

Additional file 3

Name	Sequence	Description
Assembly of pF, pFD, pFDA, and pFDA3		
SH1	AAAACCTAGGATGGTTGCTGTTGAAAGAGTTG	AtANS Adh1-cassette FW
SH2	AAAAGGCCGGCCTTAGTCGTTCTTTTCAGAAAC	AtANS Adh1-cassette RV
SH3	AAAAGTCGACATGGAAGAAGACTCTCCAGC	GhDFR Tdh3-cassette FW
SH4	AAAAGCGGCCGCTAATTACATGATTGACCTTC	GhDFR Tdh3-cassette RV
SH5	AAAACCTAGGATGGCTCCAGGTACTTTGAC	GhDFR Adh1-cassette FW
SH6	AAAAGGCCGGCCTTAAGCGAAGATTGGTCAACTG	GhDFR Adh1-cassette RV
SH7	TACGACTCACTATAGGGCGAATTGGGTACCGGGCCCCCTCGAGATTCTTCTAGGCTTTGATGCAAGGTCC	AtF3H HR to pD FW
SH8	GATGAATCTCGGTGTGTATTTATGTCTCAGAGGACAACCTGACCAGTTTATCATTATCAATACTCGCC	AtF3H HR to pD RV
SH9	AAAGTTTACAAATGAATTTTTCCGCCAGGATTTAATTAGAGCTGGAATGATCGTTACGCGCTTCTCGG	At3GT HR to pFDA FW
SH10	CCAAGCGCGCAATTAACCTCACTAAAGGGAACAAAAGCTGGAGCTCGCCTACGGTTCGCCGAAGTATGCTGCTG	At3GT HR to pFDA RV
SH11	ATTATACTAGTATGACTAAGCCATCTGACCCAAC	At3GT to p426-GPD FW
SH12	ATAAACTCGAGTTAGATGATGTTAACAACAGC	At3GT to p426-GPD RV
Assembly of pCPW002,		
CP49	ACTATATGTGAAGGCATGGCTATGGCACGGCAGACATCCGCCAGATCATCAATAGGCACGCTTTCGGTGATGACGGTGAAAACCTC	A-TRP1-B FW
CP50	GTTGAACATTCTTAGGCTGGTGAATCATTTAGACACGGGCATCGTCTCTCGAAAGTGCTGGAACAACACTCAACCCTATCTC	A-TRP1-B RV
CP43	CACCTTTCGAGAGGACGATG	B-AtF3H-N FW
CP44	TTCTAGGCTTTGATGCAAGGTC	B-AtF3H-N RV
CP45	GATCAGCAGCCACGATTGAGTC	N-AtANS-F FW
CP46	TGCCGAACCTTCCCTGTATGAAG	N-AtANS-F RV
CP47	CATACGTTGAAACTACGGCAAAGGATTG	F-2µm-G FW
CP48	AAGGGCCATGACCACCTGAT	F-2µm-G RV
CP93	GCCAGAGGTATAGACATAGCCAGACC	G-GhDFR-O FW
CP94	ATACTCCCTGCACAGATGAGTCAAG	G-ghDFR-O RV
ML108	GTGCCTATTGATGATCTGGCGGAATGTCTGCCGTGCCATAGCCATGCCTTCACATATAGTTGCGCGGAACCCCTATTG	A-E.coli-I FW
ML109	TATTCACGTAGACGGATAGGTATAGCCAGACATCAGCAGCATACTTCGGGAACCGTAGGCGAGAGGCGGTTTGCGTATTGG	A-E.coli-I RV
Assembly of pCPW006 and pCPW007		
CP129	GTCAAGACAACGGTCACTAATCATGTAATTAGTTATGTCACGCTTACATTACG	AaDFR-CYct FW
CP130	CGTGAATGTAAGCGTGACATACTAATTACATGATTAGTGACCGTTGTCTTGAC	CYct-AaDFR RV
CP131	GAAAGCATAGCAATCTAATCTAAGTTTATGATGCACAAGGGTACTG	TEFp-AaDFR FW
CP132	CAGTACCCTTGTCATCATAAACTAGATTAGATTGCTATGCTTTC	AaDFR-TEFp RV
CP133	GTTCTGTTAAGTCTACTAACAAGTAATCATGTAATTAGTTATGTCACGCTTACATTCACG	MtDFR1-CYct FW
CP134	CGTGAATGTAAGCGTGACATACTAATTACATGATTACTTGTTAGTAGACTTAACGAAC	CYct-MtDFR1 RV
