATCATGCAAGAAGGGGCAGAGTTGAT ACCGTGTTTTGGAGCTGCGGAGGTCGA	<i>M. xanthus</i>	<i>S. cerevisiae</i>

	TGCCGAGGGTAGAACGAATATGACGGCCT CTGGTTCATTGGACAAACCGCGTACCAAG TTCCCAGGTGTAGCTGGTGCAGCAACTCT AAGACAATGGGTAAGGAGGCCAGTCTTAC TAGTGCCTAGACAATCCCGTCGTAACCTA GTTCCAGAAGTCCAAGTCGCTACAACTAG GGACCCTCGTAGGCCTGTAACACTGATT CTGACCTTGGGTGTTGAACTGGCGCT TCAGGGGCAAGACTACTAGCTAGACATCC TTGGGCTAGTGAAGAACATATTGCCGAAA GAACCGGTTTGCTTCCAAGTGTCTGAAG CTCTTCTGTCACCTCTTGCCTGACGCTC GTACAGTTGCGGCAATCAGAGCGATTGAC CCACATGGATAACAGAGACGCCCTAGTGGG TGCATAA		
<i>aibC</i>	ATGAAGGCCGTCGTTTACGTTCTTGGT GAGGCCGGAAACCTAAAGATGGAAACCAT GCCAATGCCACGTCCAGGTCGTGGGAA GTGTTACTACGTGTGCATGCTGTGGTGT TGTTACCATGATGTCATTAACAGAAGAGGT AATCTACCTAGGACTTCAGTGCCAGCAATT CTTGGGCATGAAGCGGCCGGAGAAGTTAT AGAAGTGGGTCCAGACACGCCCTGGGTGG	<i>M. xanthus</i>	<i>S. cerevisiae</i>

	AAAACCGGAGACCGTGCTGCGACGTTGCA GAGAATGTCTTGTGGTGATTGTGCTCTTG CAGGTCTGGGAGAAACAGTTATGTAAGA CTGATAATAGATTTTCGGTGAGGAACCTC CAGGTGGCTATGCTCAATTGTTGCTC CTGTCGGCGGATTGGGCAGGGTTCCAGC CTCATTACCATGGAATGAGGCTGCCACGG TTTGTGTACAACCGGTACCGCAGTCACA CCGTCAGGACGAGAGGAAAGGTTAGAGCC GGTGAGACCGTTTGATTACCGGGCTAG TGGAGGTGTGGTTATCATCCGTTCA TAGCCAGACTAGACGGAGCAAGGGTGATT GCTGTTACTAGTTCCGAAGCTAAAGTTCAA GCGCTTAAAGAACGCTGGTGCAGATGAGGT TATAGTCTCTAGAGGTTGGATTTGCTTC AGATGTAAGAAAGAGAACACAAGGCGCGG GCGTTGATGTTGCAGTGGAAATAGTGGGT TCTGCAACATTGATCAAACCTTAAAATCCA TGGCTCCAGGAGGTCGTGTCGTAGTAGTA GGTAACCTAGAGTCGGAATGGTCAGCT TAATCCGGGATTAGTCATAGTTAAGGAGTT GGAAATTTAGGTGCATATGCAACAACTCA AGCCGAACTGGATGAAGCCTTAAGATTGA		
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	CTGCTACTGGAGGCAGTTAGGCAATTGTC ACGGACGCGGTTCCATTGGCAGAGGCAG CCAAAGCGCATTCAGACTGGAAAATAGA GAAGTGGCTGGTAGATTGGTCTTAGTCCC CCCAGAGGCATAA		
<i>liuC</i>	ATGCCAGAATTAAAGTCGATGCCAGAGG CCCGATTGAAATCTGGACAATTGATGGTGA AAGCAGACGTAACGCAATCAGTCGTGCTA TGTAAAAGAGTTAGGCGAATTAGTAACAA GAGTTAGTTCTAGTAGAGATGTCAGGGCG GTGGTAATAACGGGTGCGGGTGATAAGGC CTTCTGCGCAGGCGCTGACTAAAAGAAA GGGCAACAATGGCCGAAGATGAAGTTAGA GCCTTCTAGACGGTCTCGTCGTACTTTT AGAGCTATTGAAAATCAGATTGCGTGTTC ATTGCAGCCATCAACGGTGCAGCGTTGGG CGGTGGTACCGAATTGCCCTGGCATGCG ATCTAAGGGTGGCTGCTCCGCTGCTGAA TTAGGATTAACGGAGGTAAAGCTGGTATT ATACCCGGCGGTGGGGCACACAGCGTT GGCGAGGCTAGTCGGCCCTGGAAGGGCA AAGGACCTAATTAAACAGCTAGAAGAATA AACGCTGCAGAAGCCTTTCTGTCGGACT	<i>M. xanthus</i>	<i>S. cerevisiae</i>

	AGCAAATAGATTAGCTCCAGAAGGTCACCT ACTTGCAGTTGCTTATGGGCTGGCAGAGT CAGTTGTTGAGAATGCCCTATAGCTGTTG CTACCGCAAAGCACCGCATTGATGAGGGT ACCGGATTAGAGTTAGACGATGCATTGGC ATTAGAGTTAAGGAAATATGAAGAAATTCT AAAGACCGAAGATAAGGCTGGAGGGTTAA GAGCGTTGCTGAAAAAAGAGCACCAAGTA TACAAAGGCAGGTAG		
accA1	ACTAGTAAAAAAATGGGTTCAAGTCACCAC CACCAACCACCACTCATCAGGTTAGTTCCA AGAGGTTCACACATGAGAAAGGTTTAATC GCCAACAGAGGTGAAATCGCTGTAAGAGT CGCTAGAGCATGCAGAGATGCCGGTATTG CTAGTGTGCA GTATATGCCGATCCAGACA GAGATGCATTGCATGTTAGAGCTGCAGAC GAAGCCTTGCTTAGGTGGTGACACTCCT GCAACATCTTATTGGACATGCCAAGGTT TTGAAGGCCGCTAGAGAATCAGGTGCAGA TGCCATCCACCCAGGTTACGGTTCTGTC CGAAAATGCCGAATTGCTCAAGCAGTTT GGACGCTGGTTAATTGGATAGGTCCAC CTCCACATGCTATAAGAGATAGAGGTGAAA	<i>S. coelicolor</i>	<i>S. cerevisiae</i>

	AAGTTGCAGCCAGACACATCGCACAAAGA GCCGGTGCTCCTTGGTAGCCGGTACACC TGACCCAGTCTCAGGTGCTGATGAAGTTG TAGCATTGCCAAAGAACATGGTTACCAA TCGCTATTAAGGCTGCATTGGTGGTGGT GGTAGAGGTTGAAGGTTGCAAGAACTTTA GAAGAAGTACCTGAATTGTATGATTCCGCT GTCAGAGAACAGTTGCCGCTTTGGTCG TGGTGAATGTTCGTAGAAAGATACTTGGA TAAACCAAGACATGTCGAAACCCAATGCTT AGCTGACACTCACGGTAACGTCGTTGTAG TCAGTACTAGAGATTGTTCTTGCAAAGAA GACATCAAAAGTTGGTTGAAGAACGCTCCT GCACCATTTTATCAGAAGCACAAACAGAA CAATTGTATTCTTCATCCAAGGCCATATTG AAGGAAGCTGGTTACGGTGGTGCAGGTAC CGTAGAATTTGGTCGGTATGGATGGTAC TATTTCTTTGGAAGTTAACACAAGATTG CAAGTTGAACATCCAGTAACAGAACAGTC GCTGGTATTGATTGGTTAGAGAAATGTTTC AGAATTGCTGACGGTGAAGAACATTAGGTTAC GATGACCCAGCATTGAGAGGTCACTCTT CGAGTTAGAATTAAATGGTGAAGATCCTGG	
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TAGAGGTTCTGCCTGCACCAAGGTACCG TTACTTATTGATGCCCTACTGGTCCAG GTGTTAGATTGGACGCTGGTGTGAATCA GGTCCGTTATTGGTCCAGCCTGGGACTC TTTGTAGCTAAATTGATAGTTACCGTAG AACTAGAGCCGAAGCATTGCAAAGAGCAG CCAGAGCTTGGATGAGTTACTGTTGAG GGTATGGCAACCGCCATTCCATTTCATAGA ACAGTTGTAAGAGACCCTGCTTCGCACC AGAATTAACAGGTTCAACCGATCCTTCAC TGTTCACACAAGATGGATCGAAACTGAATT TGTAAACGAAATTAAACCTTCACTACACC AGCTGACACAGAAACCGATGAAGAATCCG GTAGAGAACAGTCGTTGAGAAGTCGGT GGTAAAAGATTGGAAGTTAGTTACCAAGT TCTTGGCATGTCTTAGCTAGAACCGGT TTGGCTGCAGGTGCAAGACCTAAAAGAAG AGCCGCTAAAAAGTCAGGTCCAGCAGCCT CCGGTGACACCTTAGCTAGCCTATGCAA GGTACTATAGTTAAGATCGCAGTAGAAGAA GGTCAAGAAGTTCAAGAGGGTGACTTGAT TGTCGTTTAGAAGCTATGAAAATGGAACA ACCTTGAACGCACATAGATCAGGTACTAT		
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	AAAGGGTTAACCGCCGAAGTAGGTGCCT CATTAACATCTGGTGCCGCAATCTGTGAAA TCAAGGACTAAAAGCTT		
pccB1	ACTAGTAAAAAAATGGTAGTAGTCATCAC CACCACCACCACTCATCAGGTTAGTACCA AGAGGTAGTCACATGTCAGAACAGAAGA ACAACAACCAGATATCCATACTACAGCCG GTAAATTAGCTGACTTGAGAAGAAGAATTG AAGAAGCAACTCATGCCGGTAGTGCAAGA GCCGTTAAAAGCAACACGCTAAGGGTAA ATTGACAGCAAGAGAAAGAATTGATTGTT ATTGGACGAAGGTAGTTTGTGAATTGGA TGAATTGCCAGACACAGATCAACTAATT TGGTTGGATGCAAACAGACCACGGTG ACGGTGTGTAACCGGTTACGGTACTGTT GATGGTAGACCTGTTGCTGTATTTCTCAA GACTTACCGTTTGGTGGTGCATTAGGT GAAGTATATGGTCAAAAGATTGTCAAGGTT ATGGATTCGCTTGAAAACAGGTTGTCCA GTCGTTGGTATAATGATTCAAGGTGGTGCA AGAATCCAAGAAGGTGTTGCATCCTTAGGT GCCTATGGTGAATTTTAGAAGAAACACA CATGCTTCAGGTGTAATCCCACAAATTCC	S. coelicolor	S. cerevisiae

