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Blazeck et al.

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(54) **COMPOSITIONS AND METHODS FOR LIPID PRODUCTION**(71) Applicant: **Board of Regents, The University of Texas System**, Austin, TX (US)(72) Inventors: **John Blazeck**, Austin, TX (US); **Andrew Hill**, Austin, TX (US); **Leqian Liu**, Austin, TX (US); **Hal Alper**, Austin, TX (US)(73) Assignee: **Board of Regents, The University of Texas System**, Austin, TX (US)

(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 241 days.

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(51) **Int. Cl.**

| | |
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| C12N 15/81 | (2006.01) |
| C07K 14/39 | (2006.01) |
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| C12N 9/02 | (2006.01) |

(Continued)

(52) **U.S. Cl.**

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| CPC | C12N 15/815 (2013.01); C07K 14/39 (2013.01); C12N 1/16 (2013.01); C12N 9/0006 (2013.01); C12N 9/0008 (2013.01); C12N 9/0042 (2013.01); C12N 9/0077 |
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(2013.01); **C12N 9/1007** (2013.01); **C12N 9/78** (2013.01); **C12N 9/88** (2013.01); **C12N 9/93** (2013.01); **C12N 15/01** (2013.01); **C12N 15/52** (2013.01); **C12P 7/64** (2013.01); **C12P 7/6409** (2013.01); **C12P 7/6463** (2013.01); **C12R 1/645** (2013.01)(58) **Field of Classification Search**

None

See application file for complete search history.

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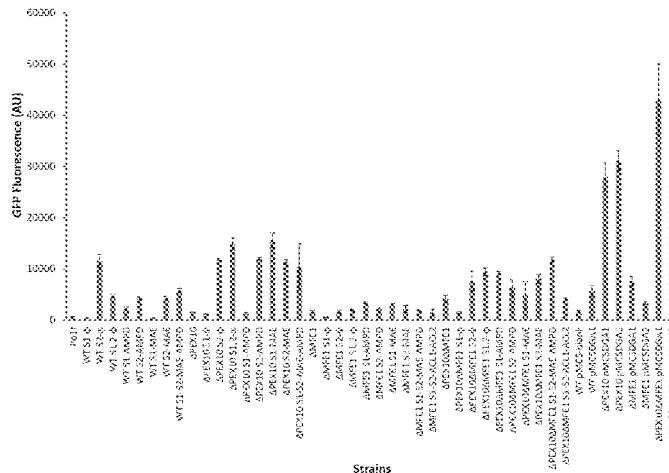
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(57) **ABSTRACT**

Described herein, inter alia, are compositions, oleaginous organisms, and methods useful for producing lipids, lipid precursors, and/or oleochemicals.

10 Claims, 23 Drawing Sheets



- (51) **Int. Cl.**
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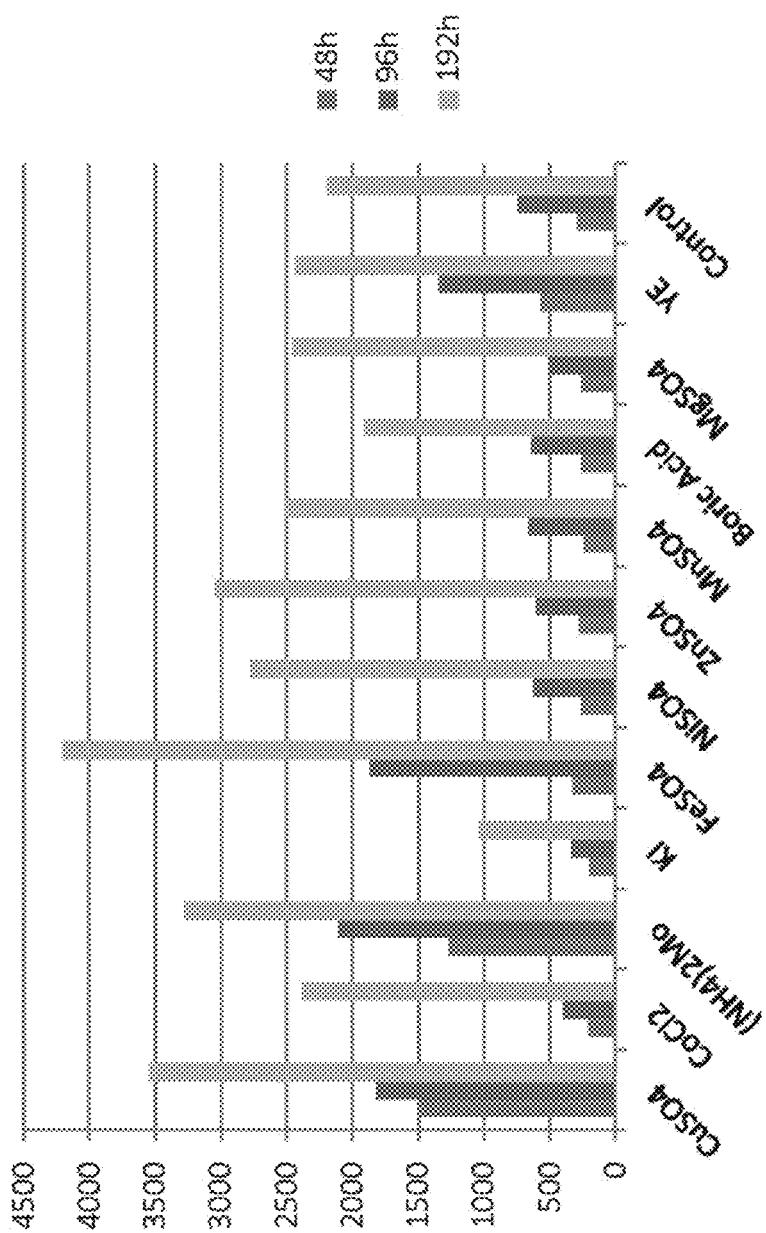


FIG. 1

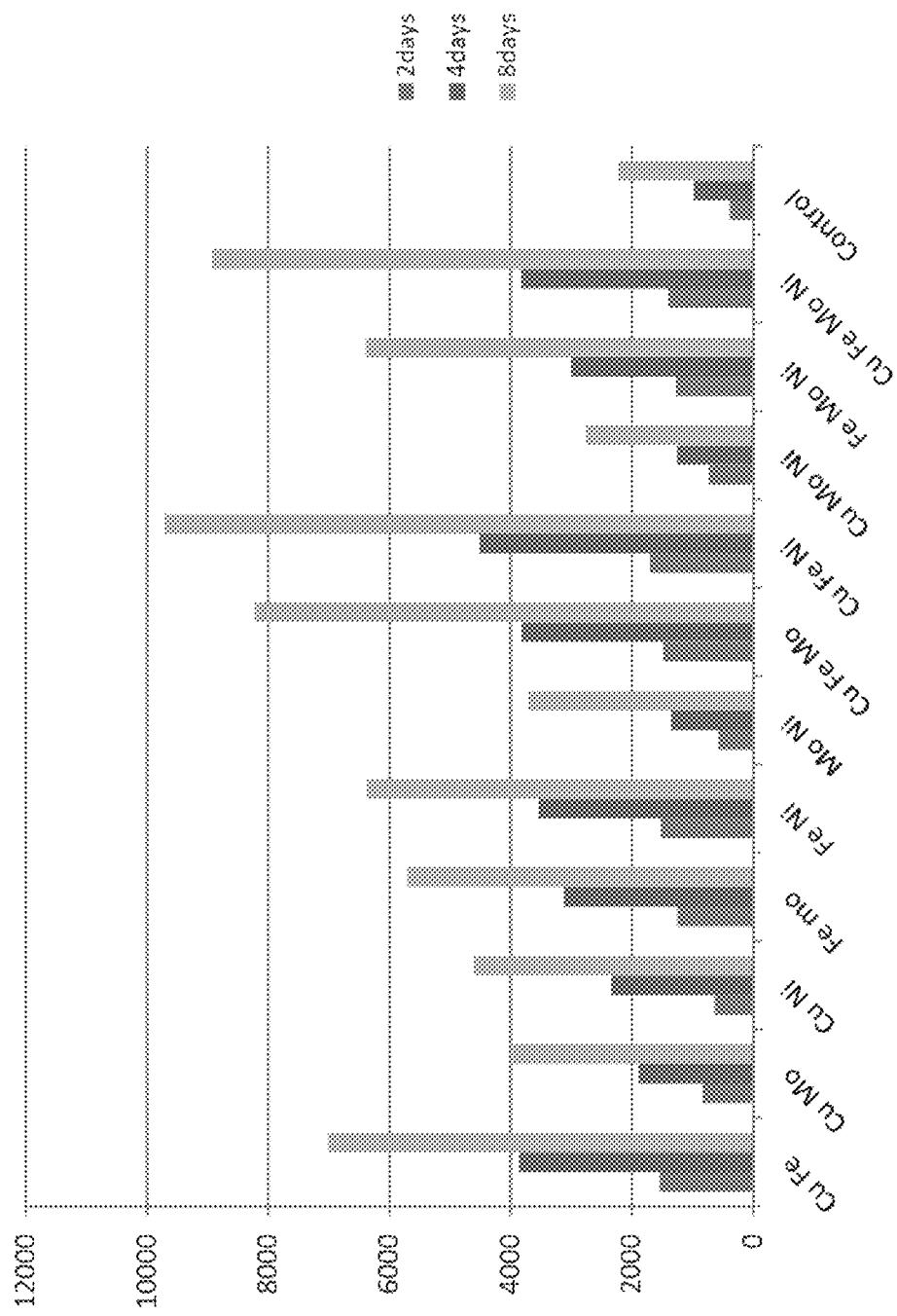
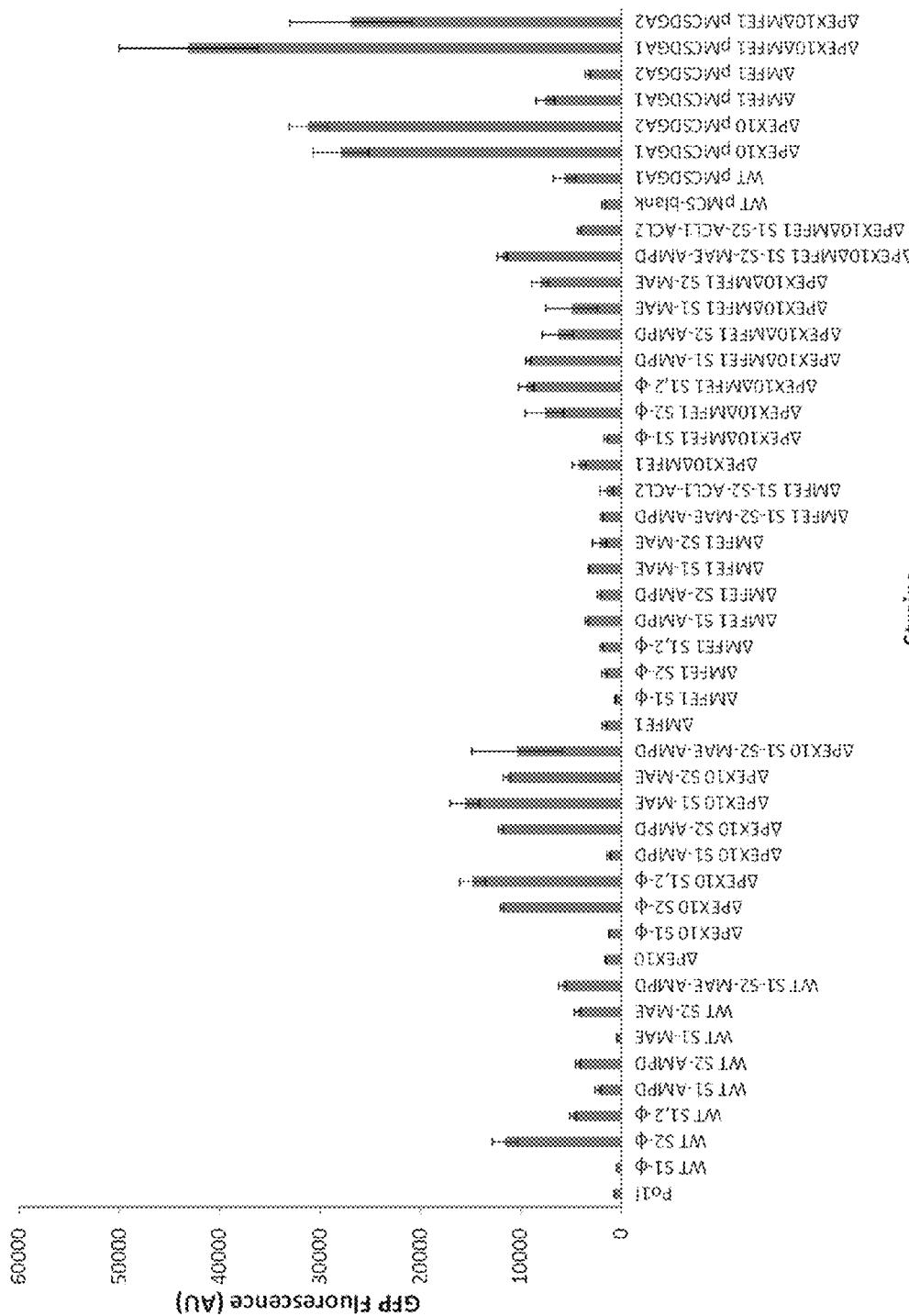


