

Supplemental Data

Mutations in the substrate-binding pocket of DiacylGlycerol AcylTransferase alter the fatty acid composition of Triacylglycerides in yeast

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Table S1: Codon optimized sequence of DGAT used

Name	Sequence
AtDGAT	ATGGCGATTCTAGATTCTGGCTGGTGTCAACAACAGTTACCGAGAATGGTGGAGGTGAATTTGTCTGACTTGGACAGATTGAGAAGAAGAA AGTCAAGATCCGATAGTAGTAATGGCCTATTATTATCCGGTTCTGATAACAACCTCCCCTAGTGATGATGTAGGAGCTCCTGCAGATGTT AGAGACAGGATAGATTCTGTGGTAAATGATGATGCCCAAGGTACAGCTAATTTGGCCGGGGACAATAATGGCGGTGGTGACAATAATG GTGGAGGAAGAGGTGGCGGTGAAGGTAGGGGTAAATGCGGATGCCACATTACGTACAGACCATCAGTCCCTGCTCATAGACGTGCTAG AGAATCTCCACTAAGTAGTGACGCCATATTCAAGCAGTCTCATGCTGGATTATTCAATTTGTGTGTAGTCGTAATAATTGCTGTGAACTC AAGACTAATTATTGAAAATCTGATGAAGTATGGCTGGTTGATTAGAAGTACTTTTGGTTTTCTCAAGATCTTTGAGAGATTGGCCGCT ATTCATGTGCTGTATATCGCTTTCCATATTTCCACTGGCTGCATTTACGGTTGAAAAGCTAGTCTTACAGAAATACATCTCAGAACCAGT GGTGATTTTCTGACATTATCATCACCATGACTGAAGTCTTATATCCTGTATATGTGACTTTAAGATGTGATTCTGCTTTCTTGAGTGCC GTTACCTTATGTTACTGACATGTATTGTGTGGTTGAAGTTAGTGTCTTATGCGCATACTTCATACGACATTCGTTCTTTGGCAAATGCCG CTGATAAAGCTAACCCAGAAGTTAGCTACTACGTGAGCTTGAAATCACTTGCGTATTTTCATGGTTGCACCCACCTTGTGCTATCAACCTT CATACCCAAGATCTGCATGTATCAGGAAAGGATGGGTTGCAAGACAATTTGCCAACTGGTTATCTTTACAGGGTTCATGGGCTTTATA ATAGAGCAGTACATTAATCCCATTTGTTAGGAACTCAAACATCCGTTAAAAGGTGATCTGCTTTATGCCATAGAACGTGTCTTAAACT TAGCGTACCGAACTTATATGTCTGGTTGTGTATGTTCTATTGCTTCTTCACTTATGGTTGAACATTTTGGCAGAGTTGTTGTGCTTTGGC GACAGAGAGTTTACAAGGATTGGTGGAAACGCAAAATCCGTTGGTGATTATTGGAGAATGTGGAACATGCCAGTTCATAAATGGATGG TTAGGCACATTTACTTTCCATGCTTACGTTTCAAGATACCTAAAACCTCTAGCCATTATTATAGCTTTCTTAGTCTCTGCAGTTTCCATGA ACTTTGTATCGCTGTACCTTGCAGATTATTCAAGCTTTGGGCATTTCTTGGGATCATGTTCCAAGTTCCCTTAGTATTCAATTACGAACTAC TTGCAAGAAAGGTTTGGATCTACTGTTGGGAATATGATATTTTGGTTTATCTTTGTATCTTTGGTCAACCAATGTGTGTTTTGTTGTATT ATCACGATTTGATGAATCGTAAAGGTTTCGATGAGCCATCATCATCACCACCATAA
CeDGAT	ATGGGAAACAAGGAGACCATGGAGACACCCAAGACCAATACGAGACCTCCTCTGCTGAAAAGCATTGTCTGCCCTCTTCCTGTGGATGG GAATAATCCAGCTCAATGTAGTATTGTTGGTGACCGCTCTCTTTGTGCCGCTCCGGATCGGAACCATGATTATCGGCTTCCTGTGCTCA TGCGACTGCTCCCCGTCAACCCGAAATCGAAGTTCGGCTCCAAGGTGGCCAAGTTTATTGACAAGAATGCTTATGGATACTTCCCAATC ACTGTTTCTGAGGACGAAAAGGCATTTCGACCCGAACCAAGGCATACGTGTTTGGGTACGAACCCCATACAATCTACGCCCTAGGTG CCTGGGCTCTTACCGATCGGAGCAGTCTCGCTCCCGTGCCCAAGATCAAGTTCAGTCTGCTTCTATTGCGTTCAACATTCCCATTCTAC GACATATCTGGACCTGGCTGGGTCTGGTGCCTGTCACTCGAAAGTCTTTCATTAAACAACCTTTCTGCGGGTAACTCGTGCATCGTTGTCC CAGGAGGGGTGCAAGAAATGCTGCACCTCGAGCAGGACTCCGAAGTTGCCTTTCTGAAATCCAGGAAGGGTTTTGTCAAGCTGGCCAT TGAGATGGGCTCACCTCTGGTTCCTGTCTTCTCGTTTGGCCAGTCTCAGGCCTACAAATGGTGGAGACCTCAAGGCAAGATCTTTCACA AAATCTATAAGATTGTGAAGCAGCCATTGTCTGTCTGTTGGGGCCGTTTCGGCTCACCTATCCCCTTCCGAGTTCCCATGCACATTGTGA TTGGTAAGCCCATCCAGTTAAAGAAGAACGCCAGCCAACGAACGATGAGGTGAACGAGGTCCACGCTCAGTTCGTTGCAGCCATGCA GGAGTTATTTGAGAAGCACAAAGGCGCAGTACGGATCCAAGGATCTGCAACTTCGCGTTTTGTGA