	TTGGTAGTCGGCCTTGCCTGGTGGTGC AGTCTACTCACCAAGCTATAACAGATTTAC CGTAATGGTCGACCAAACCTCCCACATGTT CATTACAGGTCCGTGATGTTATTAAAACCGT CACTGGTGAAGACGTTGGTTTGAAAGAATT GGGTGGTGCTAGAACACACAATTCTACCT CAGGTGTAGCTCATCACATGGCAGGTGAC GAAAAAGACGCAGTAGAATATGTCAAGCA ATTGTTGTCCTACTTACCAAGTAACAACCT GTCTGAACCACCTGCTTCCCTGAAGAAG CTGATTGGCTGTCACCGATGAAGACGCA GAATTGGATACTATAGTCCAGACAGTGCC AACCAACCTTACGATATGCATTCTGTTATC GAACACGTATTAGATGACGCTGAATTTC GAAACACAACCATTGTTCGCACCTAACATC TTGACCGGTTCGGTAGAGTTGAAGGTAG ACCAGTTGGTATCGTAGCCAACCAACCTAT GCAATTGCTGGTTGGATATTACTGC ATCAGAAAAAGCTGCAAGATTGTTAGAAC TTGCGATGCTTCAATGTTCCAGTATTAAC ATTGTCGATGTTCCAGGTTCTGCCTGG TGTTGATCAAGAACATGACGGTATAATCAG AAGAGGTGCTAAGTTAATATTGCTTATGC	
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	AGAAGCCACTGTCCCATTGATAACTGTTAT CACAAAGAAAAGCCTCGGTGGTGCTTACG ATGTTATGGGTTCCAAGCATTGGGTGCA GACTTAAATTGGCCTGGCCTACAGCACAA ATTGCCGTAATGGGTGCTCAAGGTGCAGT CAACATCTTACACAGAAGAACCATTGCTGA TGCAGGTGACGACGCCGAAGCTACTAGAG CCAGATTGATACAAGAATACGAAGATGCTT TGTTGAACCCATACACAGCCGCTGAAAGA GGTTACGTGATGCTGTTATTATGCCTTCA GACACCAGAAGACATATAGTTAGAGGTTG AGACAATTAAGAACTAAGAGAGAACCTTG CCACCTAAGAACACCGTAACATCCCTTA TGAAAGCTT		
<i>pccE</i>	ATGACTATCAAAGTAGTGAGAGGAAATCC GACGCCCGAAGAGAGCTGGCAGCCGCACTA ACAGTAGTCAGAGCGAGGGCGGTCACTGC CGCAGCCGAACCCCTCTACAACGGATCGTC CCCATGACGCTTGGTCAGACCCAAGCAGA ATAGCCACGCACCACATGCCTCATCCAGG TCCGACGGCCTGGGGAGGACATATTGG CCAACATGA	<i>S. coelicolor</i>	<i>S. cerevisiae</i>

<i>prpE</i>	ATGTCTTCTCTGAATTTACCAAAGAAGTA TTAATGAACCTGAAGCTTTGGGCAGAAC AAGCCAGAAGGATTGACTGGAGACAACCA TTTACGCAGACTCTTGACCCTCAAGGCC CCCGTTGCCAGATGGTTGTGGAGGCA CTACAAACCTGTGCCATAATGCTGTGGACA GATGGAGGGACAAACAACCCGAAGCCCTG GCGCTTATAGCAGTATCAAGTGAAACTGAT GAAGAGAGGACCTTACTTCTCACAACTG CATGATGAAGTTAATATCGTAGCAGCCATG TTGTTGAGTTGGGTGTCCAAAGAGGTGAT AGGGTTTAGTATATGCCTATGATGCC GAAGCCCAGATTACATTACTGGCTTGCT AGGATAGGAGCCATTCTTCATTCTGTGGCTTC GGAGGCTCGCGTCCCATAAGTGTGGCTGC TCGTATTGACGATGCAAGACCTGCTTAAT CGTGTCCGCTGATGCTGGTGCAAGGGGC GGCAAAATTCTGCCTTACAAAAAGCTGTTA GATGATGCCATCGCTCAAGCTAACACCA GCCTAAACATGTTCTGTTAGTCGATCGTGG TCTGGCAAAAATGGCTGGTAGATGGAA GAGATTAGACTCGCCACTCTGCGTCAG CAGCACCTGGCGCCAGTGTACCGGTTGC	S. <i>typhimurium</i>	<i>S. cerevisiae</i>
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	ATGGTTAGAAAGCAACGAGACTAGTTGCAT ATTGTATACATCAGGCACCACGGGGAAAC CTAAGGGTGTCAAAGAGAGACGTAGGTGGT TATGCTGTTGCCTTAGCAACGAGCATGGA CACTATCTCGGTGGTAAAGCAGGGGGTG TGTTTTTTGTGCATCAGATATAGGTTGGG TGGTTGGTCATTCTTATATTGTATATGCGC CTTATTAGCCGGTATGGCGACAATTGTT ATGAGGGTTGCCCACTTACCCGATTGC GGTGTGGTGGAAAGATAGTTGAGAAATA CCAAGTTAATAGGATGTTCTCCGCCCTAC AGCGATAAGGGTATTGAAGAAATTCCCTAC CGCGCAGATTGTAATCACGATTGAGCT CATTAGAACGATTATTTAGCAGGTGAAC CATTAGACGAGCCAAGTGCATCCTGGTA ACGGAAACGTTGGGTGTGCCGTGATCGA TAATTATTGGCAAACAGAAAGTGGTTGGCC TATTATGGCTTGCAGGGCTCTGGATGA TAGACCATCTAGATTAGGTAGTCCGGGAG TCCCGATGTACGGTTACAATGTTCAATTGT TGAATGAAGTCACAGGTGAGCCATGTGGC ATAAACGAGAAGGGTATGTTGGTGATAGA AGGCCCTTCCACCTGGATGTATCCAGA		
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	CGATATGGGGGGATGACGCAAGATTGTT AAAACCTTACTGGTCCCTTTAATCGTCAA GTTTATGCAACCTTGACTGGGTATTAGG GATGCGGAAGGTTATTATTCATACTGGGA AGAACTGATGACGTAATCACACATTGCTGGA CATAGACTTGGTACGCGTGAGATAGAGGA AAGTATCTCAAGCTACCCTAACGTTGCTGA GGTAGCTGTGGTGGGATAAAAGATGCC TAAAGGGTCAAGTAGCAGTTGCATTGTAA TCCCGAAACAGTCAGATACTTGGCCGAC AGAGAAGCCGCTAGAGACGAGGAGAATGC TATCATGGCATTGGTGGACAACCAAATCG GTCATTTGGCAGACCGGCCATGTTGG TTTGTCAGCCAATTACCAAAGACGAGGTCT GGTAAAATGTTGAGAAGAACTATACAAGCA ATTTGTGAAGGTCGTGATCCAGGGATCT TACGACCATAGATGATCCTGCTTCCTGCA GCAAATTAGACAAGCAATAGAAGAATGA		
<i>birA</i>	ATGAAGGATAACACCGTGCCACTGAAATT GATTGCCCTGTTAGCGAACGGTGAATTCA CTCTGGCGAGCAGTTGGGTGAAACGCTGG GAATGAGCCGGCGGCTATTAATAAACAC ATTCAAGACACTGCGTGACTGGGGCGTTGA	<i>E. coli</i>	-