FIG. 2



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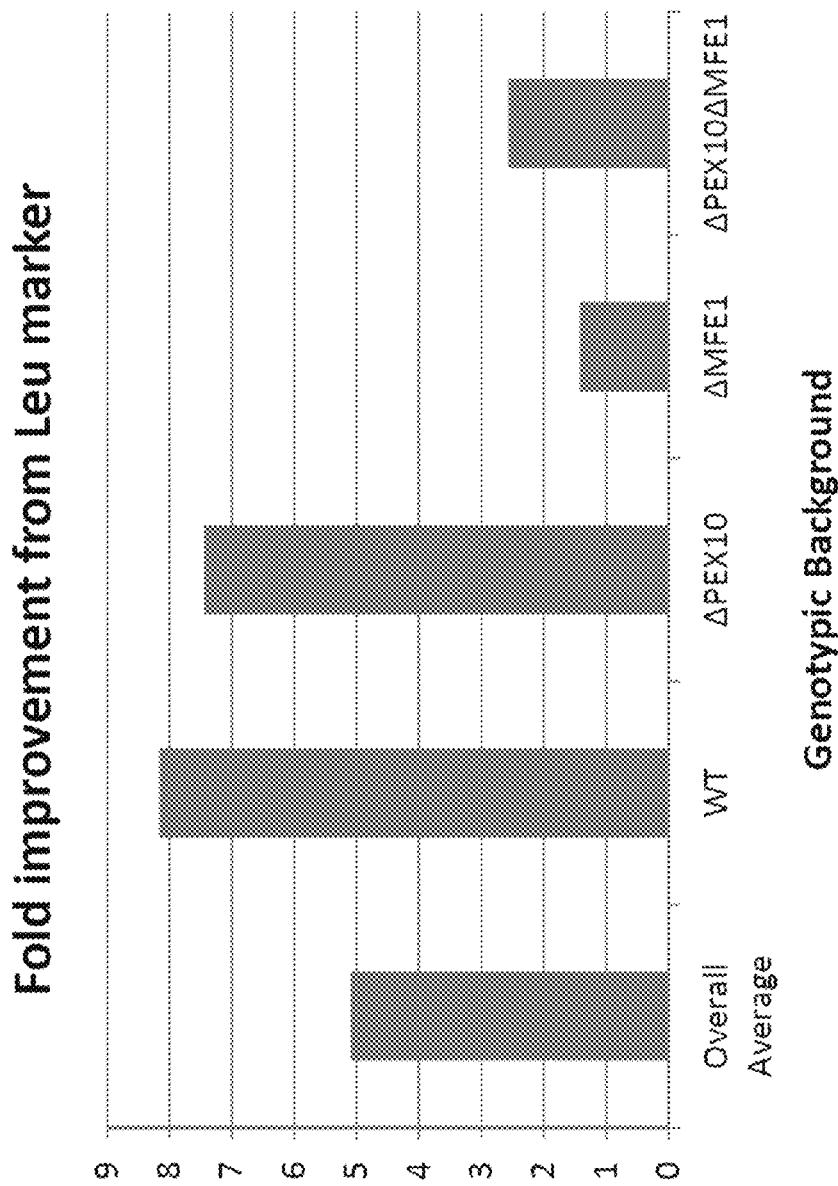