TaDGAT	ATGGAACCTATTGCCTACAAGGAGGGCGGTCTGCCGCCAAGAGCCGACGGAAAGCGAGCCTCTTCAGGAAATGTGCGAGCTACCAATG GTGCAGCCGGCGGCCCTCGCCTAGGACCTGTTAGTCCTAGAAAAGGCCGAAACCAGGGAAGCTTTGACTCCGGCGAGGCTACACCCAA CGGCATGACCACCCCTTCTCAGGGTTCCGTCTCGAGCTACAGTTCTGAGGATGCCCACACCAACGGAGGCGGGGGCTCTCCCCGGTCTT CAGGCAGCGCTGGTTCTGGCGGCGCCAACGGTGGCCTCAAGCTCCGAGCTCCCCGAGGTGGCCAGTTCTTCGCGACGTTTCAGCTGCT CCTCCGGCCCCGACACCGTCTGACACGGGAGGAGCGCGAAATGTTCTTCGAATCGAGCGAGAGTGGCGAGAAGAGGACGAGTGGGCA AAGCAGGAACCCGGTGTGGTGTCCACCATGATCTGGACCCCCATTCTGATTGGACTACGATGCTTCAATATCTGGCTGTCTGTGGTCAC CTGGCCCCCTGTCGTTCTTGGCTCGAGTGGTTTTTCGGAATGGAGATGAAGAAGGCCTCCTTCTGGGATGTACCATTGGAGCGGAGAAAAC AAACGGTGGCCGTCGCCGGTTTTGTGATGCTGCTCCCTTGTGTCTCCTGGCGTATGTCTGGAGCCTTGTCTGCTGGTTTTCCCTCTGAC CACGCTTCCCATGCTCGGCTACTACATCTGGATTTTCAAGATTGACAAGTCGCCCCGAGAACGGTCAGCGAACACCCTTTCTGAGATACT GGTCCGCCTGGAGACACTTTGCGTCTTACTTTCCGCTACGGCTCATCAAACTCACAACCTCGACCCTTCCCGAAAGTACGTCTTTGCAT ACCATCCCCATGGAATCATTTTCGATTGGTGCCTTTGGCAACTTTGCCACCAACGCCACGGGCTTTTCACGAAAGTTCCCGGGCATTGATC TGCGACTGCTCACGCTGGAGATGAACTTCTGGTGTCCCTGGATAAGGGAGTTTTTGCTTAGCATGGGTGTCTGCTCTGCCGCAAAACGT TCGTGCAACAAGATCCTTTTCGAAGGGCCCCGGTAGCGCTATCATGTTAGTTGTGGGAGGGGGCCGCCGAATCTCTTGATACAGAGCCTGG AACTTACCGACTCACTCTCGGAAGGAAAGGTTTCATCCGAGTCGCTCTGGACAATGGAGCTGATCTCGTACCAGTGCTGGCGTTCCGAG AAAACGACATTTTCGACACCATCTACTATGAGTCCGGTACTGTATGCGGAAAATCCAGGAGGTGGTACGTAAGCGGCTTGGGTTTGGC ACTCCCGTGTTCTCGGGACGTGGCTTCTTCAACTACTCGTTTGGCTTCTTGCTCATCGACGACCTGTTATTGTCTGTTTGTGGAAGACCC ATCAAGGTCCCCAAGCTCCCCGAACATCTGCGAGGATCTGCGCTCAGTACAACTCCTGAAGGAGTCGCTTTGGTGGACCAGTACCACCA GAAGTATGTTGCTGAATTACGACGGGTGTGGGACCTGTATAAGTCCAAGTGGGCGGTTTCGAGAGCTGAGTCTCTGATGATTAAGGGTG TGCAGAACCCCGCCCTGCCCAGAAGTCCGTCCCGCCGTATCCCCCAGCTCAAAGAGTGCCTGCAAGTGCAGCATCTCTATCATTCCGA GAGGTCGACGAGGCGGAGTTCGAGGCCAAGGAAGATGGAGCCACTTCCTCACCCAGTCCATGTCTGCGGCTCTCTACACCGAGGGTT GA
OsDGAT	ATGGTCGGCAGCGATGGCGATGGAGATGGTGGCGGTGGCGAGGCTCATGCTCCTGCTGCACCAGCTCACCACCATCGGCGTCTCTCCA GACCCAGAGGAGGCAGTGGAGCTATTGTTGAAGGTTTTGCCGCCGCACTCCGGAGACGAATCAGGTCGGGTGCCGCGCTGCTGCCCG AGCGTCTTTGGGGGAGACTCGGGTGATGAAGCAGCGTCTGGAGAGCCTTCGTCCTCTTCCTCGAGCTCCCCCTCTCGTCGACGAGGAG GTGATTCCAACGAGCCGAGGCCTCATCCGCGGCTGGAGGTGGAGGAGGGAGAGGAGGTGGAGGAGACTTCTCAGCTTTCACGTTTCG AGCAGCTGCACCAGTACACAGAAAGGCCAAGGAGTCCCCACTGAGCTCGGATGCCATTTTCAAGCAGTCCCATGCCGTTTGTTTAATC TCTGCATCGTCGTGCTCGTGGCTGTCAACTCGCGGCTGATCATTGAGAACCTGATGAAGTACGGTCTACTGATACGAGCCGGCTTCTGG TTCAACGACAAGAGTTTGCAGACTGGCCCCTGCTCATGTGCTGTCTCTCTTCCGGCATTCCCTCTGGGCGCCTTTGCGGTGGAGAAG CTCGCCTTCAACAACGTCATCACCGACGCCGTCGCCACCTGCCTCCATATTTTCTATCCACTACCGAAATTGTTTATCCGTTTCTGGTG ATTCTTAAGTGTGACTCTGCTGTACTTTTCAAGGCTTTCTTTTAAATCTTTATCGCCTGTATCGTGTGGCTAAAGCTGGTTAGTTTTGCTCACA CTAACCACGATATTCGACAGCTGACAATGGGCGGCAAAAAGGTGGACAACGAGCTCTCGACAGTTGACATGGACAATCTTCAACCTCC TACCCTAGGCAACCTAATCTACTTTCATGATGGCCCCACGCTGTGCTACCAGCCGTCTTACCCTCGAACCTCGTGCCTCCGAAAGGGCT GGCTCATCCGGCAGATTATCTTGTACCTGATTTTCACTGGGCTGCAGGGCTTTATCATCGAGCAGTACATCAACCCCATTTGTCGTCAATA GCCAACATCCGCTGAAAGGCGGATTGCTCAATGCGGTTGAGACAGTCCCTCAAATTGTCTCTCCCCAACGTGTATCTGTGGCTGTGTATG TTTTATGCTTTCTTCCACCTTTGGCTGTCTATTCTTGCTGAGATTCTGCGATTCCGGCGATCGAGAATTCTACAAGGACTGGTGGAAACGCC AAGACTATTGACGAGTACTGGCGTAAGTGGAACATGCCCGTCCACAAGTGGGTGGTGCGCCATATTTATTTTCCCTGCATGCGAAACGG CATCAGCAAAGAGGTGGCCGTCCTCATCTCTTCTGCTGTGCTTCACGAGATCTGTGTTGCCGTGCCTTGTGCAATTCTGAA ATTCTGGGCTTTTCTGGGTATCATGCTGCAGATCCCCCTCATCGTTCTGACCGCCTACCTCAAGTCCAAGTTCGAGACACCATGGTGGG

	AAACATGATTTTCTGGTTTTTTTTCTGCATTTACGGCCAGCCCATGTGTTTGCTGCTGTACTACCACGACGTCATGAACCGCATAGAAAA GGCCCGGTGA
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Table S2: Primer list used for site saturation mutagenesis of 10 sites in the AtDGAT sequence

Name	Sequence
RF-ATDGAT_f_F235C	ATGTGATTCTGCTTGTTTGAGTGGCGTTACC
RF-ATDGAT_f_F235D	ATGTGATTCTGCTGATTTGAGTGGCGTTACC
RF-ATDGAT_f_F235E	ATGTGATTCTGCTGAATTGAGTGGCGTTACC
RF-ATDGAT_f_F235G	ATGTGATTCTGCTGGTTTGAGTGGCGTTACC
RF-ATDGAT_f_F235H	ATGTGATTCTGCTCATTTGAGTGGCGTTACC
RF-ATDGAT_f_F235I	ATGTGATTCTGCTATCTTGAGTGGCGTTACC
RF-ATDGAT_f_F235K	ATGTGATTCTGCTAAATTGAGTGGCGTTACC
RF-ATDGAT_f_F235L	ATGTGATTCTGCTCTTTTGAGTGGCGTTACC
RF-ATDGAT_f_F235M	ATGTGATTCTGCTATGTTGAGTGGCGTTACC
RF-ATDGAT_f_F235N	ATGTGATTCTGCTAACTTGAGTGGCGTTACC
RF-ATDGAT_f_F235P	ATGTGATTCTGCTCCATTGAGTGGCGTTACC
RF-ATDGAT_f_F235Q	ATGTGATTCTGCTCAGTTGAGTGGCGTTACC
RF-ATDGAT_f_F235R	ATGTGATTCTGCTCGTTTGAGTGGCGTTACC
RF-ATDGAT_f_F235S	ATGTGATTCTGCTAGCTTGAGTGGCGTTACC
RF-ATDGAT_f_F235T	ATGTGATTCTGCTACGTTGAGTGGCGTTACC
RF-ATDGAT_f_F235V	ATGTGATTCTGCTGTTTTGAGTGGCGTTACC
RF-ATDGAT_f_F235W	ATGTGATTCTGCTTGTTTGAGTGGCGTTACC
RF-ATDGAT_f_F235Y	ATGTGATTCTGCTTATTTGAGTGGCGTTACC
RF-ATDGAT_f_V239C	TTCTTGAGTGGCTGTACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239D	TTCTTGAGTGGCGATACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239E	TTCTTGAGTGGCGAAACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239F	TTCTTGAGTGGCTTTACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239G	TTCTTGAGTGGCGGTACCCTTATGTTACTGACAT