	TGTCTTACCGTTCCGGGTAAAGGATACAG CCTGCCTGAGCCTATCCAGTTACTTAATGC TAAACAGATATTGGGTCAAGCTGGATGGCG GTAGTGTAGCCGTGCTGCCAGTGATTGAC TCCACGAATCAGTACCTTCTGATCGTATC GGAGAGCTTAAATCGGGCGATGCTTGCAT TGCAGAATACCAGCAGGCTGGCCGTGGTC GCCGGGGTCGGAAATGGTTTCGCCTTT GGCGCAAACTTATTTGTCGATGTTCTGG CGTCTGGAACAAGGCCCGGCGGCGGA TTGGTTAACGTCTGGTTATCGGTATCGTGA TGGCGGAAGTATTACGCAAGCTGGTGCA GATAAAGTCGTGTTAAATGGCCTAATGAC CTCTATCTGCAGGATCGCAAGCTGGCAGG CATTCTGGTGGAGCTGACTGGCAAAACTG GCGATGCGCGCAAATAGTCATTGGAGCC GGGATCAACATGGCAATGCGCCGTGTTGA AGAGAGTGTGTTAACAGGGGTGGATCA CGCTGCAGGAAGCGGGGATCAATCTCGAT CGTAATACGTTGGCGGCCATGCTAACAG TGAATTACGTGCTGCGTTGGAACTCTCGA ACAAGAAGGATTGGCACCTTATCTGTCGC GCTGGAAAAGCTGGATAATTATTAAATC	
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	GCCCAGTGAAACTTATCATTGGTGATAAAG AAATATTGGCATTTCACGCGGAATAGACA AACAGGGGGCTTATTACTTGAGCAGGAT GGAATAATAAAACCTGGATGGGCGGTGA AATATCCCTGCGTAGTGCAGAAAATAA		
3hpcd	ATGGAGTTCGAAACGATCGAGACCAAGAA AGAAGGCAACTTATTCTGGATTACCTGAA TCGTCCAGATAAGCTGAACCGCGCTGAACG CTAAGCTGCTGGAGGAGCTGGACCGTGC GGTTTCCCAGGCAGGAAAGCGACCCGGAAA TTCGCGTGATCATCATCACCGTAAAGGC AAAGCGTTCTGCGCGGTGCCGATATCAC TCAGTTAACCAACTGACCCCGGCTGAAG CATGGAAGTTAGCAAGAAGGGTCGCGAA ATCATGGACAAAATCGAGGCGCTGTCTAA GCCGACCATTGCGATGATTAATGGTTATGC CCTGGTGGTGGCCTGGAGCTGGCGTTG GCTTGTGATATCCGTATTGCAGCGGAGGA AGCACAATTGGGTCTGCCGGAGATTAACCT TGGGCATCTACCCGGGCTATGGTGGTACG CAACGCCTGACGCGCGTGATTGGTAAAGG CCGTGCGCTGGAAATGATGATGACCGGCG ACCGTATTCCGGGTAAAGATGCCGAGAAA	<i>S. tokodaii</i>	<i>E. coli</i>

	TACGGTCTGGTGAATCGTAGTTCCGCT GGCGAATTGGAGCAGGAAACCCGTAAAC TGGCAGAGAAGATTGCCAAGAAGAGCCG ATTAGCCTGGCACTGATCAAAGAGGTTGT CAATCGCGGTCTGGATAGCCCGCTGCTGA GC GCCCTGGCCTTGGAGAGCGTGGGCTG GGGTGTCGTTTTAGCACGGAGGACAAGA AGGAAGGTGTCTCCGCATTCTGGAGAAA CGTGAGCCTACCTCAAAGGCAAATAA		
3 ^h pcs	ATGACCGAGAAACTGAGCGAGCAACTGCA ACAAC TGGCGAACAAAACCTGGAGGAAA AAGCCGACTATAACATGCGCTATTACAAGT ACCTGTACAAGAAGAGCATCGAAGAACCG GATAAGTTCTGGGCGAGCTGGCGGAGG AGCTGATTACCTGGTATGAGCCTT GGAAG CAGGC GTT CGTGCAAGAGGAAGGCCTGCT GACTAAATGGTCGTTGGCGGTAAACTGA ACGCGAGCTATAATGCCGTGGATCGTCAC CTGAACAGCCATCGTAAATACAAGGCGGC GATCTTTGGAAAGCGAGAAAGGTGAAA AGAAAGTCGTACCTACCAGGACCTGTTT ACGAAGTGAATAAGTGGCAAATGCGCTG CGTGAGCTGGCGTGAAAAAGGGTGACC	<i>M. sedula</i>	<i>E. coli</i>

	GTTGACTATTCACATGCCGCTGACCCCG GAGGGTGTGATTGCGAAACTGGCCGTCGC CCGCTGGGTGCGATCCACTCCGTGGTT TTGCTGGTTCGGTGCGCAGGCACTGGCC GATCGTATTGCAGATGCAGGTGCGAAAGT GGTTATCACCGCGGATGCGTATTATGCC GTGGCAAATTGGTTGAAC TGAAAAAGACT GTTGACGAAGCGCTGAATATCTTGGGTGA CAAAAGCCCGTGCAAAAGTTTGGTGT ACAAACGTACCGGCACGGAGATTCCATT AAAGAGGGCCGTGACGTGTACTTGACGA GGTTGGTAAGTACAAATACATCGAGCCGG TGCCGGTTGAAGCCACCGAGCCGCTGTT ATTCTGTACACGAGCGGTACGACGGTAA GCCAAAGGGTATCGTCCACAGCACGGGC GGTTATCTGGTGGGTACGGCGGTAAATGCT GCTGTGGAGCTATGGCTTGTGCAAGAGA ACGACGTTCTGTTAACACCTCCGACATTG GCTGGATTGTCGGCCATAGCTATATCACG TACAGCCCGCTGGTCATGGTCGCTCTAT CGTATTGAAAGCGCACCGGACTACC CGTATCCGGACAAATGGGCAGAGATGATT GAGAAGTATCGTGCAACCACCTCGGCAC	
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CAGCGCTACCGCGATCCGTACCTTGATGA AGTACGGTGAGGATTATGTCAAGCAGCAC GATCTGAGCAGCCTGCGCATTATCGTTAC CAACGGCGAACCGCTGAATTATGCGCCGT GGAAGTGGGTCTGGAAGTCGTTGGTGGT GGTAAGGTGTTATGTCTCATCAGTGGTG GCAGACGGAAACCGGTGGTCCAAATATCG GCTACATTCCGGTGTGTGTATTGCCTA TGAAATCCGGTCCGGCAGTGGGTTCGCG CTGCCGGTAATAAGGTTACGGTTGTTAAT GAGGAAGGTAAAGAAACCAAACCGCGCGA GCGCGGTTACCTGGTCATGCTGCCGCCGT TTCCGCCGATGATGATGATCGGCATGTGG AACGACCCGGACAACGAGCGCCTGAAGAA AACGTACTTAGCAAGTTCCCAGGCATCTA CTACCCGGGTGATTACGCAATGATTGATG AGGATGGCTACATTGGTTATGGTCGT GCAGACGAAACCATCAAGGTTGCCGCTCA CCGCATCGGTGCCGGTGAGGTCGAGTCC ATTGTCACCAGCCATCCGGCGGTCGCTGA AGCCGCAGCTGTCGGTATCCCAGATCCGG TCAAAGGTGAAGCGGTCCACTTGTGTTG TGTTGAAGGTGGCTACAAGCCTAGCCCCG		
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	CAGCTGGCTCGCGAGATTCAAGAACATGT GCGTAAGTATATGGGCGCGATTGTTACGC CTGAGGTCCACTTCGTTGATAAACTGCCG AAAACGCGTAGCGGTAAAATCATGCGTCG TGTGATTAAAGCCGTGATGATGGGCCAAT CTGCAGGCGATATTACCACCCCTGGAAGAT GAAGCCAGCATGGACGAGATCAAGAAGGC TGTCAAGAGTTCAAGAAATCCCTGTCCCA GTAA		
<i>mcr_{Ca}</i>	AAAACAATGAGTGGTACAGGTAGATTAGCA GGTAAAATAGCATTGATAACAGGTGGTGC CGGTAAACATAGGTTCCGAATTAACAAGAAG ATTTTGCGAGAAGGTGCCACCGTTATTAT CTCTGGTAGAAACAGAGCAAAGTTAACTG CCTTGGCTGAAAGAATGCAAGCAGAAGCC GGTGTCCCTGCTAAGAGAATTGATTGGAA GTTATGGATGGTTCTGACCCAGTCGCTGT AAGAGCAGGTATTGAAGCCATAGTAGCTA GACATGGTCAAATCGATATCTGGTTAACAA ACGCAGGTTCAGCTGGTGCACAAAGAAGA TTGGCTGAAATTCTTAACTGAAGCAGAA TTGGGTCCAGGTGCCGAAGAAACATTACA TGCATCCATTGCCAATTGTTGGGTATGGG	C. <i>aurantiacus</i>	<i>S. cerevisiae</i>