FIG. 4

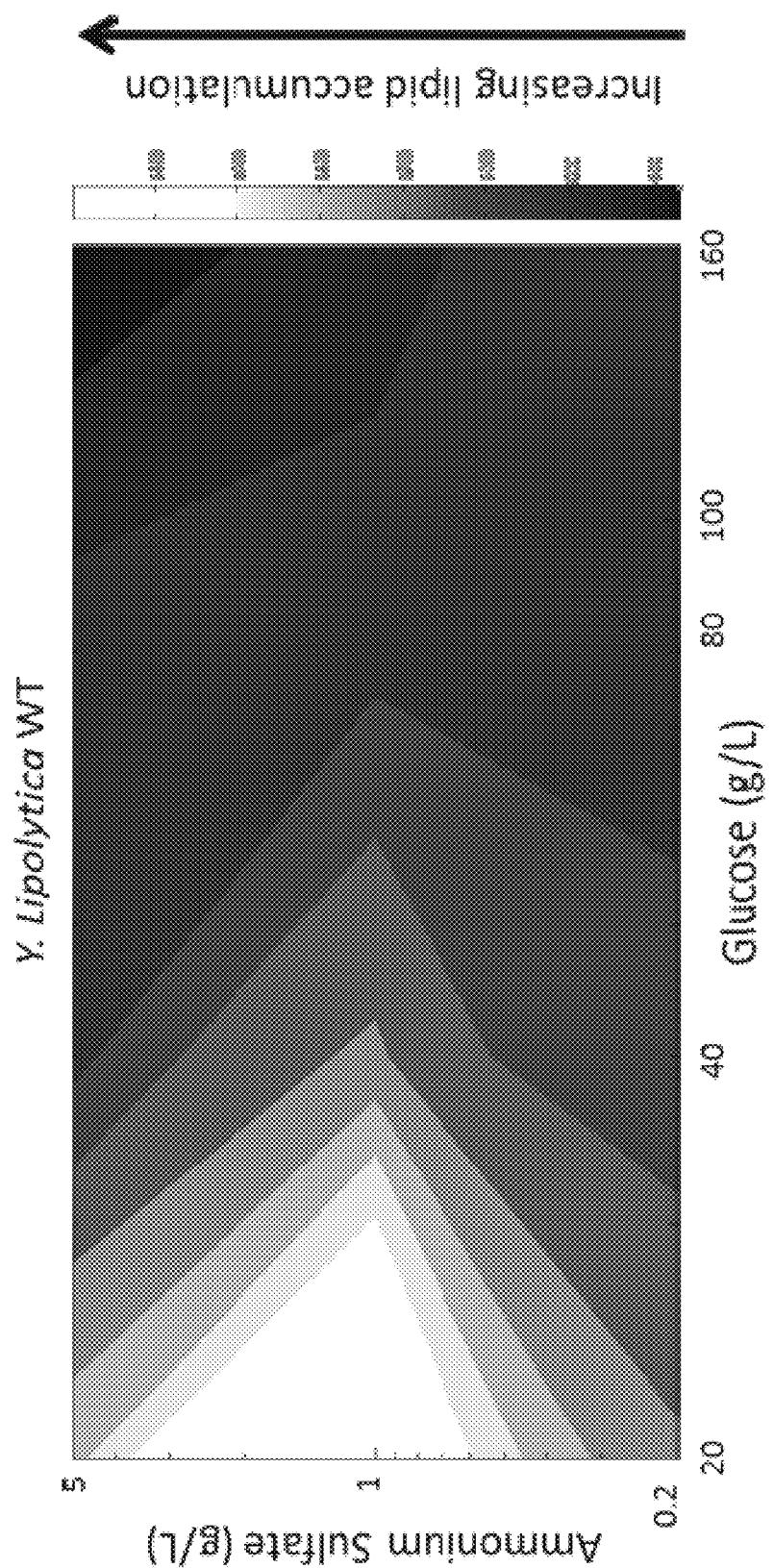


FIG. 5

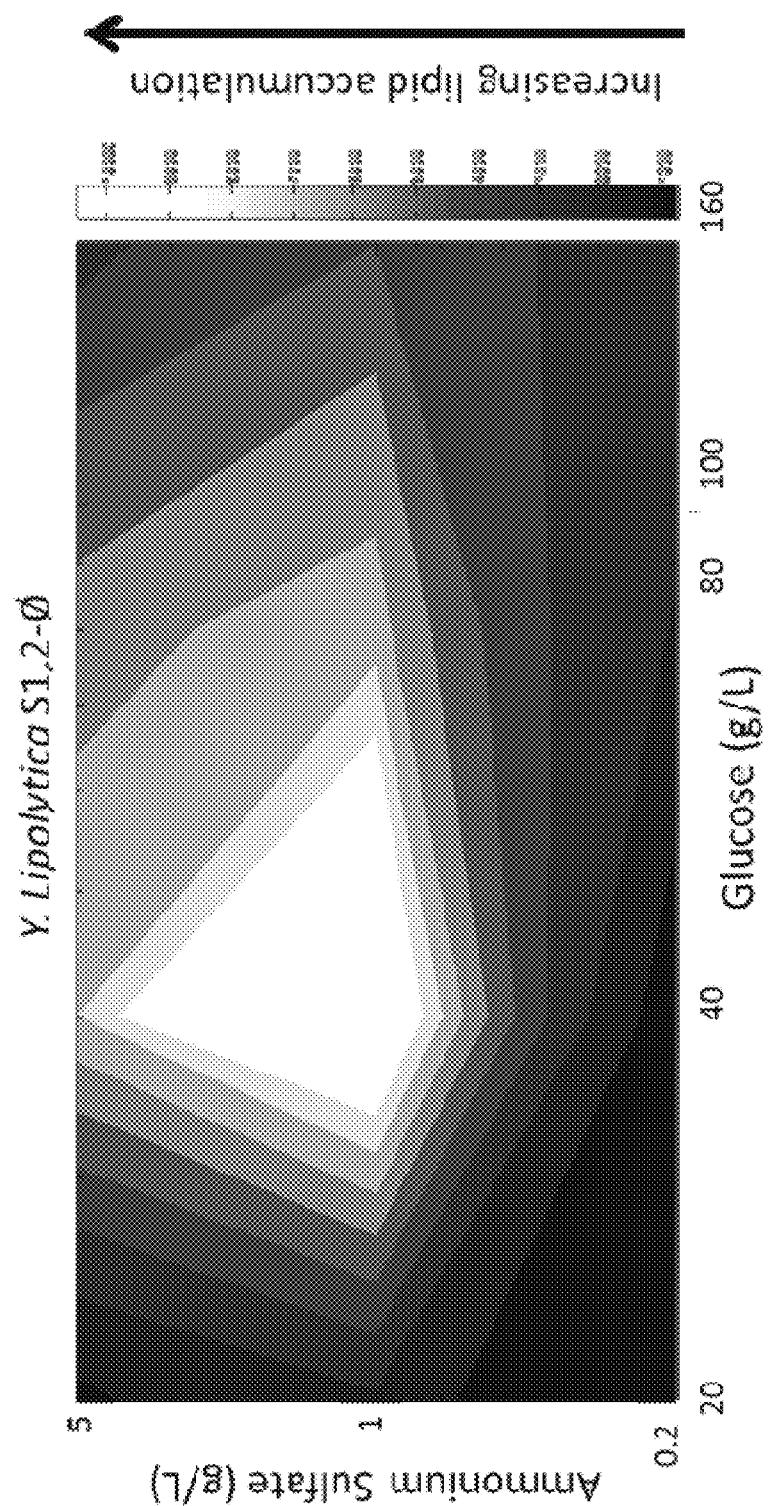


FIG. 6

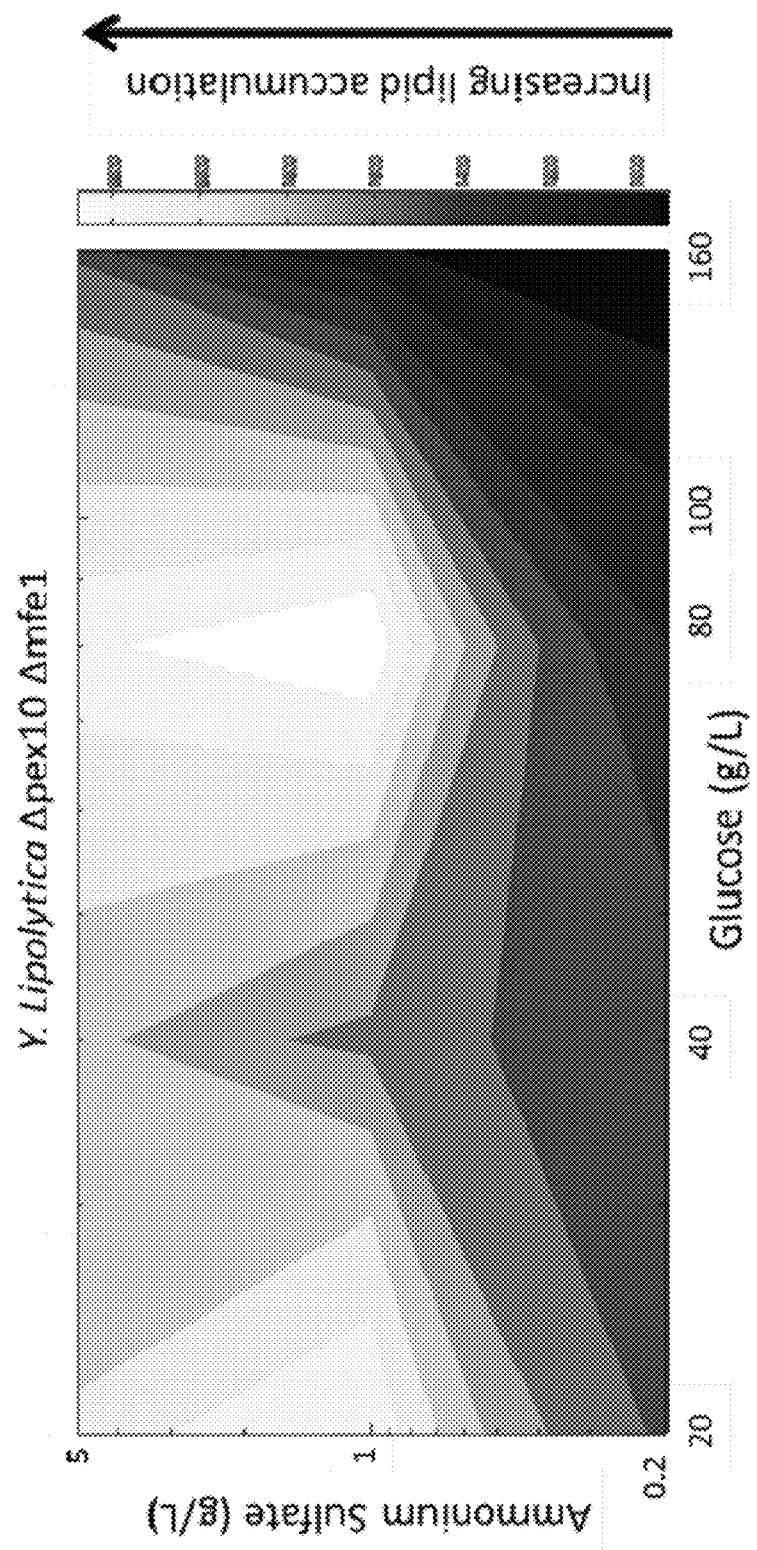


FIG. 7