RF-ATDGAT_f_V239H	TTCTTGAGTGGCCATACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239I	TTCTTGAGTGGCATCACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239K	TTCTTGAGTGGCAAAACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239L	TTCTTGAGTGGCCTTACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239M	TTCTTGAGTGGCATGACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239N	TTCTTGAGTGGCAACACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239P	TTCTTGAGTGGCCCAACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239Q	TTCTTGAGTGGCCAGACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239R	TTCTTGAGTGGCCGTACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239S	TTCTTGAGTGGCAGCACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239T	TTCTTGAGTGGCACGACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239W	TTCTTGAGTGGCTGGACCCTTATGTTACTGACAT
RF-ATDGAT_f_V239Y	TTCTTGAGTGGCTATACCCTTATGTTACTGACAT
RF-ATDGAT_f_T240C	GAGTGGCGTTTGTCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240D	GAGTGGCGTTGATCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240E	GAGTGGCGTTGAACTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240F	GAGTGGCGTTTTTCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240G	GAGTGGCGTTGGTCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240H	GAGTGGCGTTCATCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240I	GAGTGGCGTTATCCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240K	GAGTGGCGTTAAACTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240L	GAGTGGCGTTCCTTCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240M	GAGTGGCGTTATGCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240N	GAGTGGCGTTAACCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240P	GAGTGGCGTTCCACTTATGTTACTGACATGTATTGT

RF-ATDGAT_f_T240Q	GAGTGGCGTTCAGCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240R	GAGTGGCGTTCGTCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240S	GAGTGGCGTTAGCCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240V	GAGTGGCGTTGTTCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240W	GAGTGGCGTTTGGCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_T240Y	GAGTGGCGTTTATCTTATGTTACTGACATGTATTGT
RF-ATDGAT_f_L244C	GTTACCCTTATGTTATGTACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244D	GTTACCCTTATGTTAGATACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244E	GTTACCCTTATGTTAGAAACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244F	GTTACCCTTATGTTATTTACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244G	GTTACCCTTATGTTAGGTACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244H	GTTACCCTTATGTTACATACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244I	GTTACCCTTATGTTAATCACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244K	GTTACCCTTATGTTAAAAACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244M	GTTACCCTTATGTTAATGACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244N	GTTACCCTTATGTTAAACACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244P	GTTACCCTTATGTTACCAACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244Q	GTTACCCTTATGTTACAGACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244R	GTTACCCTTATGTTACGTACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244S	GTTACCCTTATGTTAAGCACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244T	GTTACCCTTATGTTAACGACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244V	GTTACCCTTATGTTAGTTACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244W	GTTACCCTTATGTTATGGACATGTATTGTGTGGTTG
RF-ATDGAT_f_L244Y	GTTACCCTTATGTTATATACATGTATTGTGTGGTTG
RF-ATDGAT_f_I247C	GTTACTGACATGTTGTGTGTGGTTGAAGTTAGTG

RF-ATDGAT_f_I247D	GTTACTGACATGTGATGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247E	GTTACTGACATGTGAAGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247F	GTTACTGACATGTTTTGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247G	GTTACTGACATGTGGTGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247H	GTTACTGACATGTCATGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247K	GTTACTGACATGTAAAGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247L	GTTACTGACATGTCTTGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247M	GTTACTGACATGTATGGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247N	GTTACTGACATGTAAAGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247P	GTTACTGACATGTCCAGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247Q	GTTACTGACATGTCAGGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247R	GTTACTGACATGTCTGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247S	GTTACTGACATGTAGCGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247T	GTTACTGACATGTACGGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247V	GTTACTGACATGTGTTGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247W	GTTACTGACATGTTGGGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_I247Y	GTTACTGACATGTTATGTGTGGTTGAAGTTAGTG
RF-ATDGAT_f_V248C	TTACTGACATGTATTTGTTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248D	TTACTGACATGTATTGATTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248E	TTACTGACATGTATTGAATGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248F	TTACTGACATGTATTTTTTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248G	TTACTGACATGTATTGGTTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248H	TTACTGACATGTATTCATTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248I	TTACTGACATGTATTATCTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248K	TTACTGACATGTATTAATGGTTGAAGTTAGTGTCTTATGC

RF-ATDGAT_f_V248L	TTACTGACATGTATTCTTTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248M	TTACTGACATGTATTATGTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248N	TTACTGACATGTATTAAC TGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248P	TTACTGACATGTATTCCATGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248Q	TTACTGACATGTATT CAGTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248R	TTACTGACATGTATT CGTTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248S	TTACTGACATGTATTAGCTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248T	TTACTGACATGTATTACGTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248W	TTACTGACATGTATTTGGTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_V248Y	TTACTGACATGTATTTATTGGTTGAAGTTAGTGTCTTATGC
RF-ATDGAT_f_M289C	ACTTGCGTATTTCTGTGTTGCACCCACCTT
RF-ATDGAT_f_M289D	ACTTGCGTATTT CGATGTTGCACCCACCTT
RF-ATDGAT_f_M289E	ACTTGCGTATTT CGAAGTTGCACCCACCTT
RF-ATDGAT_f_M289F	ACTTGCGTATTTCTTTGTTGCACCCACCTT
RF-ATDGAT_f_M289G	ACTTGCGTATTT CGGTGTTGCACCCACCTT
RF-ATDGAT_f_M289H	ACTTGCGTATTTCCATGTTGCACCCACCTT
RF-ATDGAT_f_M289I	ACTTGCGTATTT CATCGTTGCACCCACCTT
RF-ATDGAT_f_M289K	ACTTGCGTATTTCAAAGTTGCACCCACCTT
RF-ATDGAT_f_M289L	ACTTGCGTATTTCCTTGTTGCACCCACCTT
RF-ATDGAT_f_M289N	ACTTGCGTATTTCAACGTTGCACCCACCTT
RF-ATDGAT_f_M289P	ACTTGCGTATTTCCAGTTGCACCCACCTT
RF-ATDGAT_f_M289Q	ACTTGCGTATTTCCAGTTGCACCCACCTT
RF-ATDGAT_f_M289R	ACTTGCGTATTTCCGTGTTGCACCCACCTT
RF-ATDGAT_f_M289S	ACTTGCGTATTT CAGCGTTGCACCCACCTT
RF-ATDGAT_f_M289T	ACTTGCGTATTT CACGGTTGCACCCACCTT

RF-ATDGAT_f_M289V	ACTTGCGTATTTTCGTTGTTGCACCCACCTT
RF-ATDGAT_f_M289W	ACTTGCGTATTTCTGGGTTGCACCCACCTT
RF-ATDGAT_f_M289Y	ACTTGCGTATTTCTATGTTGCACCCACCTT
RF-ATDGAT_f_V290C	TTGCGTATTTTCATGTGTGCACCCACCTTGTG
RF-ATDGAT_f_V290D	TTGCGTATTTTCATGGATGCACCCACCTTGTG
RF-ATDGAT_f_V290E	TTGCGTATTTTCATGGAAGCACCCACCTTGTG
RF-ATDGAT_f_V290F	TTGCGTATTTTCATGTTTGCACCCACCTTGTG
RF-ATDGAT_f_V290G	TTGCGTATTTTCATGGGTGCACCCACCTTGTG
RF-ATDGAT_f_V290H	TTGCGTATTTTCATGCATGCACCCACCTTGTG
RF-ATDGAT_f_V290I	TTGCGTATTTTCATGATCGCACCCACCTTGTG
RF-ATDGAT_f_V290K	TTGCGTATTTTCATGAAAGCACCCACCTTGTG
RF-ATDGAT_f_V290L	TTGCGTATTTTCATGCTTGCACCCACCTTGTG
RF-ATDGAT_f_V290M	TTGCGTATTTTCATGATGGCACCCACCTTGTG
RF-ATDGAT_f_V290N	TTGCGTATTTTCATGAACGCACCCACCTTGTG
RF-ATDGAT_f_V290P	TTGCGTATTTTCATGCCAGCACCCACCTTGTG
RF-ATDGAT_f_V290Q	TTGCGTATTTTCATGCAGGCACCCACCTTGTG
RF-ATDGAT_f_V290R	TTGCGTATTTTCATGCGTGCACCCACCTTGTG
RF-ATDGAT_f_V290S	TTGCGTATTTTCATGAGCGCACCCACCTTGTG
RF-ATDGAT_f_V290T	TTGCGTATTTTCATGACGGCACCCACCTTGTG
RF-ATDGAT_f_V290W	TTGCGTATTTTCATGTGGGCACCCACCTTGTG
RF-ATDGAT_f_V290Y	TTGCGTATTTTCATGTATGCACCCACCTTGTG
RF-ATDGAT_f_M369C	GTCTGGTTGTGTTGTTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369D	GTCTGGTTGTGTGATTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369E	GTCTGGTTGTGTGAATTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369F	GTCTGGTTGTGTTTTTCTATTGCTTCTTTCACTTATGG