	TTGGCATTGATGAGAATAGCTGCACCACA CATGCCTGTTGGTAGTGCAGTTATAAACGT CTCCACCATCTTCAGTAGAGCTGAATATTA CGGTAGAATTCCCTATGTTACTCCAAAAGC CGCTTAAATGCATTGTCTCAATTAGCAGC CAGAGAATTAGGTGCTAGAGGTATTAGAG TTAACACCATATTCCAGGTCCTATCGAAT CAGATAGAATTAGAACTGTCTTCAAAGAA TGGATCAATTAAAGGGTAGACCTGAAGGT GACACAGCTCATCACTTTAAACACCATG AGATTGTGTAGAGCAAACGATCAAGGTGC CTTGGAAAGAAGATTCCATCTGTAGGTGA CGTTGCTGACGCTGCAGTCTCTTAGCCT CCGCTGAAAGTGCCGCTTGTCAAGGTGAA ACTATTGAAGTTACACATGGTATGGAATTG CCAGCCTGCTCTGAAACATCATTGTTAGCA AGAACCGATTGAGAACTATTGACGCTTCT GGTAGAACTACATTGATCTGTGCTGGTGA CCAAATTGAAGAAGTCATGGCTTGACAG GCATGTTAAGAACCTGCGGTTCTGAAGTAA TCATTGGTTTAGATCAGCAGCCGCTTAG CTCAATTGAAACAAGCAGTTAATGAATCAA GAAGATTGGCAGGTGCCGATTTACACCA	
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	CCTATAGCTTACCATTAGATCCAAGAGAT CCAGCAACCATCGATGCCGTATCGACTG GGGTGCTGGTAAAATACAGGTGGTATAC ATGCAGCCGTTATCTTACCAGCTACCTCTC ACGAACCAGCACCTGTGTCAAGAAGTA GATGACGAAAGAGTTTGAACTTCTTAGCT GATGAAATCACAGGTACCATTGTCAAGCT TCCAGATTAGCAAGATATTGGCAAAGTCAA AGATTGACTCCTGGTGCTAGAGCAAGAGG TCCAAGAGTAATCTTTGTCTAATGGTGC TGATCAAAACGGTAACGTTACGGTAGAAT TCAATCAGCTGCAATAGGTCAATTAATCAG AGTTGGAGACATGAAGCTGAATTGGATTA CCAAAGAGCATCTGCCGCTGGTGACCACG TCTTACCACCTGTATGGCCAATCAAATTG TTAGATTGCTAACAGATCCTGGAAAGGTT TAGAATTGCCCTGTGCTGGACTGCTCAAT TGTTGCATAGTCAAAGACACATCAACGAAA TCACATTGAACATACCAGCCAACATCTCCG CTACCACTGGTGCTAGATCTGCATCAGTTG GTTGGGCTGAAAGTTGATCGGTTGCATT TGGGTAAAGTCGCATTGATCACAGGTGGT TCCGCCGGTATCGGTGGTCAAATTGGTAG	
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	ATTGTTAGCATTAAGTGGTGCCAGAGTTAT GTTGGCAGCCAGAGATAGACATAAGTTAG AACAAATGCAAGCTATGATTCAATCAGAAT TGGCAGAAGTTGGTTACACTGATGTTGAA GACAGAGTCCACATAGCTCCAGGTTGCGA TGTCTCTTCAGAAGCCCAATTGGCTGACTT AGTAGAAAGAACTTGTCTGCTTCGGTAC AGTTGATTATTGATTAATAACGCAGGTAT AGCCGGTGTAGAAGAAATGGTTATAGATAT GCCTGTTGAAGGTTGGAGACATACATTGTT CGCAAATTGATCTCCAACACTACAGTTGAT GAGAAAGTTGGCTCCATTAATGAAAAAGCA AGGTTCCGGTTACATATTGAACGTTCCAG TTACTTCGGTGGTGAAAAAGATGCTGCAAT ACCATATCCTAACAGAGCTGACTACGCAGT CTCTAAGGCAGGTCAAAGAGCAATGGCCG AAGTATTGCTAGATTCTTAGGTCCCTGAAA TCCAAATTAAATGCTATTGCACCAAGGTCTG TTGAAGGTGACAGATTAAGAGGTACTGGT GAAAGACCAGGTTGTTGCCAGAAGAGC TAGATTGATCTGGAAAATAAGAGATTGAA CGAATTACATGCCGCTTGATTGCAGCCG CTAGAACAGATGAAAGATCTATGCACGAAT	
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TAGTAGAATTGTTGTCCTAATGACGTTG		
CAGCCTTGGAACAAAACCCTGCTGCACCA		
ACTGCCTTGAGAGAATTAGCTAGAAGATT		
AGATCTGAAGGTGACCCAGCCGCTTCTTC		
ATCCAGTGCATTGTTAACAGATCAATAGC		
AGCCAAGTTATTGGCTAGATTACATAACGG		
TGGTTATGTTTGCCTGCTGATATTTC		
AAATTGCCTAACCCACCTGACCCATTTT		
CACAAGAGCCCCAAATTGATAGAGAAGCTA		
GAAAGGTTAGAGACGGTATCATGGGCATG		
TTGTACTTGCAAAGAACGCCAACCGAATT		
GATGTTGCCATGGCTACTGTCTATTACTTA		
GCTGACAGAAATGTTCCGGTGAAACTTTT		
CATCCTAGTGGTGGTTGAGATATGAAAGA		
ACTCCAACAGGTGGTGAATTGTCGGTTA		
CCATCTCCTGAAAGATTGGCTGAATTAGTC		
GGTTCAACAGTATACTTAATAGGTGAACAT		
TTGACCGAACACTTAAATTGTTGGCAAGA		
GCCTATTGGAAAGATAACGGTGCAAGACA		
AGTTGTCATGATTGTTGAAACCGAAACTGG		
TGCTGAAACAATGAGAAGATTATTGCATGA		
TCACGTTGAAGCTGGTAGATTGATGACCAT		
TGTTGCTGGTGACCAAATAGAAGCTGCAAT		

	CGACCAAGCTATTACTAGATATGGTAGACC AGGTCCCTGTAGTTGACTCCTTTAGACC ATTACCTACAGTCCATTGGTCGGTAGAAA AGATTCTGACTGGTCAACAGTTTATCAGA AGCAGAATTGCCGAATTATGCGAACATCA ATTGACTCATCACTTCAGAGTCGCCAGAAA GATTGCTTGTCTGATGGTGCTTCATTAGC ATTGGTAACCCCAGAAACAACCGCTACTTC CACTACAGAACAAATTGCTTGGCAAACCTT CATCAAGACCACTTGCATGCCTCACAGC TACCATTGGTAGAAAGTGAAAGAACTGC TCAAAGAACATTAATCAACCAAGTTGATT GACAAGAACAGGCCAGAGCTGAAGAACCTA GAGACCCACACGAAAGACAACAAGAACATTA GAAAGATTCAATTGAAGCAGTATTGTTGGTT ACTGCCATTGCCACCAGAACAGACAC AAGATAACGCAGGTAGAATCCACAGAGGTA GAGCCATTACAGTCTAA		
acr	ATGAAAGCAATCGTCGCCCCAGGCCAAA ACAAGGCTACAAACTGGAGGAAGTGCCGG ACCCGAAACCGGGTAAGGATGAAGTCATT ATTCGCGTTGATCGTGCAGGCAGTGTGTTA CCCGCGACTTGCTGCAGTTGCAGGGTTACT	<i>M. sedula</i>	<i>E. coli</i>

ATCCCGTATGAAATATCCGGTCATCCTG GGCCATGAGGTCGTGGTACGATTGAAGA GGTTGGCGAGAACATTAAGGGTTCGAAG TCGGTGATAAAGTTATCAGCCTGTTGTATG CGCCGGACGGTACGTGTGAGTACTGCCAG ATTGGTGAAGAGGCCTACTGCCACCACG CCTGGCTATTCCGAGGAGCTGGACGGCT TCTTGCTGAGAAGGCGAAGATCAAGGTG ACCAGCTTGGTTAAAGTTCCGAAAGGTAC GCCGGATGAAGGCGCGGTTCTGGTGCCG TGC GTTACGGGTATGATCTATCGTGGCATT CGCCGTGCGGTGGTATCCGTAAAGGTG AGCTGGTCCTGGTGACGGGTGCGAGCGG TGGTGTAGGTATTACGCTATCCAAGTGG CAAAGCACTGGTGCGAAGGTGATTGGC GTCACCACCTCTGAAGAAAAGGCAAAGAT CATCAAACAGTACGCGGACTACGT CATCG TTGGCACCAAATT CAGCGAAGAAGCCAAG AAGATCGCGATGTGACCTTGGTTATCGA CACCGTAGGCACCCCGACCTTGACGAGA GCCTGAAAAGCCTGTGGATGGCGGT CGT ATTGTGCAGATTGGTAATGTTGATCCGAGC CAAATCTACAATCTCGGTCTGGTTACATT		
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	ATTCTGAAAGACCTGAAAATTGTTGGCCAT GCCAGCGCTACGAAGAAAGATGCCGAAGA TACCCTGAAACTGACCCAAGAGGGCAAGA TTAACCGGTTATTGCGGGTACTGTCTCC CTGGAGAACATCGACGAGGGTTATAAGAT GATCAAGGACAAAAACAAGGTGGGTAAAG TGCTGGTGAAGCCTTAA		
<i>ccrCc</i>	ATGACCATCCAGACACTTGAAACAAACAGCA TTGAAAGATTATATGAGATTGGTGAAATT CCACCTGCCTTCATGTTCCAAAAACAATG TATGCATGGAGCATCAGAAAAGAAAGACA CGGAAAACCCACACAAGCAATGCAAGTCG AAGTAGTACCAACGTGGAAATTGGCGAA GACGAAGTTCTAGTTGGTAATGGCTGCT GGTGTAAATTATAACGGTGGTGGCAGC CTTAGGAGAACCAATTCTCCATTGGATGG CCACAAACAACCATTCACATCGCAGGGT CCGATGCTAGTGGATTGTGTGGAAAGTC GGTGCAGGGTAAAAGATGGAAGCTAGG TGATGAAGTGGTTATTCACTGCAACCAAGA TGATGGTGATGACGAAGAATGTAATGGGG GAGATCCAATGTTCTTCACAGCGTA TCTGGGGTTACGAGACTCCTGATGGCTCA	C. <i>crescentus</i>	<i>S. cerevisiae</i>