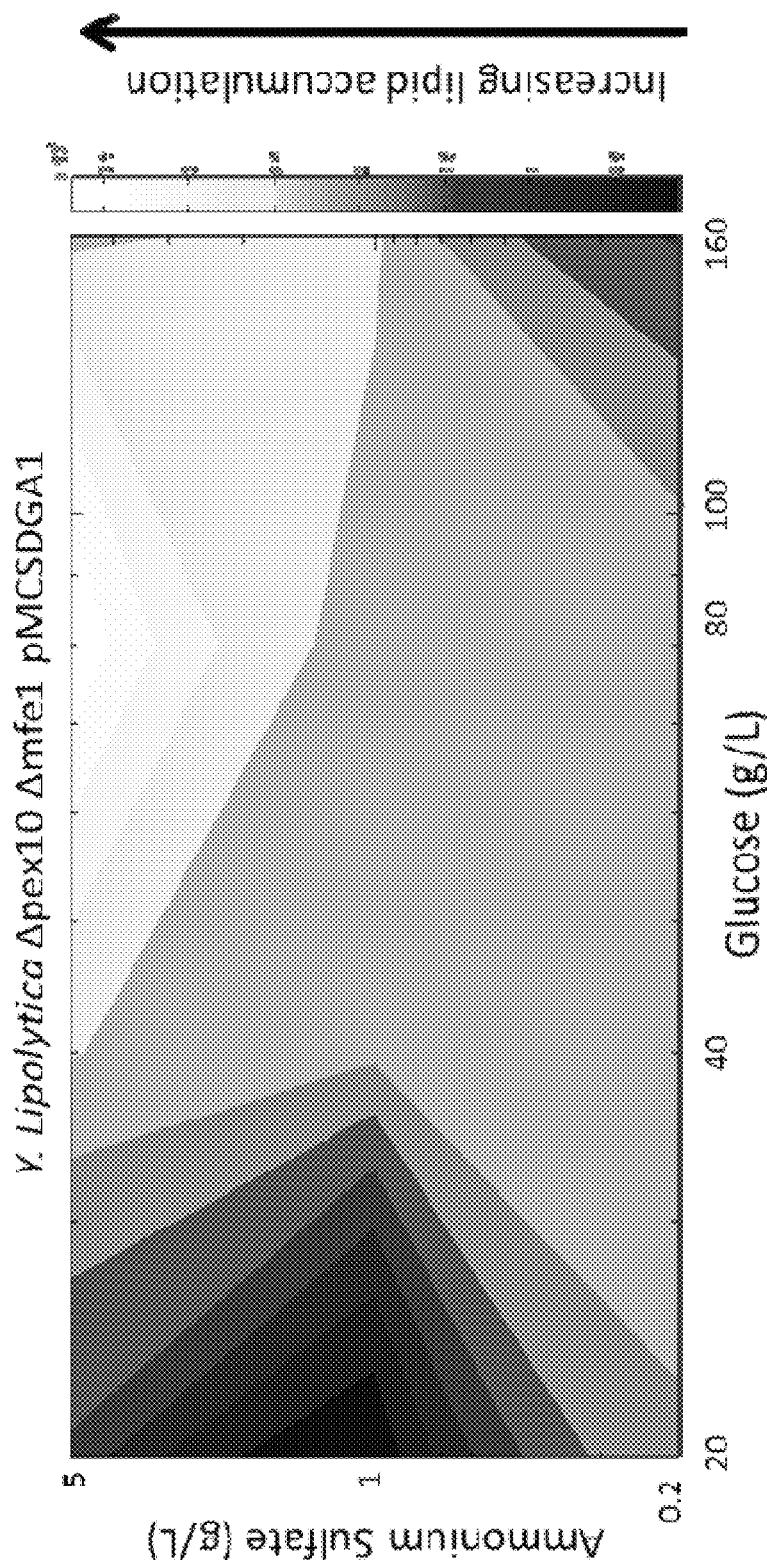


FIG. 8

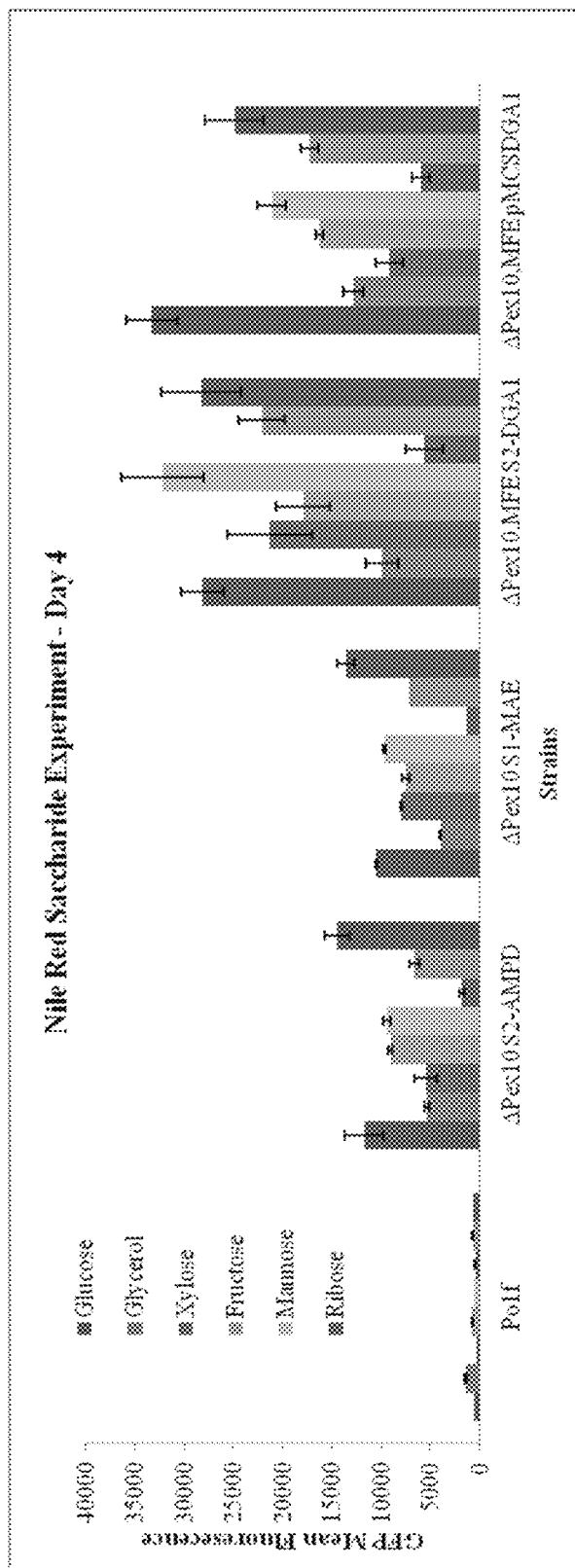


FIG. 9

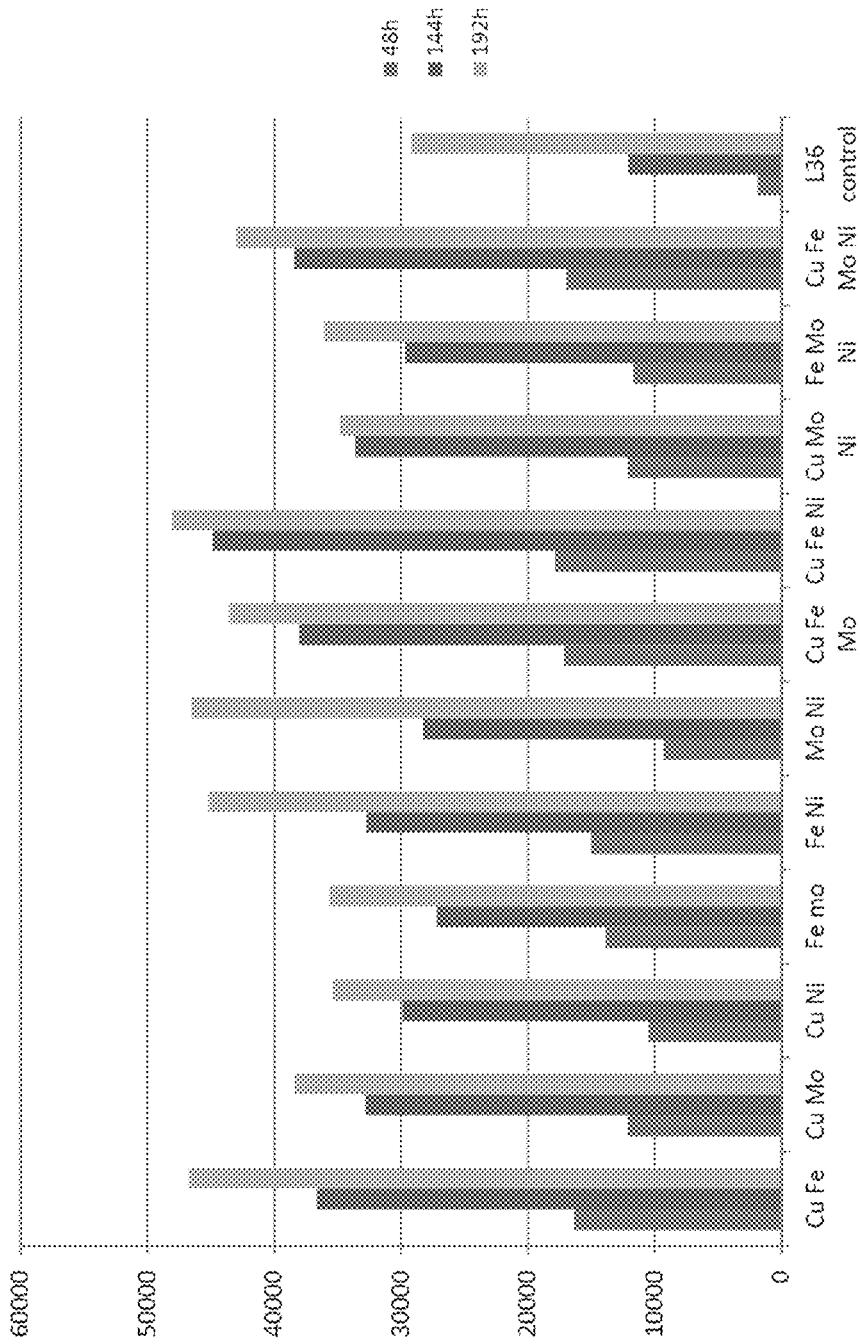
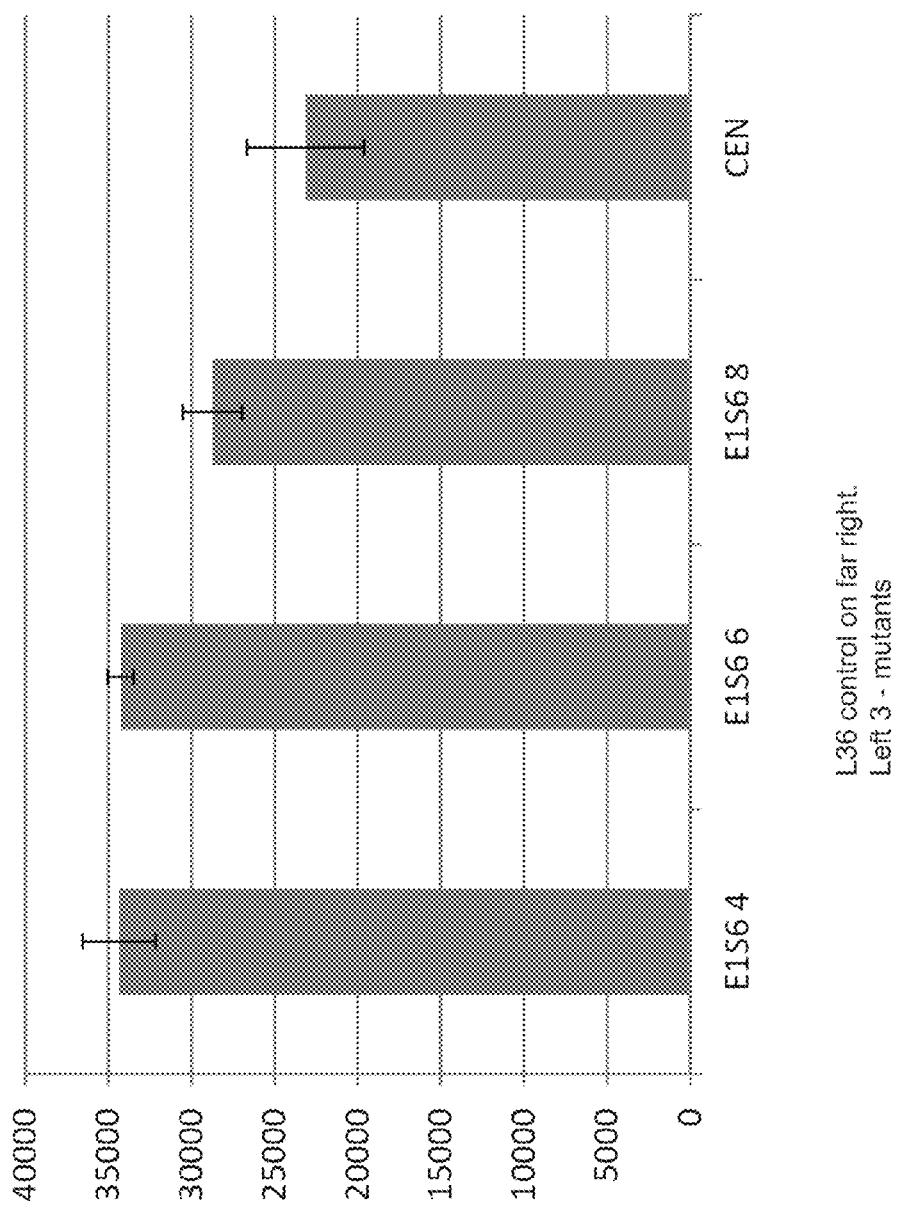