RF-ATDGAT_f_M369G	GTCTGGTTGTGTGGTTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369H	GTCTGGTTGTGTCATTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369I	GTCTGGTTGTGTATCTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369K	GTCTGGTTGTGTAAATTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369L	GTCTGGTTGTGTCTTTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369N	GTCTGGTTGTGTAACCTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369P	GTCTGGTTGTGTCCATTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369Q	GTCTGGTTGTGTCAGTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369R	GTCTGGTTGTGTCGTTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369S	GTCTGGTTGTGTAGCTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369T	GTCTGGTTGTGTACGTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369V	GTCTGGTTGTGTGTTTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369W	GTCTGGTTGTGTTGGTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_M369Y	GTCTGGTTGTGTTATTTCTATTGCTTCTTTCACTTATGG
RF-ATDGAT_f_F373C	TGTATGTTCTATTGCTGTTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373D	TGTATGTTCTATTGCGATTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373E	TGTATGTTCTATTGCGAATTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373G	TGTATGTTCTATTGCGGTTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373H	TGTATGTTCTATTGCCATTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373I	TGTATGTTCTATTGCATCTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373K	TGTATGTTCTATTGCAAATTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373L	TGTATGTTCTATTGCCTTTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373M	TGTATGTTCTATTGCATGTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373N	TGTATGTTCTATTGCAACTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373P	TGTATGTTCTATTGCCCATTTCACTTATGGTTGAACATTT

RF-ATDGAT_f_F373Q	TGTATGTTCTATTGCCAGTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373R	TGTATGTTCTATTGCCGTTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373S	TGTATGTTCTATTGCAGCTTTCACCTTATGGTTGAACATTT
RF-ATDGAT_f_F373T	TGTATGTTCTATTGCACGTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373V	TGTATGTTCTATTGCGTTTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373W	TGTATGTTCTATTGCTGGTTTCACTTATGGTTGAACATTT
RF-ATDGAT_f_F373Y	TGTATGTTCTATTGCTATTTTCACTTATGGTTGAACATTT

Table S3: 77 Identified TAGs

Rt (min)	[M+NH4] ⁺ m/z	TG ACN:DB	TG*	TG type	[AA]	[AA] ⁺ m/z	[AB]	[AB] ⁺ m/z	[AC]	[AC] ⁺ m/z	[BC]	[BC] ⁺ m/z
7.72	738.6602	TG 42:1	TG 14:0/14:0/14:1	AAB	DG 14:0/14:0	495.4328	DG 14:0/14:1	493.4196				
7.85	738.6602	TG 42:1	TG 16:0/16:0/10:1	AAB	DG 16:0/16:0	551.4988	DG 16:0/10:1	465.3859				
7.72	738.6602	TG 42:1	TG 16:0/16:1/10:0	ABC			DG 16:0/16:1	549.4805	DG 16:0/10:0	467.4018	DG 16:1/10:0	465.3875
7.85	738.6602	TG 42:1	TG 18:0/16:1/8:0	ABC			DG 18:0/16:1	577.5100	DG 18:0/8:0	467.4020	DG 16:1/8:0	437.3611
7.72	738.6602	TG 42:1	TG 18:1/14:0/10:0	ABC			DG 18:1/14:0	549.4805	DG 18:1/10:0	493.4188	DG 14:0/10:0	439.3775
7.74	738.6602	TG 42:1	TG 18:1/16:0/8:0	ABC			DG 18:1/16:0	577.5098	DG 18:1/8:0	465.3875	DG 16:0/8:0	439.3710
6.86	736.6448	TG 42:2	TG 16:0/16:1/10:1	ABC			DG 16:0/16:1	549.4860	DG 16:0/10:1	465.3857	DG 16:1/10:1	463.3741
6.73	736.6448	TG 42:2	TG 16:1/16:1/10:0	AAB	DG 16:1/16:1	547.4678	DG 16:1/10:0	465.3873				
6.73	736.6448	TG 42:2	TG 18:1/16:1/8:0	ABC			DG 18:1/16:1	575.4968	DG 18:1/8:0	465.3873	DG 16:1/8:0	437.3558
9.26	766.6918	TG 44:1	TG 16:0/16:1/12:0	ABC			DG 16:0/16:1	549.4841	DG 16:0/12:0	495.4371	DG 16:1/12:0	493.4169
9.26	766.6918	TG 44:1	TG 18:0/16:1/10:0	ABC			DG 18:0/16:1	577.5115	DG 18:0/10:0	495.4371	DG 16:1/10:0	465.3856
9.26	766.6918	TG 44:1	TG 18:1/16:0/10:0	ABC			DG 18:1/16:0	577.5115	DG 18:1/10:0	493.4169	DG 16:0/10:0	467.4017
7.98	764.6761	TG 44:2	TG 16:1/16:1/12:0	AAB	DG 16:1/16:1	547.4633	DG 16:1/12:0	493.4160				
7.98	764.6761	TG 44:2	TG 18:1/14:1/12:0	ABC			DG 18:1/14:1	547.4633	DG 18:1/12:0	521.4539	DG 14:1/12:0	465.3881
8.12	764.6761	TG 44:2	TG 18:1/16:0/10:1	ABC			DG 18:1/16:0	577.5167	DG 18:1/10:1	491.4056	DG 16:0/10:1	465.3852
7.98	764.6761	TG 44:2	TG 18:1/16:1/10:0	ABC			DG 18:1/16:1	575.4941	DG 18:1/10:0	493.4160	DG 16:1/10:0	465.3881
8.01	764.6761	TG 44:2	TG 18:1/18:1/8:0	AAB	DG 18:1/18:1	603.5283	DG 18:1/8:0	465.3863				
11.10	794.7227	TG 46:1	TG 14:0/14:0/18:1	AAB	DG 14:0/14:0	495.4322	DG 14:0/18:1	549.4789				
11.10	794.7227	TG 46:1	TG 16:0/16:0/14:1	AAB	DG 16:0/16:0	551.5011	DG 16:0/14:1	521.4484				
11.10	794.7227	TG 46:1	TG 16:0/16:1/14:0	ABC			DG 16:0/16:1	549.4789	DG 16:0/14:0	523.4640	DG 16:1/14:0	521.4479
11.10	794.7227	TG 46:1	TG 18:0/14:0/14:1	ABC			DG 18:0/14:0	551.4974	DG 18:0/14:1	549.4775	DG 14:0/14:1	493.4263
11.16	794.7227	TG 46:1	TG 18:0/16:1/12:0	ABC			DG 18:0/16:1	577.5088	DG 18:0/12:0	523.4681	DG 16:1/12:0	493.4178
11.16	794.7227	TG 46:1	TG 18:0/18:1/10:0	ABC			DG 18:0/18:1	605.5494	DG 18:0/10:0	495.4345	DG 18:1/10:0	493.4178
11.10	794.7227	TG 46:1	TG 18:1/16:0/12:0	ABC			DG 18:1/16:0	577.5081	DG 18:1/12:0	521.4479	DG 16:0/12:0	495.4322
9.51	792.7070	TG 46:2	TG 16:0/16:1/14:1	ABC			DG 16:0/16:1	549.4792	DG 16:0/14:1	521.4482	DG 16:1/14:1	519.4343