	TTTGCTCAGTTCTGTAGAGTTCAATCCCGT CAGCTGCTGCCTAGACCAAAGCATCTGAC CTGGGAAGAAAGTGCCTGCTATACATTAAC CTTAGCTACCGCCTATAGAATGCTTTTGG ACATAAGCCTCATGAATTAAAACCAGGCCA GAATGTGCTTGTGGGTGCATCTGGCG GCTTGGGTGTTTGCACACTCAGCTGGCT GCAGTAGCAGGTGCAAATGCTATTGGTGT GGTTTCTTCTGAAGACAAGAGGGAGTCG TCTTGTCTATGGGTGCAAAGGCCGTATTAA ATAGAGGTGAATTCAATTGTTGGGACAG CTACCAAAGGTTAACGGCCCAGAGTTCAA TGATTACATGAAAGAAAGCAGAAAATTGG AAAAGCCATTGGCAGATCACCGGCAATA AGGATGTTGATATGGTATTGAACATCCGG GTGAACAAACATTCCCCGTGTCCGTGTTCT TAGTTAAGAGGGTGGATGGTCGTCATT TGCGCAGGTACAACAGGTTCAACCTTACT ATGGATGCCAGGTTTGTGGATGAGACA AAAACGTGTGCAAGGAAGCCATTTGCAA CTTGATGCAGGCTTCCGCTGCTAATCAACT AGTCATCGACAGGAGAGTGGATCCATGTC TAAGTGAAGTTTCCGTGGATCAGATAC	
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	CAGCTGCTCACGAAAAGATGTTAGCCAAT CAACATCTTCCAGGTAAATATGGCAGTACTG GTGTGCGCACAAAGACCAGGCCTTAGGAC ATTCGAAGAAGTTCAAGAATTGAGTGGTGC CCCATAA		
ter	CAAATCGAACATACACTAAAAAGAGAATTACC GCTGAAGTTAAAGCTGGTGCTAAGGCC AAAGAACGTTTGGTTTGGTTGGTCCAA CGGTTACGGTTGGCCTCCAGAATTACTG CTGCTTTGGTTACGGTGCCGCTACCATC GGTGTCTTTGAAAGGCCGGTCCGA AACTAAGTACGGTACTCCAGGTTGGTACAA TAACCTGGCTTCGATGAAGCTGCTAAGAG TATTCCGTTACTATTGACGGTGATGCCTT TCTGACGAAATCAAAGCTCAAGTCATCGAA GAAGCCAAAAAGAAAGGTATCAAGTTCGAT TTGATTGTCTACTCTTAGCCTCTCCTGTTA GAAC TGATCCAGATACTGGTATTATGCACA AATCCGTTTGAAGCCATTGGTAAGACCT TCACTGGTAAA ACTGTCGATCCTTCAGT GTGAATTAAAGGAAATCTCTGCTGAACCTG CCAACGACGAAGAAGCTGCTGCCACTGTT AAGGTTATGGGTGGTGAAGACTGGGAAAG	<i>T. denticola</i>	<i>S. cerevisiae</i>

	ATGGATCAAGCAATTATCTAAGGAAGGTTT GTTGGAAGAAGGTTGTATCACCTTGGCTTA CTCTTACATCGGTCCAGAACGCTACCCAAG CTTTGTACAGAAAGGGTACCATGGTAAG GCTAAAGAACACTTGGAGTCAATAAAGGTT TGGTCACTAGAGCCTCTGCCGTATTCCA GTTATCCCTTATACTTGGCTTCTTGTTA AAGTCATGAAGGAAAAGGGTAACCATGAA GGTTGTATCGAACAAATCACTCGTTGTAC GCTGAACGTTATACAGAAAGGACGGTAC CATCCCTGTCGATGAAGAAAACAGAATCA GAATCGACGATTGGGAATTGGAAGAAGAT GTTCAAAAAGCCGTTCCGCCTTGATGGAA AAGGTCACCGGTGAAAATGCCGAATCCTT GAUTGACTTAGCTGGTTACAGACATGACTT TTTAGCTTCTAATGGTTCGATGTTGAAGG TATTAACTATGAGGCTGAAGTCGAAAGATT TGACAGAATCGGATCCTAA		
crt	ATGACTAGAGAAGTTGTCGTCGTTCCGGT GTCCGTACCGCTATCGGTACTTCGGTGG TTCCTTAAAGGATGTTGCTCCTGCTGAATT GGGTGCTTAGTTGTTAGAGAAGCTTG CCAGAGCCCCAAGTCTCCGGTGACGACGTT	C. <i>acetobutylicum</i>	<i>S. cerevisiae</i>

	GGTCACGTCGTTTCGGTAACGTCAATCCAA ACTGAACCACGTGACATGTACTTGGGTAG AGTCGCCGCTGTTAACGGTGGTGTACCCA TCAACGCTCCTGCCTTAACGTAAACAGAT TATGTGGTTCCGGTTACAAGCTATTGTCT CTGCCGCCAAACTATCTTGGTGGTGATA CTGACGTTGCTATTGGTGGTGGTGCTGAA TCTATGTCTAGAGCTCCATACTTGGCTCCA GCTGCCCGTTGGGTGCTAGAATGGGTGA CGCCGGTTGGTCGATATGATGTTGGTG CCTTGCATGATCCTTCCACAGAACCCACA TGGGTGTTACCGCTGAAAACGTTGCTAAG GAATACGATATCTCTAGAGCTAACAAAGAT GAAGCCGCTTAGAATCTCACAGACGTGC CTCCGCCGCTATTAAGGCTGGTTACTCAA GGACCAAATTGTTCCAGTTGCTCTAAGGG TCGTAAAGGTGATGTTACCTTGATACTGA CGAACACGTTAGACACGACGCCACTATTG ACGATATGACTAAATTAAAGACCAGTCTTG TTAAGGAGAATGGTACCGTTACTGCTGGTA ACGCTTCTGGTTGAACGATGCCGCCGCT GCCGTTGTTATGATGGAAAGAGCTGAAGC CGAAAGACGTGGTTAAAGCCATTGGCCA	
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	GATTAGTCTCCTACGGTCACGCTGGTGT GACCCAAAGGCTATGGGTATCGGTCCAGT TCCTGCTACTAAGATTGCTTAGAAAGAGC TGGTTTGCAAGTTCTGACTTGGACGTCAT CGAAGCCAACGAAGCCTCGCTGCTCAAG CTTGTGCTGTCACCAAGGCTTGGGTTG GATCCAGCTAAAGTTAACCTAATGGTTCT GGTATTTCCTGGGTACCCAATCGGTGC TACCGGTGCTTAATCACTGTTAACGCCTT ACACGAATTGAACAGAGTTCAAGGTAGATA CGCTTGGTCACTATGTGCATCGGTGGTG GTCAAGGTATCGCTGCTATCTCGAAAGAA TCGGATCCTAA		
<i>bktB</i>	ATGGAATTGAACAACGTTATTTGGAAAAG GAAGGTAAGGTCGCTGTCGTTACTATCAA CAGACCAAAGGCTTAAACGCTTGAACTC TGACACCTTGAAAGAAATGGATTATGTTAT CGGTGAAATCGAAAATGACTCTGAAGTTTT GGCCGTTATCTGACTGGTGCCTGGTGAAGA AATCTTCGTTGCTGGTGCCTGACATTCTG AAATGAAGGAGATGAATACCATTGAAGGTA GAAAGTTCGGTATCTGGGTACAAGGTTT TTAGAAGATTGGAATTGTTGGAAAAACCAG	<i>R. eutropha</i>	<i>S. cerevisiae</i>

	TCATCGCTGCTGTTAACGGTTCGCTTAG GTGGTGGTTGTGAAATCGCTATGTCCTGT GACATT CGTATGCCCTCCCAATGCTAGA TTCGGTCAACCAGAAGTTGGTTAGGTATT ACTCCAGGTTCGGTGGTACCCAAAGATT GTCTAGATTGGTCGGTATGGGTATGGCTA AGCAATTAACTTCACTGCTCAAAACATTAA GGCTGATGAAGCCTACGTATTGGTTGGT CAACAAGGTCGTTGAACCATCTGAATTGAT GAATACCGCTAAGGAAATTGCTAACAAAAT TGTTTCTAATGCCCCAGTTGCTGTCAAGTT GTCCAAGCAAGCTATTAACAGAGGTATGC AATGTGATATTGACACTGCTTGGCTTCG AATCCGAAGCTTTGGTAATGTTTCTA CCGAAGATCAAAAGGATGCTATGACCGCT TTCATCGAGAAGAGAAAGATCGAAGGTTTC AAAAACAGAGGATCCTAA		
paaH1	ATGTCCCATCAGAACTGTCGGTATTGTTGGT GCTGGTACTATGGGTAAACGGTATTGCTCA AGCCTGTGCTGTCGTCGGTTAACGTGCG TCATGGTCGACATTCTGACGCTGCTGTT AAAAGGGTGGTGTACTGTCGCTTCCTCTT TGGACAGATTAATTAAGAAGGAAAAGTTGA	C. <i>acetobutylicum</i>	<i>S. cerevisiae</i>

	CCGAAGCCGACAAGGCCTCTGCCTGGCC AGAATTAAGGGTCCACTTCTTATGACGAC TTGAAAGCTACCGACATTGTTATCGAAGCT GCTACTGAAAAC TACGATTGAAAGTTAAG ATCTTGAAGCAAATTGATGGTATCGTCGGT GAGAACGTCATTATTGCTTCTAACACTTCC TCCATTCTATCACTAAATTAGCCGCCGTC ACCTCTAGAGCCGACAGATTATCGGTATG CACTTCTTAATCCAGTTCCAGTCATGGCT TTGGTCGAATTAATTAGAGGTTGCAAACC TCCGACACCACCCACGCCGCCGTTGAAGC TTTGTCTAAGCAATTGGGTAAGTACCCAAT CACCGTTAAAATTCCCCAGGTTCGTTGT CAACCGTATTTGTGCCCAATGATCAATGA AGCTTCTGTCTGGGTGAGGGTTGG CCTCCCCAGAAGAAATCGATGAAGGTATG AAGTTAGGTTGTAACCACCCATTGGTCCT TTAGCCTGGCCGACATGATCGGTTAGA CACTATGTTGCCGTTATGGAAGTCTTGT CACTGAATTGCTGACCCAAAGTACAGAC CAGCTATGTTAATGAGAGAAATGGTTGCTG CCGGTTATTTGGGTAGAAAGACTGGTCGT GGTGT TATGTCTACTCTAAAGGGATCTAA	
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Table S2**Plasmids used in this study to build pathway plasmids (Table 3)**