CP135	GAAAGCATAGCAATCTAATCTAAGTTTATGGGTTCTATGGCTGAAAC	TEFp-MtDFR1 FW
CP136	GTTTCAGCCATAGAACCCATAAACTTAGATTAGATTGCTATGCTTTTC	MtDFR1-TEFp RV
Replace p426 GTS		
ML009	TCGGTATAGAATATAATCGGGGATGCC	2µm internal FW
ML010	GCGTTTACTGATTACTAGCGAAGCTG	2µm internal RV
ML017	TGTA AACGACGGCCAGT	M13 FW
ML018	TCACACAGGAACAGCTATGAC	M13 RV

X-2::(At4CL3, AtCHS3, AtCHI1) integration		
CP95	GATCTAGGAAGAGAGTAGTGGCGGTTTTAGAGCTAGAAATAGC	X-2 gRNA GTS FW
CP96	CTAAACCGCCCACTACTCTCTCCTAGATCATTTATCTTCACTGC	X-2 gRNA GTS RV
CP85	GTTAATTGTGCTGTTATTGTCTAGATTTTTCTCGGAGATGGCGCATCTATTGCCGTCCAGTATAGCGACCAGCATT	4CL3 CHS3 CHI1 repair FW
CP86	CATTGGAAGTTAGGTGTGGCACAAGCCGATAAGAAAAAGAAAGTAGTGAGGACAGCGCAAATTAAGCCTTCGAGC	4CL3 CHS3 CHI1 repair RV
CP109	GCTCGATCTTCTATCCTCTTTAG	X-2_int diagnosis FW
CP110	CCATGTAAGAACGGAATAAACAG	X-2_int diagnosis RV
CP51	ACGGCGAAACACACCTAATC	X-2_int diagnosis FW
CP55	ATGTCTTCATCCAACGCCTG	X-2_int diagnosis RV
XII-2::(coAtPAL1, coAtC4H, coAtCPR1, coAtCHS3) integration		
CP115	ATAAATGATCTTTGCTTTGTTCTGGTCCCTGTTTTAGAGCTAGAAATAGCAAG	XII-2 gRNA GTS FW
CP116	GCTCTAAACAGGGACCAGAACAAAGCAAAGATCATTTATCTTCACTGCG	XII-2 gRNA GTS RV
CP87	CTGGAAAGGCCTAAGTAATGTCGTAACGCATTCTATCTGTACTCAACTCTCTCTGTGTCATTATCAATACTCGCCAT	PAL1 C4H CPR1 repair FW
CP88	GGAAGAGTAAAAAGGAGTAGAAACATTTTGAAGCTATGGCGGAATTGAGCGCAAGTTC	PAL1 C4H CPR1 repair RV
CP89	CAACTGTTTTAAAGGATCATGAACCTGCGCTCAATCCGCCATAGCTTCAAAATGTTTC	CHS3 repair FW
CP90	GTTCGAGAATGAGAACTTCGTAGTCGGTTAAACAGCTGTAGTGTCTGGCCTCTTAGCAGTATAGCGACCAGCATT	CHS3 repair RV
CP111	CATATTTGTCTTTTCGCGCC	XII-2_int diagnosis FW
CP112	CCTGTTTGGTAAATGAAGGCTAG	XII-2_int diagnosis RV
CP69	ATGACTTCTGCTTTGTACGC	XII-2_int diagnosis FW
CP32	AGTATGCTGTGCTTGGGTG	XII-2_int diagnosis RV
CP100	CGGTCTTCAATTTCTCAAGTTTCAG	XII-2_int diagnosis FW
CP102	CTCCTTCCTTTTCGGTTAGAGC	XII-2_int diagnosis RV
CAN1::(coAtF3H, coAtANS, coGhDFR, coAt3GT) integration		
CP81	ATGACAAATCAAAGAAGACGCCACATAGAGGAGAAGCATATGTACAATGAGCCGGTCCATAGCTTCAAAATGTTTCTAC	DFR 3GT repair FW
CP82	GAATCATTTAGACACGGGCATCGTCTCTCGAAAGGTGGCCTACGGTCCCGAAGTATG	DFR 3GT repair RV
CP83	ATAGCCAGACATCAGCAGCATACTTCGGGAACCGTAGGCCACCTTCGAGAGGACGATG	F3H ANS repair FW