9.51	792.7070	TG 46:2	TG 16:1/16:1/14:0	AAB	DG 16:1/16:1	547.4630	DG 16:1/14:0	521.4482				
9.51	792.7070	TG 46:2	TG 18:1/14:0/14:1	ABC			DG 18:1/14:0	549.4792	DG 18:1/14:1	547.4630	DG 14:0/14:1	493.4175
9.51	792.7070	TG 46:2	TG 18:1/16:1/12:0	ABC			DG 18:1/16:1	575.4938	DG 18:1/12:0	521.4482	DG 16:1/12:0	493.4175
9.51	792.7070	TG 46:2	TG 18:1/18:1/10:0	AAB	DG 18:1/18:1	603.5281	DG 18:1/10:0	493.4175				
13.38	822.7537	TG 48:1	TG 16:0/16:0/16:1	AAB	DG 16:0/16:0	551.4972	DG 16:0/16:1	549.4791				
13.69	822.7537	TG 48:1	TG 18:0/16:0/14:1	ABC			DG 18:0/16:0	579.5286	DG 18:0/14:1	549.4775	DG 16:0/14:1	521.4537
13.38	822.7537	TG 48:1	TG 18:0/16:1/14:0	ABC			DG 18:0/16:1	577.5102	DG 18:0/14:0	551.4972	DG 16:1/14:0	521.4535
13.38	822.7537	TG 48:1	TG 18:0/18:1/12:0	ABC			DG 18:0/18:1	605.5508	DG 18:0/12:0	523.4642	DG 18:1/12:0	521.4535
13.38	822.7537	TG 48:1	TG 18:1/16:0/14:0	ABC			DG 18:1/16:0	577.5102	DG 18:1/14:0	549.4791	DG 16:0/14:0	523.4642
11.42	820.7386	TG 48:2	TG 16:1/16:1/16:0	AAB	DG 16:1/16:1	547.4631	DG 16:1/16:0	549.4775				
11.64	820.7386	TG 48:2	TG 18:0/16:1/14:1	ABC			DG 18:0/16:1	577.5085	DG 18:0/14:1	549.4774	DG 16:1/14:1	519.4379
11.47	820.7386	TG 48:2	TG 18:1/16:0/14:1	ABC			DG 18:1/16:0	577.5179	DG 18:1/14:1	547.4631	DG 16:0/14:1	521.4466
11.42	820.7386	TG 48:2	TG 18:1/16:1/14:0	ABC			DG 18:1/16:1	575.4940	DG 18:1/14:0	549.4775	DG 16:1/14:0	521.4467
11.42	820.7386	TG 48:2	TG 18:1/18:1/12:0	AAB	DG 18:1/18:1	603.5284	DG 18:1/12:0	521.4467				
9.78	818.7231	TG 48:3	TG 16:1/16:1/16:1	AAA	TG 16:1/16:1	547.4633						
9.83	818.7231	TG 48:3	TG 18:1/16:1/14:1	ABC			DG 18:1/16:1	575.4953	DG 18:1/14:1	547.4626	DG 16:1/14:1	519.4393
16.32	850.7854	TG 50:1	TG 16:0/16:0/18:1	AAB	DG 16:0/16:0	551.4937	DG 16:0/18:1	577.5084				
16.42	850.7854	TG 50:1	TG 18:0/16:0/16:1	ABC			DG 18:0/16:0	579.5245	DG 18:0/16:1	577.5083	DG 16:0/16:1	549.4791
16.59	850.7854	TG 50:1	TG 18:0/18:0/14:1	AAB	DG 18:0/18:0	607.5674	DG 18:0/14:1	549.4791				
16.42	850.7854	TG 50:1	TG 18:0/18:1/14:0	ABC			DG 18:0/18:1	605.5411	DG 18:0/14:0	551.4935	DG 18:1/14:0	549.4791
16.59	850.7854	TG 50:1	TG 20:0/16:0/14:1	ABC			DG 20:0/16:0	607.5674	DG 20:0/14:1	577.5084	DG 16:0/14:1	521.4571
16.59	850.7854	TG 50:1	TG 20:0/16:1/14:0	ABC			DG 20:0/16:1	605.5413	DG 20:0/14:0	579.5247	DG 16:1/14:0	521.4571
13.84	848.7698	TG 50:2	TG 16:0/16:0/18:2	AAB	DG 16:0/16:0	551.4991	DG 16:0/18:2	575.4937				
13.95	848.7698	TG 50:2	TG 16:1/16:1/18:0	AAB	DG 16:1/16:1	547.4634	DG 16:1/18:0	577.5089				
14.17	848.7698	TG 50:2	TG 18:0/18:1/14:1	ABC			DG 18:0/18:1	605.5416	DG 18:0/14:1	549.4776	DG 18:1/14:1	547.4632
13.84	848.7698	TG 50:2	TG 18:0/18:2/14:0	ABC			DG 18:0/18:2	603.5242	DG 18:0/14:0	551.4991	DG 18:2/14:0	547.4683
13.84	848.7698	TG 50:2	TG 18:1/16:0/16:1	ABC			DG 18:1/16:0	577.5084	DG 18:1/16:1	575.4937	DG 16:0/16:1	549.4791
13.84	848.7698	TG 50:2	TG 18:1/18:1/14:0	AAB	DG 18:1/18:1	603.5242	DG 18:1/14:0	549.4791				

11.91	846.7542	TG 50:3	TG 16:1/16:1/18:1	AAB	DG 16:1/16:1	547.4630	DG 16:1/18:1	575.4939					
11.91	846.7542	TG 50:3	TG 18:1/18:1/14:1	AAB	DG 18:1/18:1	603.5283	DG 18:1/14:1	547.4630					
20.10	878.8167	TG 52:1	TG 18:0/18:0/16:1	AAB	DG 18:0/18:0	607.5560	DG 18:0/16:1	577.5085					
20.10	878.8167	TG 52:1	TG 18:0/18:1/16:0	ABC			DG 18:0/18:1	605.5394	DG 18:0/16:0	579.5248	DG 18:1/16:0	577.5085	
20.22	878.8167	TG 52:1	TG 20:0/16:0/16:1	ABC			DG 20:0/16:0	607.5555	DG 20:0/16:1	605.5391	DG 16:0/16:1	549.4825	
20.22	878.8167	TG 52:1	TG 20:0/18:1/14:0	ABC			DG 20:0/18:1	633.5798	DG 20:0/14:0	579.5244	DG 18:1/14:0	549.4825	
17.14	876.8010	TG 52:2	TG 16:1/16:1/20:0	AAB	DG 16:1/16:1	547.4756	DG 16:1/20:0	605.5393					
17.03	876.8010	TG 52:2	TG 18:0/18:1/16:1	ABC			DG 18:0/18:1	605.5394	DG 18:0/16:1	577.5084	DG 18:1/16:1	575.4937	
16.97	876.8010	TG 52:2	TG 18:1/18:1/16:0	AAB	DG 18:1/18:1	603.5246	DG 18:1/16:0	577.5087					
16.97	876.8010	TG 52:2	TG 20:1/16:0/16:1	ABC			DG 20:1/16:0	605.5397	DG 20:1/16:1	603.5246	DG 16:0/16:1	549.4813	
14.44	874.7855	TG 52:3	TG 16:1/16:1/20:1	AAB	DG 16:1/16:1	547.4737	DG 16:1/20:1	603.5241					
14.44	874.7855	TG 52:3	TG 17:1/17:1/18:1	AAB	DG 17:1/17:1	575.4925	DG 17:1/18:1	589.5135					
14.44	874.7855	TG 52:3	TG 18:1/18:1/16:1	AAB	DG 18:1/18:1	603.5241	DG 18:1/16:1	575.4936					
24.64	906.8476	TG 54:1	TG 18:0/18:0/18:1	AAB	DG 18:0/18:0	607.5560	DG 18:0/18:1	605.5395					
24.89	906.8476	TG 54:1	TG 20:0/18:0/16:1	ABC			DG 20:0/18:0	635.5955	DG 20:0/16:1	605.5393	DG 18:0/16:1	577.5083	
24.64	906.8476	TG 54:1	TG 20:0/18:1/16:0	ABC			DG 20:0/18:1	633.5725	DG 20:0/16:0	607.5560	DG 18:1/16:0	577.5123	
20.82	904.8320	TG 54:2	TG 16:1/16:1/22:0	AAB	DG 16:1/16:1	547.4652	DG 16:1/22:0	633.5709					
20.82	904.8320	TG 54:2	TG 18:1/18:1/18:0	AAB	DG 18:1/18:1	603.5236	DG 18:1/18:0	605.5405					
20.82	904.8320	TG 54:2	TG 20:0/18:1/16:1	ABC			DG 20:0/18:1	633.5755	DG 20:0/16:1	605.5405	DG 18:1/16:1	575.4949	
20.87	904.8320	TG 54:2	TG 20:1/18:0/16:1	ABC			DG 20:1/18:0	633.5704	DG 20:1/16:1	603.5243	DG 18:0/16:1	577.5103	
20.82	904.8320	TG 54:2	TG 20:1/18:1/16:0	ABC			DG 20:1/18:1	631.5554	DG 20:1/16:0	605.5405	DG 18:1/16:0	577.5133	
17.69	902.8167	TG 54:3	TG 16:1/16:1/22:1	AAB			DG 16:1/16:1	547.4689	DG 16:1/22:1	631.5568			
17.69	902.8167	TG 54:3	TG 18:1/18:1/18:1	AAA	TG 18:1/18:1	603.5240							
17.69	902.8167	TG 54:3	TG 20:1/18:1/16:1	ABC			DG 20:1/18:1	631.5559	DG 20:1/16:1	603.5240	DG 18:1/16:1	575.4935	