Strain/Plasmid name	Description/genotype	Reference
pNK3	P_{PGK1} - <i>aibC</i> -T _{ADH1} , LEU2	This study
pNK13	P_{TEF1} - <i>aibA</i> -T _{PGK1} , HIS3	This study
pNK18 (pNK2.0)	P_{PGK1} - <i>bkdB</i> -T _{ADH1} , 2μ, TRP1	This study
pNK19 (pNK2.0)	P_{ADH1} - <i>aibB</i> -T _{HXT7} , 2μ, TRP1	This study
pNK20 (pNK2.0)	P_{FBA1} - <i>liuC</i> -T _{TDH3} , 2μ, TRP1	This study
pNK21 (pNK2.0)	P_{TDH3} - <i>ilvE</i> -T _{ACT1} , 2μ, TRP1	This study
pNK22 (pNK2.0)	P_{TEF1} - <i>ipdV</i> -T _{PGK1} , 2μ, TRP1	This study
pNK26	P_{TEF1} - <i>accA1</i> -T _{TDH3} P _{PGK1} - <i>pccB1</i> -T _{HXT7} , 2μ, TRP1	This study
pNK27	P_{TDH3} - <i>3hpcs</i> -T _{ACT1} , 2μ, TRP1	This study
pNK28	P_{FBA1} - <i>3hpcd</i> -T _{TDH3} , 2μ, TRP1	This study
pNK29	P_{ADH1} - <i>acr</i> -T _{HXT7} , 2μ, TRP1	This study
pYC06	P_{TEF1} - <i>mcr</i> -T _{CYC1} , HIS3	Shi et al., 2014
pNK43	P_{ADH1} - <i>ccr</i> -T _{HXT7} , 2μ, TRP1	This study
pWL002	P_{GAL7} - <i>ter</i> -T _{ADH1} P _{TEF2} - <i>crt</i> -T _{PGK1} , URA3	This study

pWL005	$P_{GAL1-paaH1-T_{ENO1}}$ $P_{GAL10-bktB-T_{SSA1}}$, URA3	This study
pGT18	Integrative plasmid for LEU2 locus with HIS3 selection marker	Unpublished
pGT22	Integrative plasmid for HIS3 locus with hphMX4 selection marker	Unpublished

Table S3

Primers used in this study for pathway cloning

Primer	Sequence	Amplicon
NK1	TCCACCCGGCGATTGGCGCGAATCCTTA	P_{PGK1}
NK2	CATTGTTTATTTGTTGTAAAAAGTAGATAATTAC TT	P_{PGK1}
NK3	CTACTTTACAACAAATATAAAAACAATGAAGGCCG TCGTTTACGT	<i>aibC</i>
NK4	AAAATCATAAATCATAAGAAATTGCTTATGCCTCT GGGGGGACT	<i>aibC</i>
NK5	GCGAATTCTTATGATTATGATTATTAAATA AG	T_{ADH1}
NK6	TCATAAAAAAGAGAACATCTTCTTGAAGCTCAGGTA AGGGG	T_{ADH1}
NK7	CCCCTTACCTGAGCTTCAAGAAAGATTCTCTTTTT ATGATATTGTACA	Backbone pGT18 (Keasling Lab unpublished)
NK8	CCAAATGCCCGGGTGGATGGCGCGTTAGTAT	Backbone pGT18 (Keasling Lab unpublished)
NK9	GTGTCGACCCGGGAAGCAACAGGCGCGTTGGA	P_{TEF1}
NK10	TTTGTAATTAAAACCTAGATTAGATTGCTATGCT	P_{TEF1}
NK11	AGCATAGCAATCTAATCTAAGTTTAATTACAAAATG AAGACAGCTAGATGGTGCTCATTGGA	<i>aibA</i>

NK12	AGAAAAAAAATTGATCTATCGATTCAATTCAATTCC ACGCAGCTCTCCTCGTCAGTAGTGTCT	<i>aibA</i>
NK13	GAATTGAATTGAAATCGATAGATCAATTTTTCT	T _{PGK1}
NK14	TCTGCTAGCCTCTTTGTTGCAAGTGGGATGAGCT TGGAGC	T _{PGK1}
NK15	CACTTGCAACAAAAAGAGGGCTAGCAGAATTACCC CCACG	Backbone pGT22 (Keasling Lab unpublished)
NK16	CTGTTGCTTCCCCGGGTCGACACTGGATGGC	Backbone pGT22 (Keasling Lab unpublished)
NK17	CCGCCCGCTTAATGCGCCGCTACAGGGCGCGTC GCGCCATTGAGTACTCGATTGGCGCGAATCC	P _{PGK1}
NK18	CATTGTTTATATTGTTGAAAAAGTAGATAATTAC TT	P _{PGK1}
NK19	CAAGGAAGTAATTATCTACTTTACAACAAATATAA AACAAATGGCAATTGAACAAATGACG	<i>bkdB</i>
NK20	CTTATTTTTTATAACTTTAATAATAATAAAAAATCAT AAATCATAAGAAATTGCTTAGTAAACAGATGTCTT CTCG	<i>bkdB</i>
NK21	GCGAATTCTTATGATTATGATTATTAAATA AG	T _{ADH1}
NK22	GCAGCGAGTCAGTGAGCGAGGAAGCGGAAGAGCG CCCAATACGCAAAGTACTCTGAAGCTCAGGTAAG GGG	T _{ADH1}
NK23	AGTACTTGCATTGGCG	Backbone pRS424
NK24	AGTACTCGAATGGCGCGACG	Backbone pRS424
NK25	CCGCCCGCTTAATGCGCCGCTACAGGGCGCGTC GCGCCATTGAGTACTGGGTGTACAATATGGACTT CCTC	P _{ADH1}
NK26	TGTTTATGAGATAGTTGATTGTATGCTGG	P _{ADH1}
NK27	CAATCAACTATCTCATAAACAATGAGTGCTACTTG GACATAACTCC	<i>aibB</i>
NK28	GCGTGATCATGAATTAATAAAAGTGTTCGCAAATTA TGCACCCACTAAGCGTCTGTATCC	<i>aibB</i>
NK29	TTTGCACACTTTATTAATTGATCAC	T _{HXT7}
NK30	CAGCGAGTCAGTGAGCGAGGAAGCGGAAGAGCGC CCAATACGCAAAGTACTCTCTTTAAGTGACGGGC GATG	T _{HXT7}

NK31	CCGCCCGCGCTTAATGCGCCGCTACAGGGCGCGTC GCGCCATTGAGTACTTGGGTCAATTACGTAAATAAT GATAGGAATGGGA	P_{FBA1}
NK32	CATTTGAATATGTATTACTTGGTTATGGTT	P_{FBA1}
NK33	ACCATAACCAAGTAATACATATTCAAAATGCCAGAA TTTAAAGTCGATGCCAGAGG	<i>liuC</i>
NK34	GCAAGATTAAAGTAAATTCACCTACCTGCCTTG ATACTGGTG	<i>liuC</i>
NK35	GTGAATTACTTAAATCTTGCATT	T_{TDH3}
NK36	AAGAGCGCCCAATACGCAAAGTACTGGCGCGTACA TTAATTTCACGT	T_{TDH3}
NK37	CCGCCCGCGCTTAATGCGCCGCTACAGGGCGCGTC GCGCCATTGAGTACTACAGTTATTCCCTGGCATCC ACTAAATATAATGG	P_{TDH3}
NK38	CTTATTCACTTGTTGTTATGTGTGTTATTGAA AC	P_{TDH3}
NK39	CGAATAAACACACATAAACAAACAAAATGAATAAGC TTATTGAACGAGAAAAAACTGT	<i>ilvE</i>
NK40	CGCACAAAAGCAGAGATCACACTCCACTGTCCAG TTAACCGG	<i>ilvE</i>
NK41	GAAGTGTGATCTCTGCTTTGTGCGCG	T_{ACT1}
NK42	AAGAGCGCCCAATACGCAAAGTACTTGGTTCCCA GAAACAAAGGG	T_{ACT1}
NK43	CCGCCCGCGCTTAATGCGCCGCTACAGGGCGCGTC GCGCCATTGAGTACTGAAGCAACAGGCGCGTTGG	P_{TEF1}
NK44	GTTGCCATTGTAATTAAACTTAGATTAGATTGCT ATGCT	P_{TEF1}
NK45	GCAATCTAATCTAAGTTTAATTACAAAATGGCAACT GAGTATGACGTAGTCATTCTGGG	<i>ipdV</i>
NK46	AGAAAAAAATTGATCTATCGATTTCAATTCAATTCTT AAAAATGAATGGCTTGCCATCTGCGGC	<i>ipdV</i>
NK47	GAATTGAATTGAAATCGATAGATCAATTTC TTTC	T_{PGK1}
NK48	GCAGCGAGTCAGTGAGCGAGGAAGCGGAAGAGCG CCCAATACGCAAAGTACTTTTGTGCAAGTGGGAT GAGCTTG	T_{PGK1}
NK49	ACGTTGAAAATTAAATGTACGCGCCAGTACTTGC TATTGGCGCTTCCGCTTCC	Backbone pRS424
NK50	AAATAAAGGATTGCGGCCAAATCGTTTGTGCAA GTGGGATGAGCTTGG	T_{PGK1}