FIG. 10



L36 control on far right.
Left 3 - mutants

FIG. 11

Lipid visualization with FLM

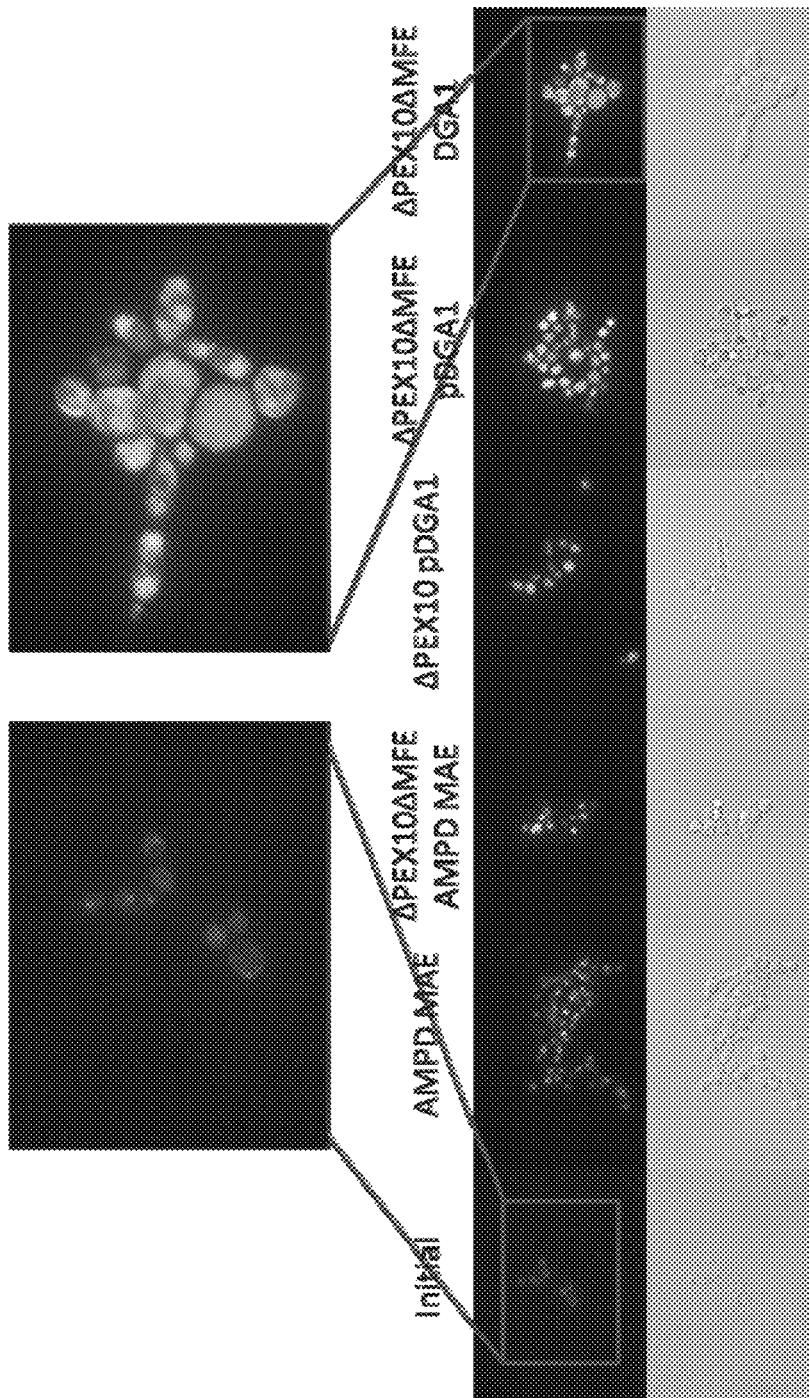


FIG. 12

Lipid accumulation mechanism in *Yarrowia lipolytica*

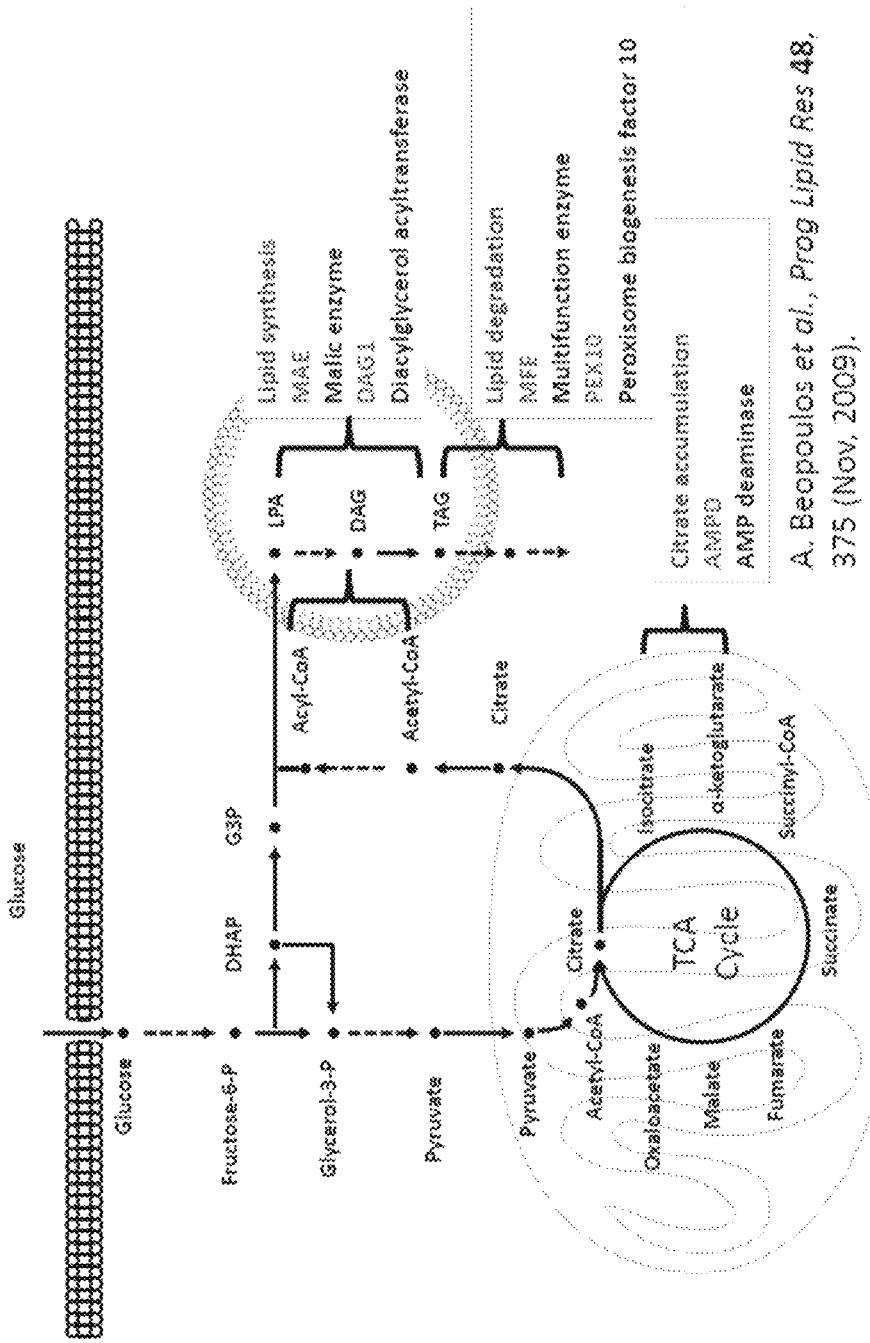


FIG. 13

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Isolation of "L36" mutant strain