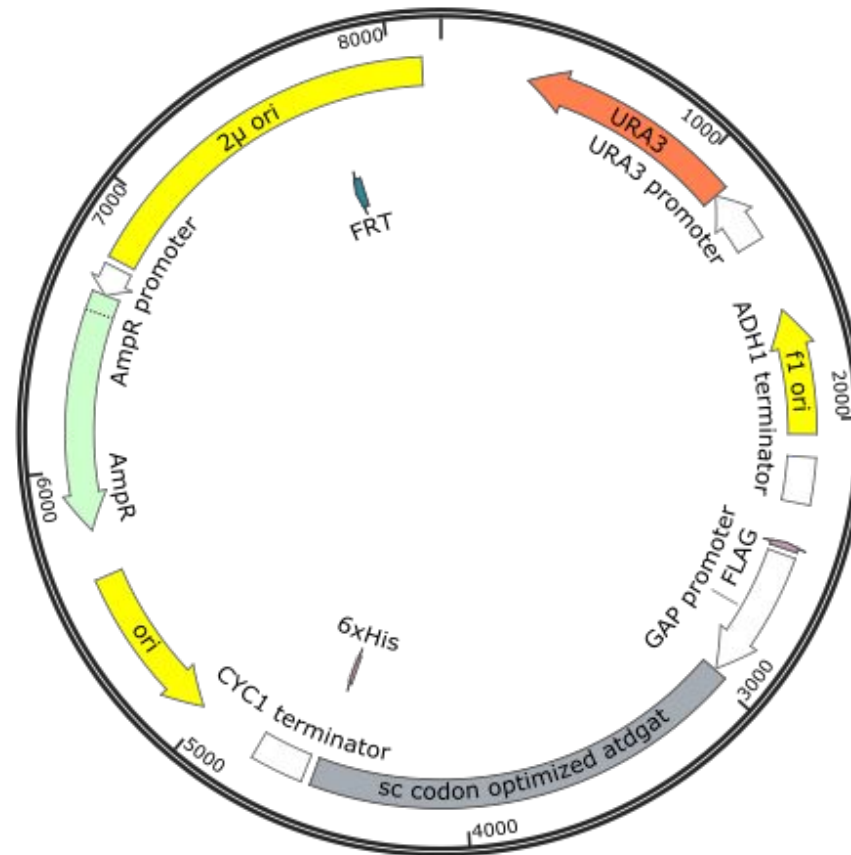


Figure S1: pESC-GAP-ATDGAT plasmid map for the expression of ATDGAT in H1246 cells. The amplified codon optimized sequence of *A. thaliana* DGAT1 (At2g19450) cDNA was ligated into pESC-pGAP vector to produce pESC-pGAP-ATDGAT expression vector

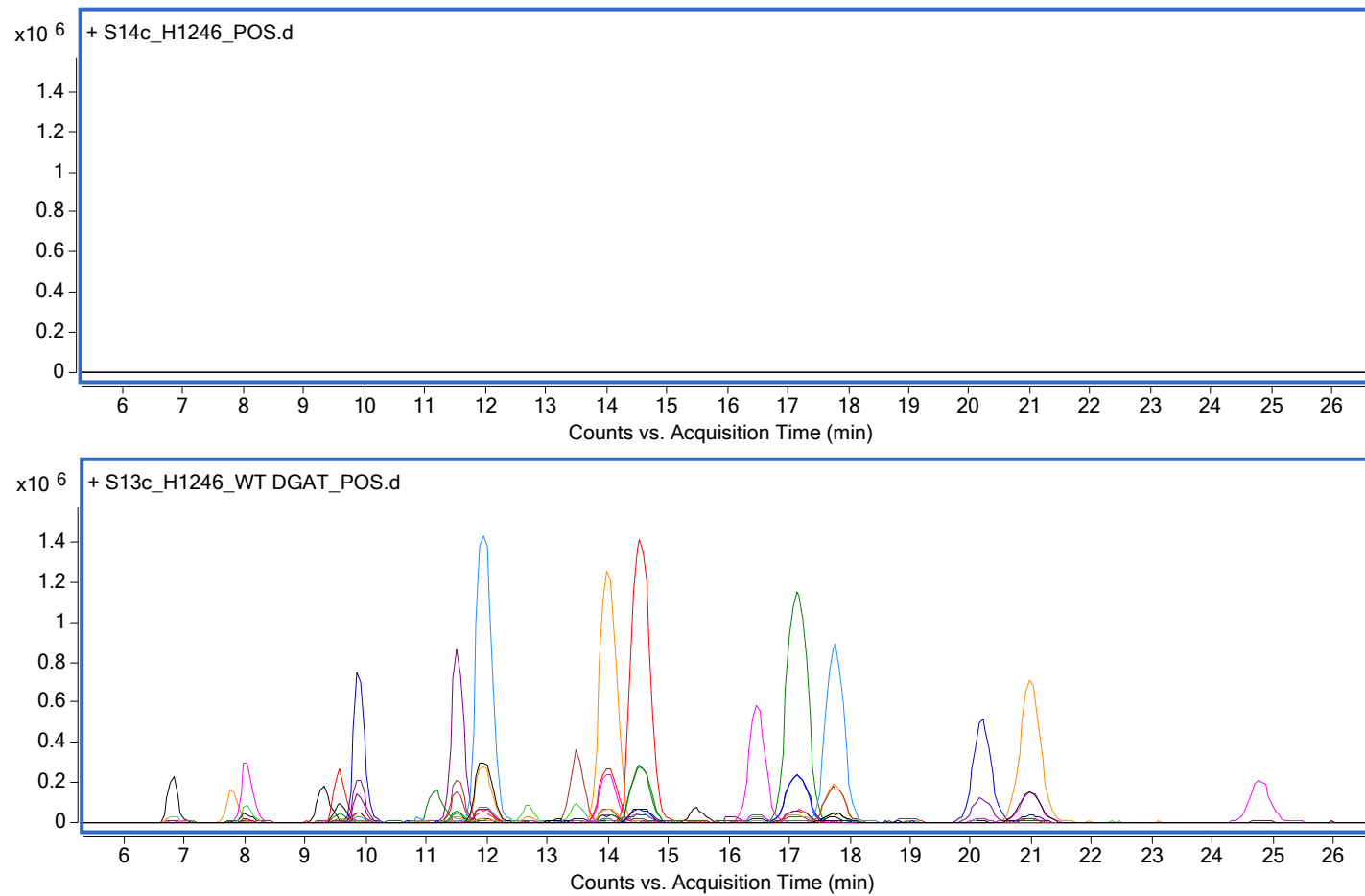


Figure S2: Overlay of extracted ion chromatograms of triacylglycerols in H1246 strain with empty plasmid (Top) and H1246 strain expressing wild type DGAT (Bottom). This validates that TAG is not produced in H1246 base strain with empty plasmid.