NK51	CAAGCTCATCCCAC TTGCAACAAAACGATTGGGC GCGAATCC	P_{PGK1}
NK52	AAGAGGAAGTCCATATTGTACACCCCTTGAAGCTCA GGTAAGGGGC	T_{ADH1}
NK53	ACTAGCCCCTACCTGAGCTTCAGGGGTGTACAA TATGGACTTCCTC	P_{ADH1}
NK54	CTATCATTATTACGTAATGACCCACTTCTTTAAGT GACGGGCGATG	T_{HXT7}
NK55	CTCATGCCCGTCACTAAAAGAAGTGGGTATTAC GTAAATAATGATAGGAATGGGA	P_{FBA1}
NK56	GCAGCGAGTCAGTGAGCGAGGAAGCGGAAGAGCG CCCAATACGCAAAGTACTGGCGCGTACATTAAATT TCAACGTATTCTATAAG	T_{TDH3}
NK57	GGGCGCGTCGCGCCATT CGGTATACGGCGCGTAC ATTTAATTTCAAC	T_{TDH3}
NK58	CTGTGAAATCAAGGACTAAAAGCTTGTGAATTACT TTAAATCTTGCATT	T_{TDH3}
NK59	AATGCAAGATTAAAGTAAATTACAAGCTTTAGTC CTTGATTCACAGATTGCGGC	$accA1$
NK60	ATCTAAC TCAAGTTTAATTACAAA ACTAGTAAAAAA ATGGGTTCAAGTCACCACC	$accA1$
NK61	GACTTGAACCCATT TTTTACTAGTTGTAAATTAAA ACTTAGATTAGATTGCTATGCT	P_{TEF1}
NK62	GACTACTACCCATT TTTTACTAGTTGTTTATATT GTTGTAAAAAGTAG	P_{PGK1}
NK63	TACTTTTACAACAAATATAAAACAAC TAGTAAAAAA ATGGGTAGTAGTCATCAC CACC	$pccB1$
NK64	ATGAATTAAT AAAAGTGT CGCAAAAGCTTCATAA AGGGATGTTACCG	$pccB1$
NK65	CGGT AACATCCC TTATGAAAGCTTTGCGAACAC TTTTATTAAATT CATGATCAC GCG	T_{HXT7}
NK66	AAGAGCGCCCAATACGCAAGTATA CCTTCTTTAAG TGACGGGCGATG	T_{HXT7}
NK67	CATCGCCC GTCACTAAAAGAAGGTATACTT GCGTA TTGGCGCTCTCCGCTTCC	Backbone pRS424
NK68	GAATAAACACACATAAACAAACAAAATGACCGAGAA ACTGAGCGAGCACTGC	$3hpcs$
NK69	AAACATACGCGCACAAAGCAGAGACTGGGACAGG GATTCTGA ACTCTCGAC	$3hpcs$

NK70	AGAGTTCAAGAAATCCCTGTCCCAGTCTCTGCTTT GTGCGCGTATG	T_{ACT1}
NK71	GTTGCTCGCTCAGTTCTCGGTCACTTGTTGTT ATGTGTGTTATTGAAAC	P_{TDH3}
NK72	CATAACCAAGTAATACATATTCAAAATGGAGTCGA AACGATCGAGACC	$3hpcd$
NK73	AATGCAAGATTAAAGTAAATTCACTTGCCTTGAA GGTAGGCTCACG	$3hpcd$
NK74	ACGTGAGCCTACCTCAAAGGCAAAGTGAATTACT TTAAATCTTGC	P_{FAB1}
NK75	TGGTCTCGATCGTTCGAACTCCATTGAAATATGT ATTACTGGTTATG	P_{FAB1}
NK76	ATACAATCAACTATCTCATAAACAAATGAAAGCAAT CGTCGCCCCAG	acr
NK77	ATGAATTAATAAAAGTGTTCGCAAAAGGCTTCACCA GCACTTACCC	acr
NK78	GGTGGGTAAAGTGCTGGTGAAGCCTTTGCGAACAC CTTTTATTATTGATGATCACG	T_{HXT7}
NK79	GGCCTGGGACGACGATTGCTTCATTGTTATGAG ATAGTTGATTGTATGCTTGG	P_{ADH1}
NK80	ATAGGGCGAATTGGGTACCGGGCGCCGCACACACC ATAGCTCAAAATGTTCTACTCC	P_{TEF1}
NK81	AAGAGGAAGTCCATATTGTACACCCGATCCGGAGG ATCTCGAGCGTCC	T_{CYC1}
NK82	GGGACGCTCGAAGATCCTCCGGATCGGGTGTACAA TATGGACTTCCTC	P_{ADH1}
NK83	CTATCATTATTACGTAATGACCCACTTCTTTAAGT GACGGGCGATG	T_{HXT7}
NK84	CTCATGCCCGTCACTAAAAGAAGTGGGTATTAC GTAAATAATGATAGGAATGGGA	T_{FBA1}
NK85	TTAGTGGATGCCAGGAATAAAACTGTGGCGCGTACA TTTAATTTCACAG	T_{TDH3}
NK86	ACGTTGAAAATTAAATGTACCGGCCACAGTTATTCT CTGGCATCCACTAAATATAATGG	P_{TDH3}
NK87	GCCTCTCCCCCGCGTGGCCGATTGGCGCCTTG GTTCCCAGAAACAAAGGG	T_{ACT1}
NK88	CTTTGTTCTGGGAACCAAGGCGCCAATCGGCCAA CGCGCGGGGAG	Backbone pRS425
NK89	AAGAGGAAGTCCATATTGTACACCCGATCCGGAGG	Backbone pRS425

	ATCTTCGAGCGTCC	
NK90	GAATAAACACACATAAACAAACAAAATGTCTTCTCT GAATTTACCAAAGAAG	<i>prpE</i>
NK91	CATAAACATACGCGCACAAAAGCAGAGATCATTCTT CTATTGCTTGCTAATTGC	<i>prpE</i>
NK92	CAAATTAGACAAGCAATAGAAGAATGATCTCTGCTT TTGTGCGCGTATG	T _{ACT1}
NK93	CTTTGGTAAAATTCAAGAGAAAGACATTTGTTGTT ATGTGTGTTATTGAAAC	P _{TDH3}
NK94	GTGGATGCCAGGAATAAACTGTGGCGCGTACATT AATTTCAACG	T _{TDH3}
NK95	CATACCCTTGTTCGGAACCAACGAATGGCGC GACGCGC	Backbone pRS424
NK96	GCTACAGGGCGCGTCGCGCCATTGTTGGTCCC GAAACAAAGGG	T _{ACT1}
NK97	GAATACGTTGAAAATTAAATGTACGCGCCACAGTT ATTCCCTGGCATCCAC	P _{TDH3}
NK98	CATACAATCAACTATCTCATAAACAAATGACCATCC AGACACTTGAAAC	<i>ccr</i>
NK99	CATGAATTAAATAAAAGTGTTCGCAAATTATGGGGCA CCACTCAATT	<i>ccr</i>
NK100	CAAGAATTGAGTGGTCCCCATAATTGCGAACACT TTTATTAATT	T _{HXT7}
NK101	GTTGTTCAAGTGTCTGGATGGTCATTGTTATGA GATAGTTGATTGTATG	P _{ADH1}
NK102	CTACAGGGCGCGTCGCGCCATTGAGTACTCCCCA TTATCTTAGCCTAAAAAACCTTC	P _{GAL1}
NK103	CAAGAAGGATAGTAAGCTGGCAAATCATAAAATTAA AGTAGCAGTACTCAACCATTAGTG	T _{SSA1}
NK104	GTTGAAGTACTGCTACTTTAATTGATTGCCAG CTTACTATC	P _{GAL7}
NK105	GAAGCGGAAGAGCGCCCAATACGCAAAGTACTACA TAGAAATATCGAATGGG	T _{PGK1}
NK106	CCCATTGATATTCTATGTAGTACTTGCCTATTG GGCGC	Backbone pRS425
NK107	GAGAAGGTTTTTAGGCTAAGATAATGGGGAGTAC TCGAATGGCGCGACG	Backbone pRS425
NK108	CGCGTCGCGCCATTGAGTACTGGTTCCCAGAAA CAAAGGGTATGAGC	T _{ACT1}

NK109	CATATTGGCCAACATGATCTCTGCTTTGTGCGCGT ATG	T _{ACT1}
NK110	GCACAAAAGCAGAGATCATGTTGGCCAATATGTCC TC	pccE
NK111	ACACACATAAACAAACAAAATGACTATCAAAGTAGT GAGAGG	pccE
NK112	CTCTCACTACTTGATAGTCATTTGTTGTTATGT GTGTTATTGAAAC	P _{TDH3}
NK113	GTATAGCATGAGGTGGCTCGTAATTACTTAAAT CTTGCATTAAATAAATTTC	T _{TDH3}
NK114	GTAAATTACGAGCGACCTCATGCTACCTGAG	T _{ADH1}
NK115	GAAGAGGCCAATACGCAAAGTACTCTCGAGCG TCCCAAAACCTCTC	T _{CYC1}
NK116	AGTACTTGCATTGGCGCTCTCCGCTTCC	Backbone pRS424
NK117	AGTACTCGAATGGCGCGACGCGCCCTG	Backbone pRS424
NK118	CCATATTGTACACCCGAGCGACCTCATGCTATAC	T _{ADH1}
NK119	AGTGAATTACTTAAATCTGCATTAAATAAATTTC C	T _{TDH3}
NK120	GAAAATTATTAAATGCAAGATTAAAGTAAATTCA CTCATTCTTCTATTGCTTGTCTAATTG	prpE
NK121	CAAGTAATACATATTCAAAATGTCTTCTCTGAATT TACCAAAAG	prpE
NK122	CTTGGTAAAATTCAAGAGAAAGACATTGAAATATG TATTACTTGGTTATGG	pFBA1
NK123	CTGCGTAGTGCAGAAAAATAATTGCGAACACTTT ATTAATTGATG	T _{HXT7}
NK124	CATGAATTAAATAAAAGTGTTCGCAAATTATTTCTG CACTACGCAGG	birA
NK125	CTATCTCATAAACAAATGAAGGATAACACCGTGCC	birA
NK126	GGCACGGTGTTCCTCATTGTTATGAGATAGT TGATTGTATGC	P _{ADH1}
NK127	GTATAGCATGAGGTGGCTCGCTGGGTACAATATGGA CTTCC	T _{ADH1}