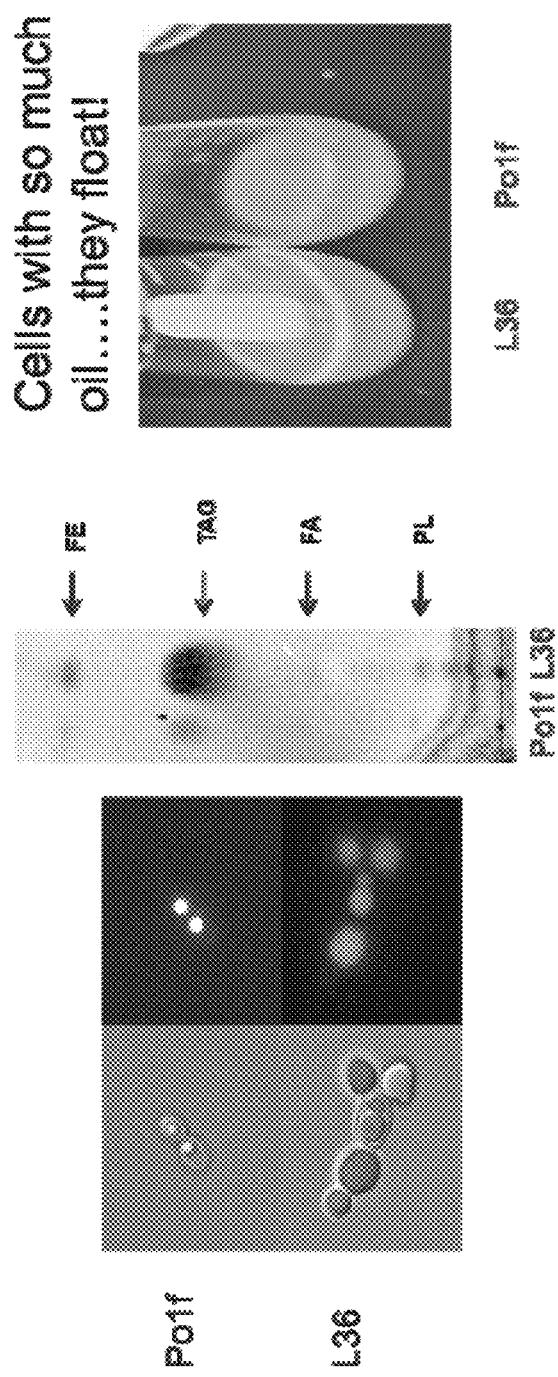


FIG. 14

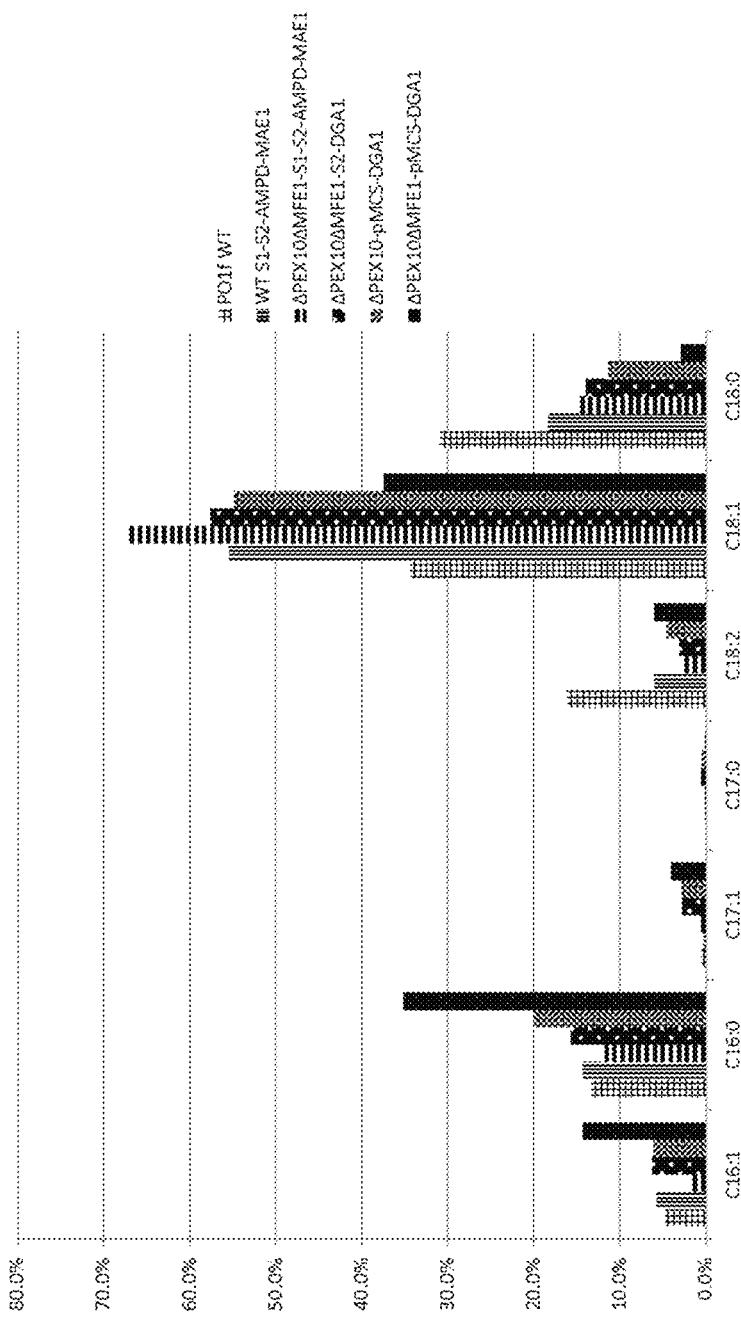


FIG. 15



FIG. 16

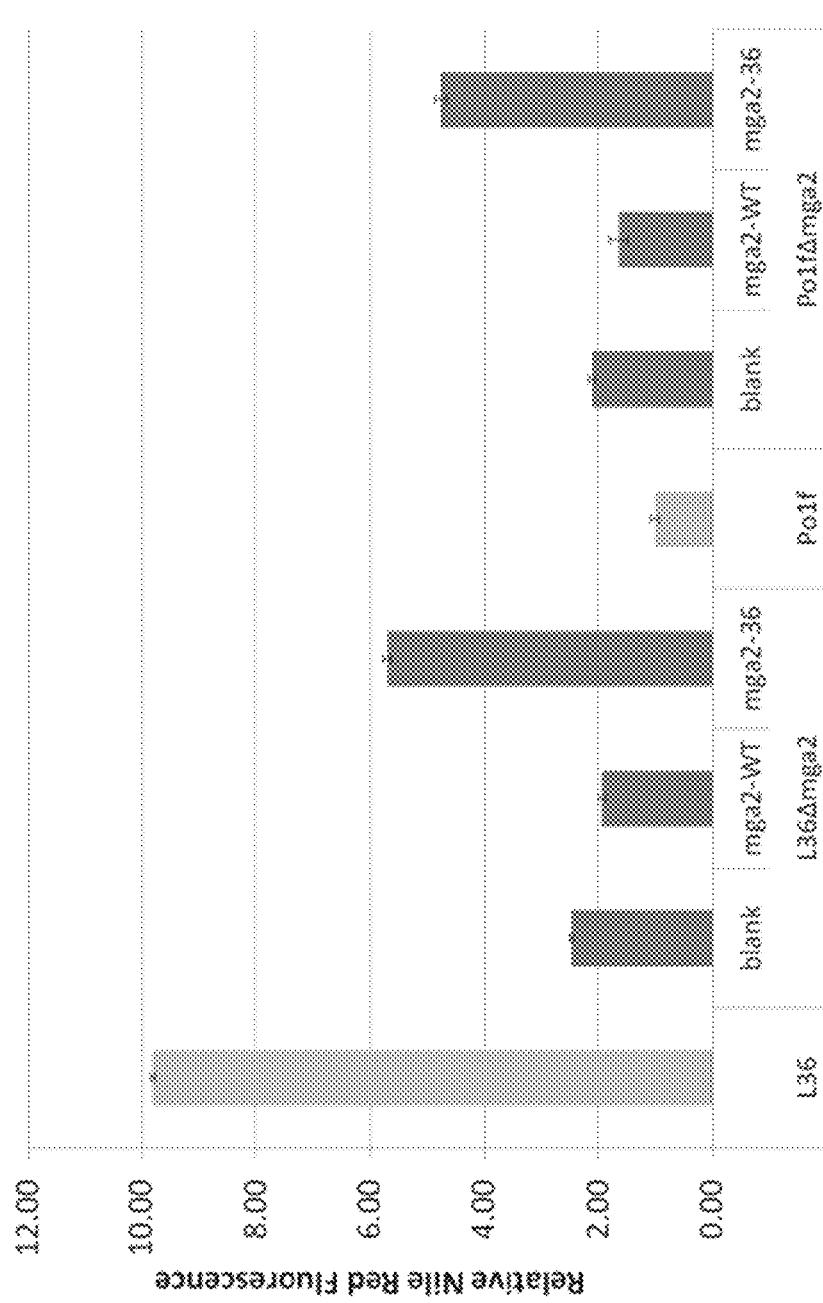


FIG. 17

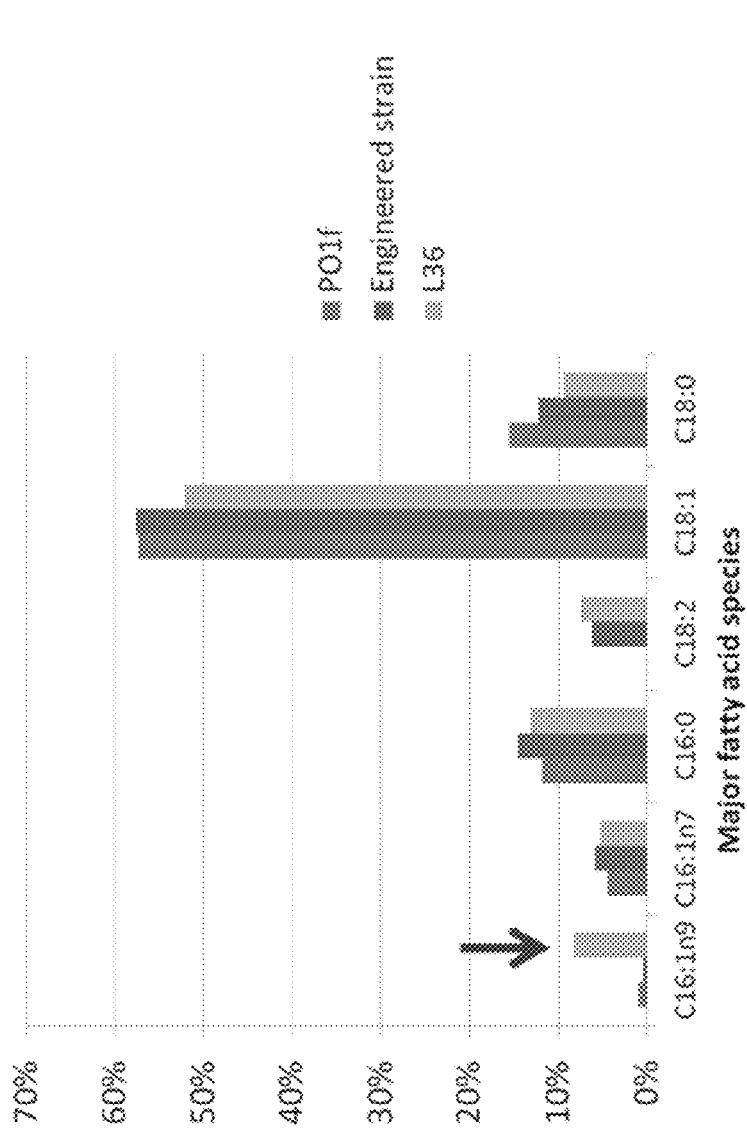


FIG. 18

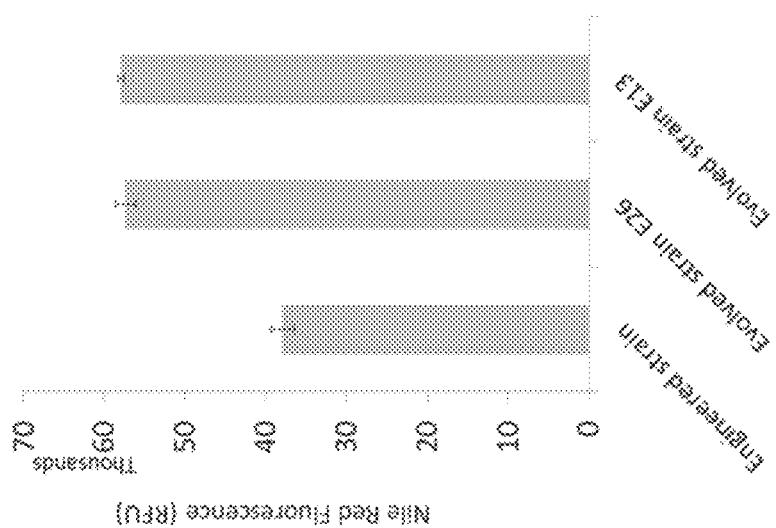


FIG. 19

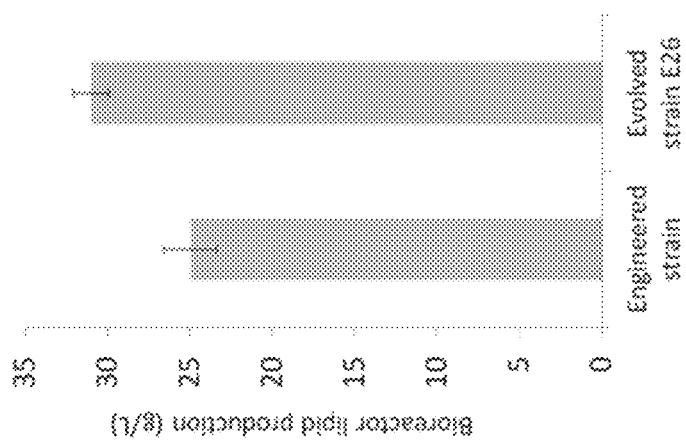


FIG. 20

| | | | | | |
|--------|----------|------|---|--------------|--|
| Yal10A | 297474 | G | A | YAL10A02354g | similar to <i>S. cerevisiae</i> CSM6; member of an oxygenated- binding protein family |
| Yal10A | 316425 | CGGA | C | YAL10A02497g | no similarity |
| Yal10C | 138994 | T | C | YAL10C01001g | no similarity |
| Yal10C | 139014 | A | G | YAL10C01001g | no similarity |
| Yal10C | 953493 | G | A | YAL10C07150g | similar to <i>S. cerevisiae</i> YCR081B; ubiquitous gene and putative Helicase |
| Yal10C | 29666661 | C | T | YAL10C22231g | weakly similar to Schizosaccharomyces pombe RPA subunit |
| Yal10C | 3047264 | G | A | YAL10C22726g | similar to <i>S. cerevisiae</i> transcription factor TFIIC subunit |
| Yal10D | 1576990 | G | A | YAL10D12628g | similar to <i>Fusarium solani</i> cutinase transcription factor 1 |
| Yal10E | 2038953 | G | A | YAL10E17215g | some similarity to <i>S. cerevisiae</i> PRM3 |
| Yal10E | 2038954 | G | A | YAL10E17215g | some similarity to <i>S. cerevisiae</i> PRM1 |
| Yal10E | 2424790 | T | G | YAL10E20449g | weakly similar to <i>S. cerevisiae</i> YOK1 |
| Yal10F | 3369592 | C | T | YAL10F26191g | similar to <i>S. cerevisiae</i> UGA2 |