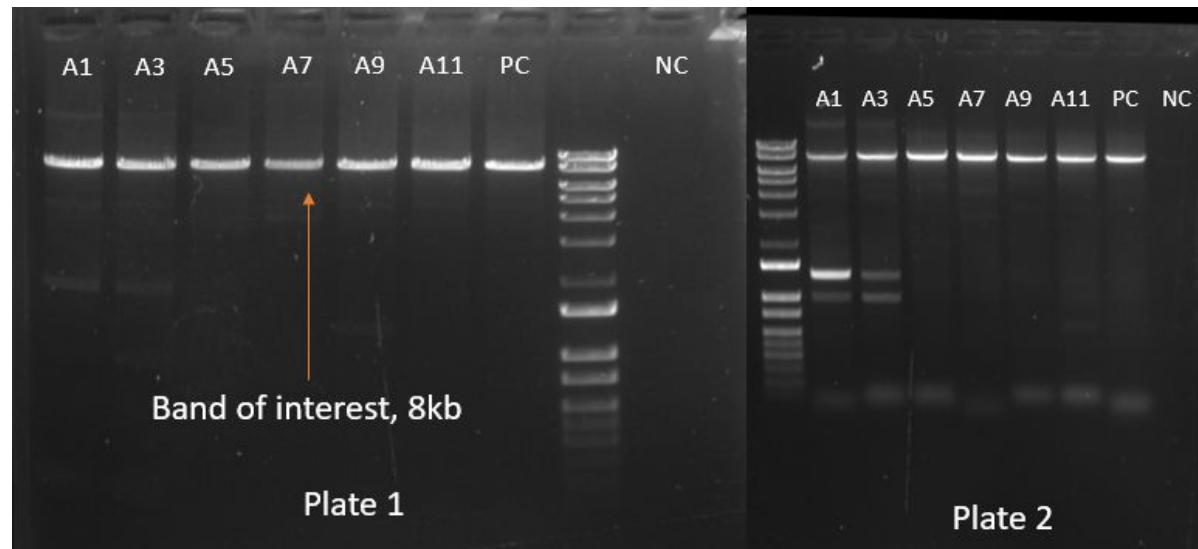


Figure S3: RF cloning random gel electrophoresis check from 190 constructs (2 plates). At least one sample per residue was analysed post-PCR to assess amplification success and detect potential artifacts, such as primer dimers

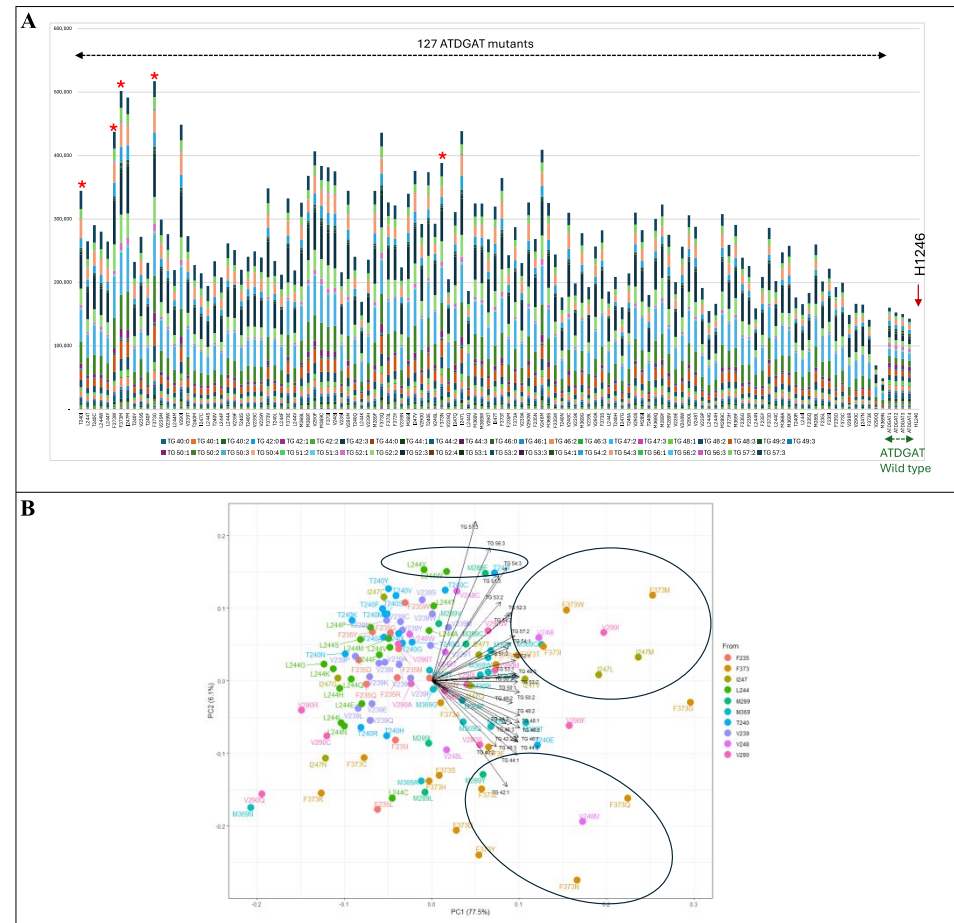


Figure S4: (A) TAG profile analysis for 127 mutants and wild type DGAT. **(B)** PCA analysis plot of TAG profile data of 127 mutants. The mutants with distinctive profile as per the PCA analysis has been circled and identified for large scale growth and lipid analysis

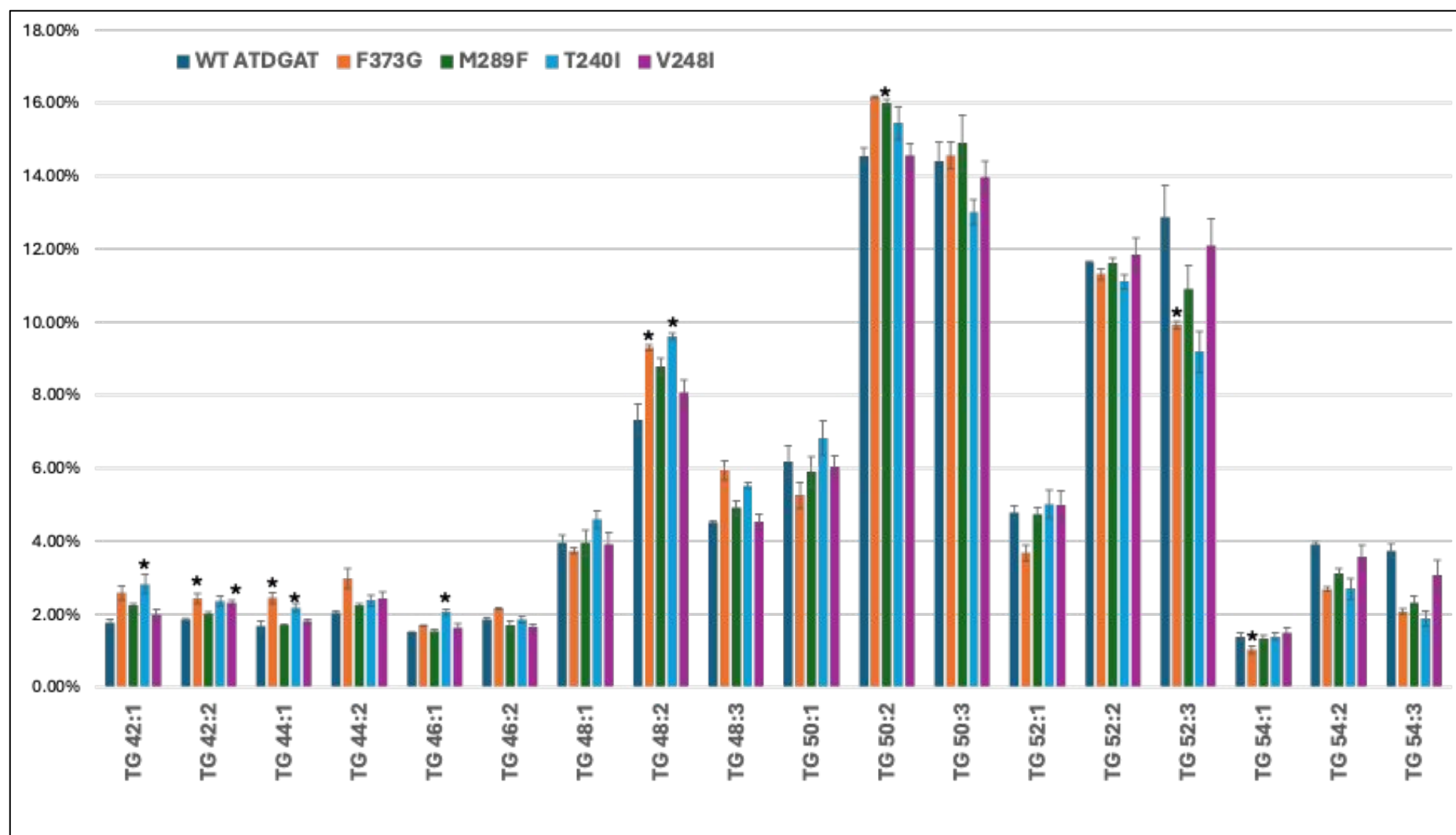


Figure S5: Complete TAG profile of 4 DGAT mutants F373G, M289F, T240I and V248I in comparison to the wild type DGAT expressing strain. * indicates changes with *p-value* < 0.05.