CP84	GAGGATGTAAACAGGATGAATGTAGCCATTTACCCAAGGACTGCGTGACAGAATATGCCAACGCAGAATTTTCGAGTTA	F3H ANS repair RV
CP146	CTTCAGACTTCTTAACCTCTG	CAN1_int diagnosis FW
CP103	CATAACCAACCAAGAACCGATG	CAN1_int diagnosis RV
CP35	GGTCAGGTGCTTTCTCAGG	CAN1_int diagnosis FW
CP26	CCTATTTTGGGCATGTACGG	CAN1_int diagnosis RV
CP108	CCACTTGGCTTACCCAGAAG	CAN1_int diagnosis FW
CP147	CTTTGTGAGAACTGTG	CAN1_int diagnosis RV
EXG1::coCHS3 integration		
CP176	TGATCCTTCTGTCAATATTTAGGTAGTTTTAGAGCTAGAAATAGCAAGTTAAAATAAGGC	EXG1 gRNA GTS FW
CP177	CTAGCTCTAAACTACCTAAATATTGACAGAAGGATCATTTATCTTCACTGCGGAGAAG	EXG1 gRNA GTS RV
CP191	GGTGGTTGTTACTTCTTGAACCATACATTACTCCATCTTGTTCGAGGCTTCCGTACACATAGCTTCAAAATGTTTCTACTCC	coAtCHS3 repair FW
CP167	GTCTAGAACAGGACCAATGGCTCATTAATCAATTCGATACCAATAACAGTGTCGAAGTACAGTATAGCGACCAGCATT	coAtCHS3 repair RV
CP164	CGTTACTGTGTACGTTGTTGACTG	EXG1_int diagnosis FW
CP165	CAAACAGTCCAGTGGGATTCATT	EXG1_int diagnosis RV
CP188	GAGACACATGCACTTGACTGAAG	EXG1_int diagnosis FW
CP189	CACAAGCAGAAGACATGTTACCG	EXG1_int diagnosis RV

SPR1::coRcTAL1 integration		
CP178	TGATCGGAAATATGGTTTGAAAGTAGTTTTAGAGCTAGAAATAGCAAGTTAAAATAAGGC	SPR1 gRNA GTS FW
CP179	CTAGCTCTAAAACTACTTTCAAACCATATTTCCGATCATTTATCTTTCACTGCGGAGAAG	SPR1 gRNA GTS RV
CP190	ACACCTTCTTTATTCGAGACTTTCGCTACTAATCCGTACAACGATGACGGTATTCTGTTGTAAAACGACGGCCAGT	coRcTAL1 repair FW
CP169	CTTCAAAAGCAAATTTTCAATCTTCCATGTCAATAACTGGACCTAACGGTTCATTGAGGCAAATTAAGCCTTCGAGC	coRcTAL1 repair RV
CP162	GGTGGGTGGCTAGTATTGGAG	SPR1_int diagnosis FW
CP163	GATGGTCAATTATGACGCCATATTCG	SPR1_int diagnosis RV
CP174	ATTAATGGAAGTTTTGAGTGGTCATG	SPR1_int diagnosis FW
CP175	CGTCTTGTGCAGGATGATC	SPR1_int diagnosis RV
TSC13::MdECR integration		
CP180	TGATCTGAAAATACCTTGATTCAATGTTTTAGAGCTAGAAATAGCAAGTTAAAATAAGGC	TSC13 gRNA GTS FW
CP181	CTAGCTCTAAACATTGAATCAAGGTATTTTCAGATCATTTATCTTTCACTGCGGAGAAG	TSC13 gRNA GTS RV
CP182	GCTATCTAGAAACCAATTGAGCTATTTGAGAGAGATACATATTTGAATTTAATTTGAAAATGAAGGTTACTGTTGTTTC	coMdECR repair FW
CP183	CCACTTCGTGAAAGCTAATATCTCTTTACCTTGCATTTGGGCATGTTGCAAACAGGAGGATTACAAGAATGGTGGCAAG	coMdECR repair RV
CP184	TGAAAAGGGACTAAGAGCGTG	TSC13_int diagnostic FW
CP185	GATGAAAGCACCGAAAGACC	TSC13_int diagnostic RV
CP186	GACTTTGCCAGTTCAACCAGG	TSC13_int diagnostic FW
CP187	TGCTACTACGCCACTTCGTG	TSC13_int diagnostic RV