FIG. 21

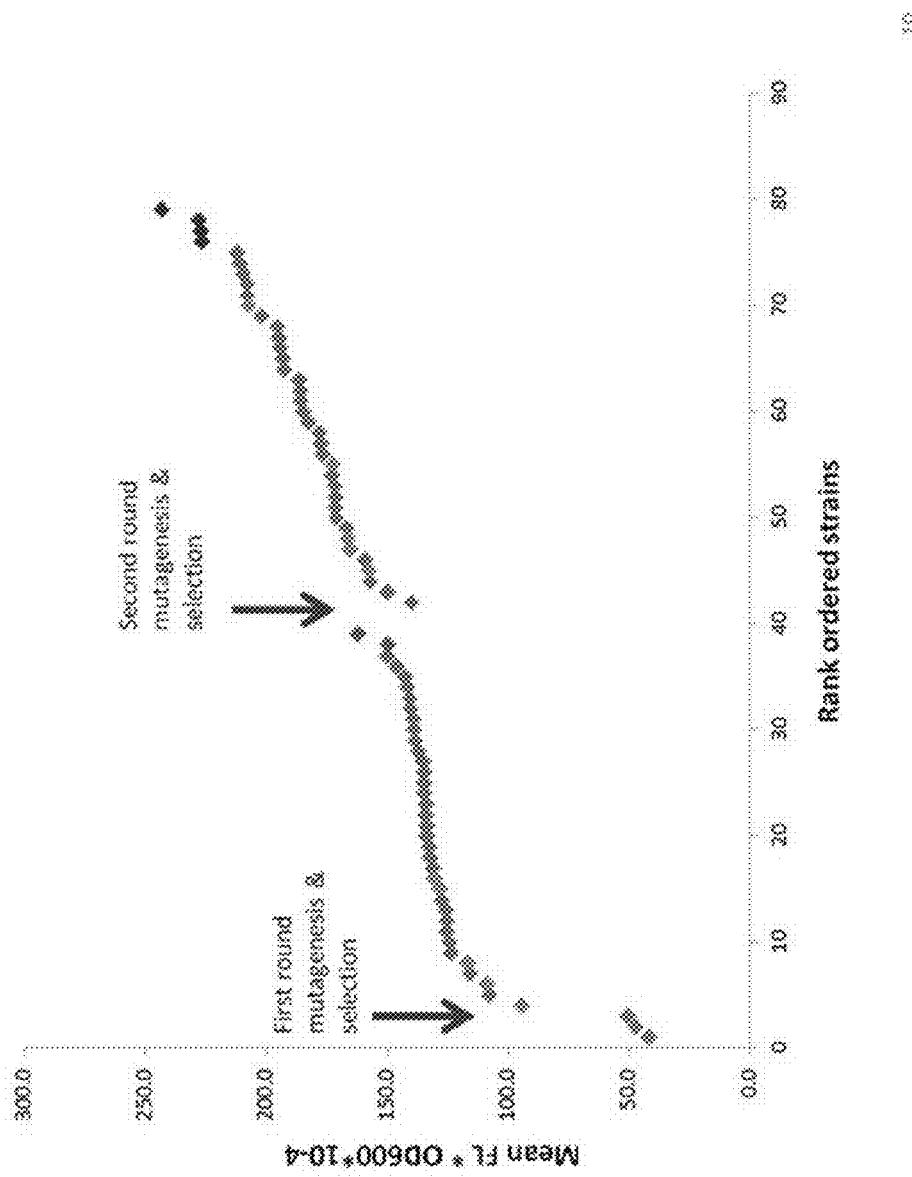


FIG. 22

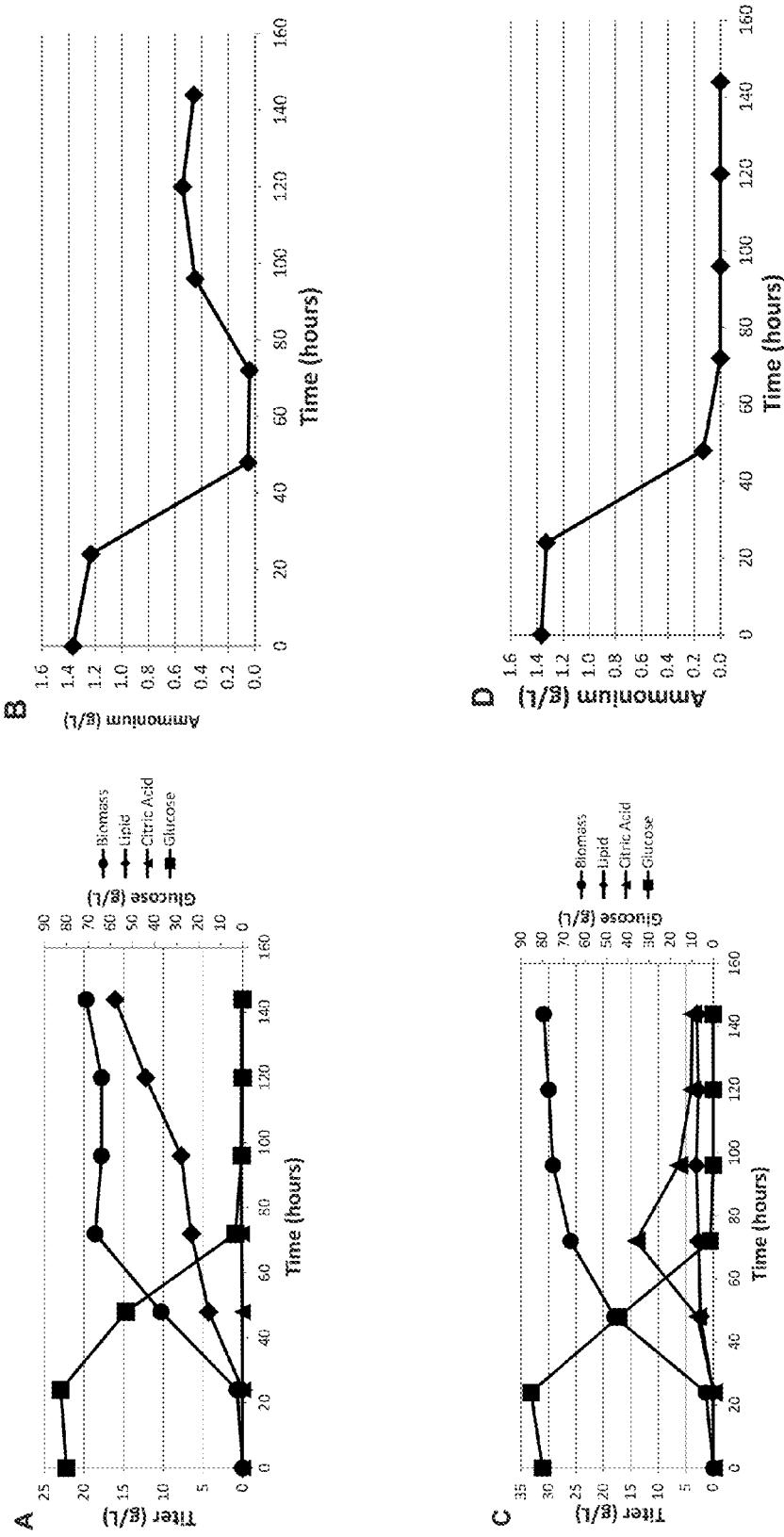


FIG. 23

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COMPOSITIONS AND METHODS FOR LIPID PRODUCTION

CROSS-REFERENCES TO RELATED APPLICATIONS

This application claims the benefit of U.S. Provisional Patent Application No. 61/819,476, filed May 3, 2013, which is incorporated herein by reference in its entirety and for all purposes.

STATEMENT AS TO RIGHTS TO INVENTIONS MADE UNDER FEDERALLY SPONSORED RESEARCH AND DEVELOPMENT

This invention was made with government support under N000141110669 awarded by Office of Naval Research. The government has certain rights in the invention.

REFERENCE TO A "SEQUENCE LISTING," A TABLE, OR A COMPUTER PROGRAM LISTING APPENDIX SUBMITTED ON A COMPACT DISK

The Sequence Listing written in file 93331-003510US-907029_ST25.TXT, created on Apr. 29, 2014, 210,560 bytes, machine format IBM-PC, MS-Windows operating system, is hereby incorporated herein by reference in its entirety and for all purposes.

BACKGROUND

Increasing oil consumption makes continued dependence on petroleum reserves untenable. Microbial production of renewable alternatives can reduce petroleum footprints through the *in vivo* synthesis of ethanol, biodiesel, and industrial precursors (Curran et al. 2013; Elshahed 2010; Li et al. 2008; Xu et al. 2013; Yim et al. 2011). Economic viability is highly dependent upon microbial choice, and an ideal host efficiently generates high titers independent of fermentation condition, through native or imported biosynthetic metabolism (Alper and Stephanopoulos 2009). In this regard, *Yarrowia lipolytica*'s genetic tractability, efficient utilization of many energy sources, and native capacity to accumulate lipids make it an ideal platform for oleo-chemical synthesis (Barth and Gaillardin 1996; Beopoulos et al. 2009a; Papanikolaou and Aggelis 2002).

Here we have employed a large-scale combinatorial approach to maximize lipid production in *Y. lipolytica* through both genomic engineering and combinatorial and inverse metabolic engineering multiplexed with phenotypic induction.