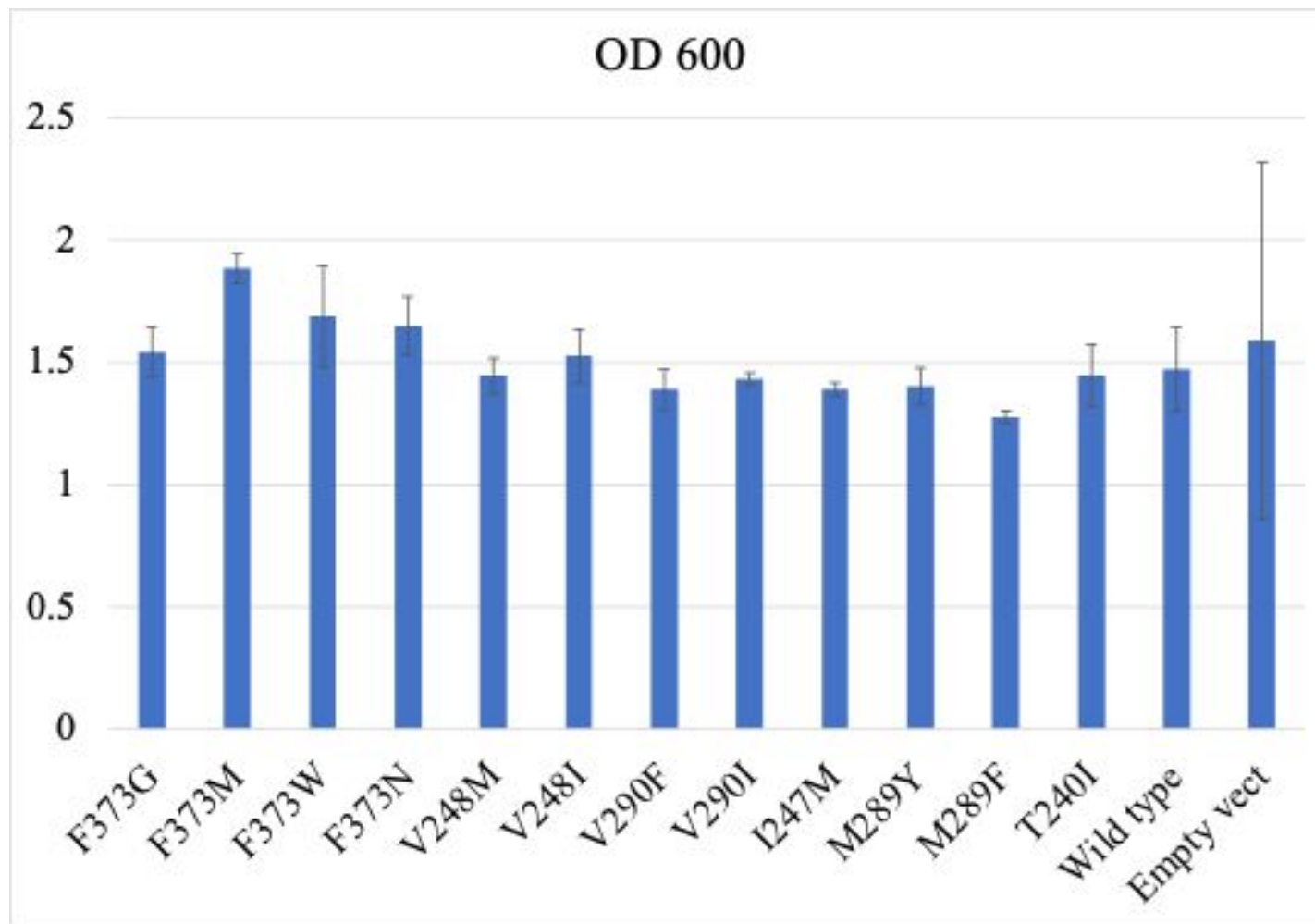


Figure S6: Growth comparison for 12 mutants, wild type and the empty vector control. OD at 600nm was measured for 2ml cultures grown for 48 hours with a starting OD of 0.2.

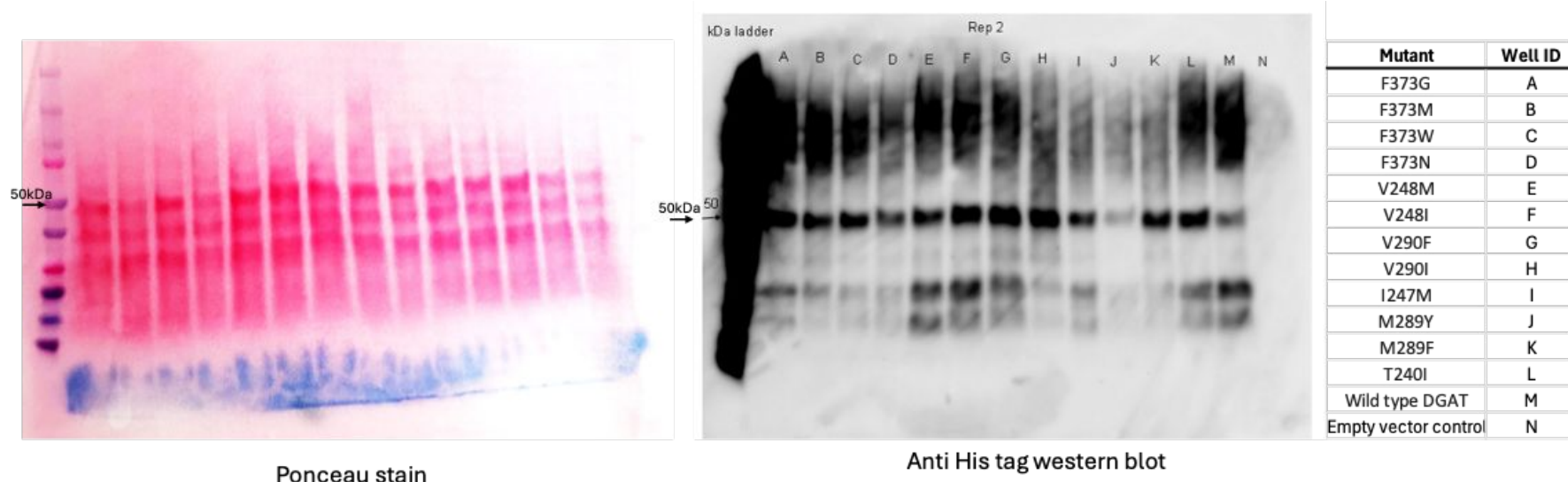


Figure S7: Comparison of protein expression for 12 mutants, wild type and the empty vector control in the insoluble fraction of cell lysate. The left hand side image shows the ponceau stain while the right side shows the Anti-His Tag western blot image. Protein was extracted from 2ml cultures grown for 48 hours with a starting OD of 0.2. The cell lysate was centrifuged to separate the soluble and insoluble fractions. SDS PAGE samples were prepared for all soluble and insoluble samples. The gel shown in this figure is of the insoluble fraction samples for all strains. The samples are loaded as follows: F373G:**A**, F373M:**B**, F373W:**C**, F373N:**D**, V248M:**E**, V248I:**F**, V290F:**G**, V290I:**H**, I247M:**I**, M289Y:**J**, M289F:**K**, T240I:**L**, Wild type DGAT:**M**, Empty vector control:**N**.