Table S4 Growth rates with propionate supplementation

Growth on propionate was compared between a *S. cerevisiae* BJ5465 strain carrying the propionyl-CoA synthase gene on plasmid pNK36 or a strain with empty vector control (pRS424). A matlab script was used to determine the growth rate between every consecutive time point.⁸⁹ The maximal growth rate in h⁻¹ as well as the time point at which the maximal growth rate was reached are listed below. As previously mentioned, feeding propionate is toxic for the cell due to propionyl-CoA accumulation from *prpE* expression and growth is reduced. Higher concentrations of propionate lead to slower maximal growth at later time points. Normal growth was observed when expressing the empty vector pRS424.

Plasmid	Propionate feed in mM	Maximal growth rate in h⁻¹	Time of maximal growth in h
pNK36	0	0.47±0.04	10.3±0.3
pNK36	5	0.12±0.04	45.6±28.2
pNK36	50	0.01±0.06	61.9±16.0
pRS424	0	0.4±0.01	11.8±1.0
pRS424	5	0.36±0.06	13.0±1.9
pRS424	50	0.45±0.03	12.6±0.3

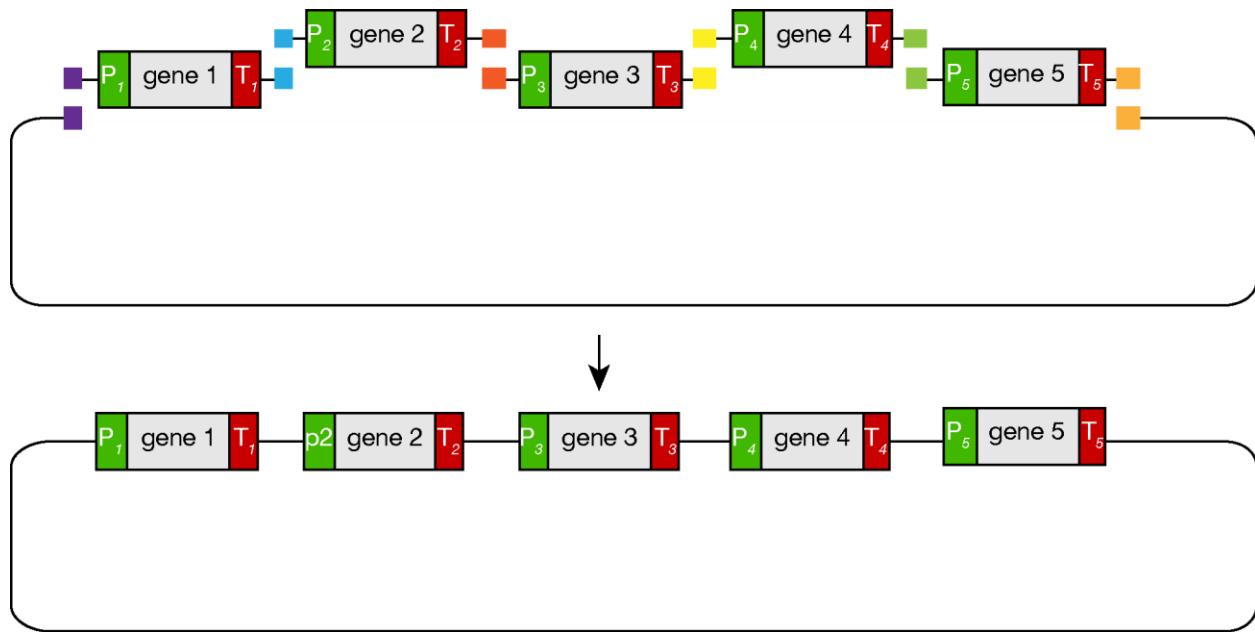


Figure S1. Pathway plasmids were assembled as modular cassettes using CPEC, Gibson assembly or yeast homologous recombination and expressed from 2μ plasmids. Each fragment contains a promoter (P_1 - P_5), gene and terminator (T_1 - T_5) as well as overlapping regions to flanking parts.

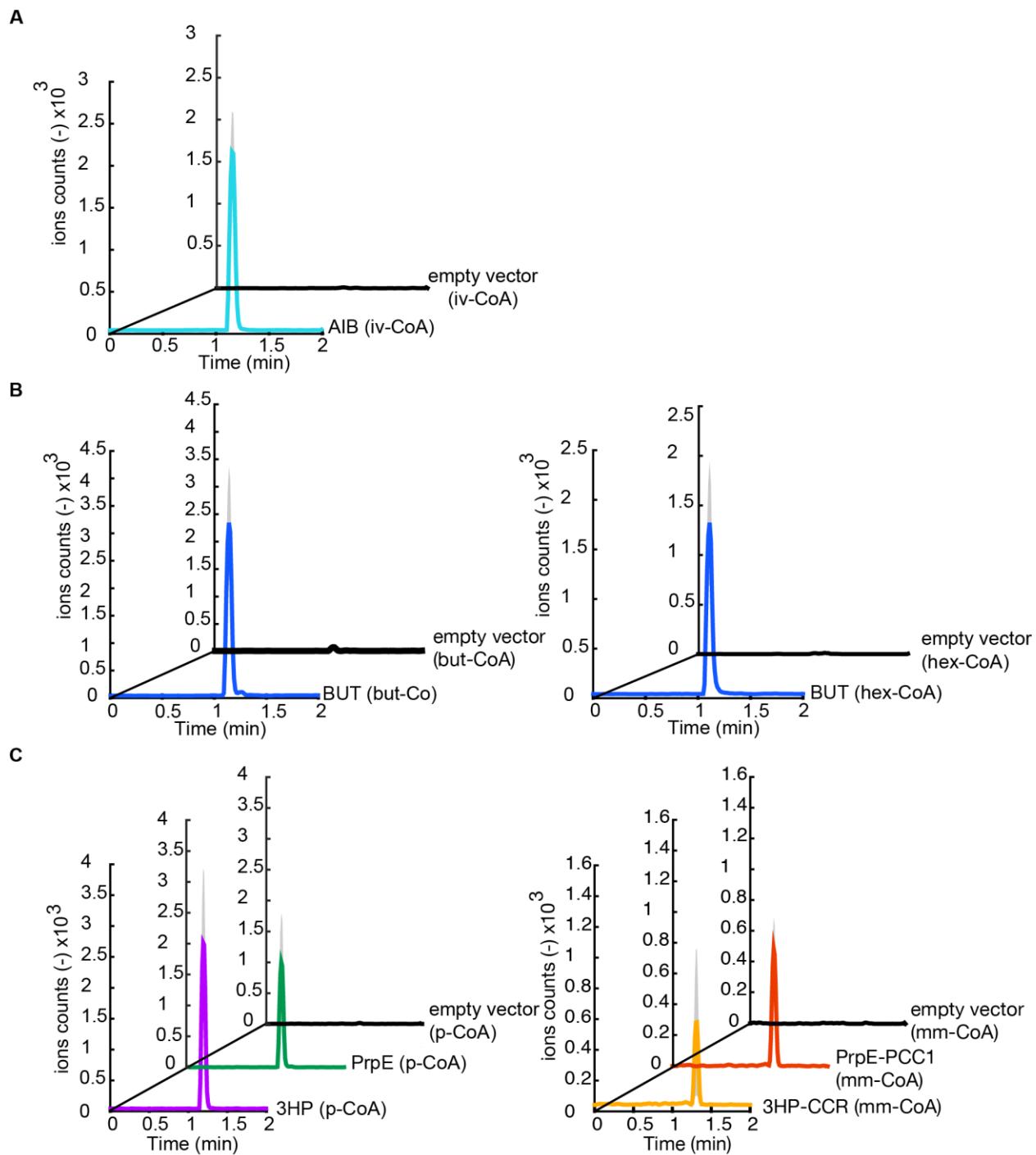


Figure S2. (A) LC-MS/MS chromatograms for strains transformed with the AIB pathway (light blue) or the empty vector control (black). Shown are the means of three biological replica and respective standard deviations (grey). (B) LC-MS/MS chromatograms for strains transformed with the BUT pathway (blue) (butyryl-CoA: left panel; hexanoyl-

CoA: right panel) or the empty vector control (black). Shown are the means of four biological replica and respective standard deviations (grey). (C) LC-MS/MS chromatograms for strains transformed with the 3HP pathway (purple), the PrpE pathway (green) or the empty vector control (black) (left panel) as well as strains transformed with the 3HP-CCR pathway (orange), the PrpE-PCC1 pathway (red) or the empty vector control (black) (right panel). Shown are the means of three biological replica and respective standard deviations (grey). MRM parameters are according to

Table 4.