Y. lipolytica has a fully defined metabolic engineering toolbox that enables intracellular flux control through genomic manipulation (Blazeck et al. 2013b; Dujon et al. 2004; Fickers et al. 2003; Juretzek et al. 2001; Matsuoka et al. 1993). *Y. lipolytica* is commonly utilized for heterologous protein excretion and to examine and manipulate lipid and fatty acid metabolism (Beopoulos et al. 2009b; Beopoulos et al. 2008; Dulermo and Nicaud 2011; Madzak et al. 2004; Thevenieau et al. 2009), and has proven amenable to downstream manipulation of its fatty acid content to alter desaturation levels (Chuang et al. 2010) or to synthesize novel oleo-chemicals (Blazeck et al. 2013a). Thus, *Y. lipolytica* lipid reserves are ideal for *in vivo* catalysis to alkanes (Schirmer et al. 2010), fatty acid esters (Shi et al. 2012) or for standard transesterification-based conversion and use as

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biodiesel. In particular, biodiesel production grants a high net energy gain compared to other alternative fuels with minimal environmental impact, and harvesting lipid reserves from a microbial source such as *Y. lipolytica* enables easily scaled-up production without compromising food supply (Christophe et al. 2012; Hill et al. 2006; Kirstine and Galbally 2012; Subramaniam et al. 2010). *Y. lipolytica*'s natural lipid content consists of predominantly C16:0, C16:1, C18:0, C18:1, and C18:2 fatty acids (Beopoulos et al. 2008; Blazeck et al. 2013a; Tai and Stephanopoulos 2013), very similar to the fatty acid content of biodiesel derived from soybeans and rapeseed (Gruzdienė and Anelauskaitė 2011; Hammond et al. 2005). Economic viability can be greatly improved by fully utilizing all sugars from lignocellulosic biomass or by using carbon from industrial waste streams. In this regard, *Y. lipolytica* can efficiently utilize hydrophobic and waste carbon sources, such as crude glycerol (Andre et al. 2009; Fickers et al. 2005; Makri et al. 2010; Rywinska et al. 2013), and has shown excellent heterologous gene expression when utilizing glucose, sucrose, glycerol, or oleic acid as a carbon source (Blazeck et al. 2013b). Finally, *Y. lipolytica* is regarded as a "safe-to-use" organism (Groenewald et al. 2013).

Lipid accumulation in *Y. lipolytica* can be induced by nitrogen starvation and has been associated with the activity of four enzymes: AMP Deaminase (AMPDp), ATP-Citrate Lyase (ACLp), Malic Enzyme (MAEp) and Acetyl-CoA Carboxylase (ACCP) (Beopoulos et al. 2009a; Dulermo and Nicaud 2011). AMPDp cleaves AMP into NH₄⁺ and inosine 5'-monophosphate to replenish intracellular nitrogen levels; AMP deficiency inhibits the citric acid cycle resulting in citric acid accumulation. ACLp cleaves citric acid into oxaloacetate and acetyl-CoA, and ACCP carboxylates acetyl-CoA into malonyl-CoA fatty acid building blocks. Fatty acid synthesis is further encouraged by a MEAp-mediated increase in NADPH levels (Beopoulos et al. 2009a). Fatty acids can be directly stored in intracellular lipid bodies or further incorporated in triacylglycerides before storage (Beopoulos et al. 2008). Triacylglyceride synthesis follows the Kennedy Pathway to fuse three fatty acids to a glycerol-3-phosphate (G3P) backbone (Kennedy 1961). The ultimate step is catalyzed by the DGA1 or DGA2 acyl-CoA:diacylglycerol acyltransferases (Beopoulos et al. 2009a; Beopoulos et al. 2012). G3P backbone is synthesized from dihydroxyacetone phosphate (DHAP) by the cytosolic, NAD⁺-dependent glycerol-3-phosphate dehydrogenase (GPD1) and recycled into glycolysis by the mitochondrial, FAD⁺-dependent glycerol-3-phosphate dehydrogenase isoform (GUT2) (Dulermo and Nicaud 2011). TAG hydrolysis mobilizes free fatty acids for peroxisomal degradation through the four step β-oxidation cycle (Beopoulos et al. 2011)—oxidation by one of six acyl-CoA oxidases (POX1-6), hydration and dehydrogenation by the multifunctional enzyme (MFE1), and thiolysis by a 3-ketoacyl-CoA-thiolase (POT1 or PAT1) (Beopoulos et al. 2009a). The PEX10p transcription factor has been implicated in peroxisomal biogenesis and ΔpeX10 mutants display increased triacylglyceride content (Blazeck et al. 2013a; Hong et al. 2012; Zhu et al. 2012).

Genomic modifications to *Y. lipolytica*'s fatty acid, lipid, and central carbon metabolism have shown promise towards increasing lipid accumulation capacity. Deletion of the six POX genes increased *ex novo* incorporation of oleic acid in *Y. lipolytica*, while deletion of the single MFE1 gene had a similar effect (Beopoulos et al. 2008; Dulermo and Nicaud 2011). Increasing G3P backbone levels by combining GUT2p deletion and GPD1p overexpression in these β-oxi-

dation deficient backgrounds further increased ex novo lipid accumulation to 65-75% triacylglyceride content (Dulermo and Nicaud 2011). Overexpression of DGA1p increased de novo triacylglyceride accumulation fourfold over control levels to 33.8% triacylglyceride content, and co-overexpression of ACC1p further increased triacylglyceride accumulation to a final yield of 41% triacylglyceride content (Tai and Stephanopoulos 2013). To date, no study has attempted to combine the beneficial effects of engineering *Y. lipolytica*'s fatty acid, lipid and central metabolism in a single strain. Additionally, *Y. lipolytica*'s dependence on media formulation for lipid accumulation has not been adequately explored, nor has its ability to randomly accumulate mutations that enhance lipid accumulation. Furthermore, no attempt has been made to utilize mutation-based evolutionary selection to identify novel lipogenic genotypes. Thus, the ultimate capacity of *Y. lipolytica* to accumulate lipids and other oleochemicals has not been unlocked. To this end, we have employed a large scale combinatorial approach to maximize lipid production while accounting for unexpected interactions between genotype and environmentally-induced phenotype. The present invention provides solutions to these and other problems in the art.

BRIEF SUMMARY

In a first aspect is provided a genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) wherein the dry weight of said yeast cell includes greater than 60% wt/wt lipids, lipid precursors, and/or oleochemicals.

In a second aspect is provided a method of producing a lipid, lipid precursor, or oleochemical (e.g., lipid, lipid precursor, oleochemical) including: 1) culturing a yeast cell as described herein (including embodiments or as described in the examples, tables, figures, and/or claims) in a growth medium; and 2) isolating the lipid, lipid precursor, or oleochemical (e.g., lipid, lipid precursor, oleochemical) (e.g. from the medium or yeast cell).

In a third aspect is provided a method of isolating a genetically modified yeast cell from a plurality of yeast cells, including greater than 60% wt/wt lipids, lipid precursors, and/or oleochemicals in dry weight, including allowing a genetically modified yeast cell to separate from a population of yeast cells within the plurality of yeast cells by floating above the population of yeast cells within an aqueous medium thereby isolating the genetically modified yeast cell, wherein the population of yeast cells includes a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals than said genetically modified yeast cell.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1. Nile Red assay quantifying lipid content of PO1f WT strain in C160N0.2 media supplemented with individual micronutrients after 2, 4, and 8 days of cultivation.

FIG. 2. Nile Red assay quantifying lipid content of PO1f WT strain in C160N0.2 media supplemented with multiple micronutrients after 2, 4, and 8 days of cultivation.

FIG. 3. Nile Red assay quantify lipid content of 46 rationally constructed genetically modified PO1f derivatives.

FIG. 4. Fold improvement of lipid accumulation (from Nile Red assay signal (RFU)) by enabling the capacity to synthesis leucine through incorporation of the LEU2 marker to different genotypic background. LEU2 expression can be from an episomal or an integrated sequence.

FIG. 5. Heat map of lipid content based on Nile Red signal of PO1f WT cultured in media formulations with different carbon to nitrogen ratios after 4 days.

FIG. 6. Heat map of lipid content based on Nile Red signal of PO1f-S1-S2-φ cultured in media formulations with different carbon to nitrogen ratios after 4 days.

FIG. 7. Heat map of lipid content based on Nile Red signal of ΔPEX10ΔMFE1 cultured in media formulations with different carbon to nitrogen ratios after 4 days.

10 FIG. 8. Heat map of lipid content based on Nile Red signal of ΔPEX10ΔMFE1-pMCS-DGA1 cultured in media formulations with different carbon to nitrogen ratios after 4 days.

15 FIG. 9. Nile Red assay quantify lipid content on Day 4 with different strains growing on different saccharides as carbon sources. Saccharide initial concentration was set at 80 g/L with 5 g/L ammonium sulfate.

20 FIG. 10. Nile Red assay quantify lipid content of isolated L36 strain cultured in C160N0.2 media supplemented with multiple micronutrients after 2, 4, and 8 days of cultivation.

25 FIG. 11. Nile Red assay quantify lipid content with EMS mutagenesis in evolved L36 strains and L36.

FIG. 12. Fluorescence light microscopy pictures of lipid accumulation in selected strains. Lipids were stained with 25 Nile Red as usual. Strain ΔPEX10ΔMFE1-pMCS-DGA1 shows almost total lipid content while PO1f WT has very little.

FIG. 13. General lipid metabolism in yeast and a portion of selected targets to engineering lipid metabolism.

30 FIG. 14. The isolation and characterization of superior lipid production strain L36.

FIG. 15. Fatty acid profiles for different strains.

FIG. 16. Lipid accumulation in strain PO1f and PO1fΔaco1 DGA1 leu+ ura+ characterized with flow cytometry using cells stained with Nile Red on 48 hour and 96 hour time point. The starting OD of the culture is 2.5 and the cells were cultivated in yeast synthetic medium with 80 g/L glucose.

40 FIG. 17. Lipid accumulation characterized with flow cytometry using cells stained with Nile Red on 192 h time point. The starting OD of the culture is 5 and the cells were cultivated in yeast synthetic medium with 160 g/L glucose and 0.2 g/L ammonium sulfate. Illustrated in the bar graph, L36Δmga2 presented a significantly reduced lipid level 45 comparing to L36 and L36Δmga2 MGA2-36 presented an elevated level of lipid accumulation comparing to L36Δmga2, indicating that mga2-36 is the reason of the high lipid accumulation phenotype in L36 strain. Combining the data with Δmga2 and Δmga2 MGA2-36 in PO1f, this set of 50 data proves that Δmga2 can lead to improved lipid accumulation and further introduce the mutant transcriptional factor MGA2-36 can further elevate the level of lipid accumulation. (All strains in the set contain an episomal plasmid with LEU2). Lipid accumulation characterized with flow cytometry using cells stained with Nile Red on 192 h time point with yeast synthetic medium containing 160 g/L glucose and 0.2 g/L ammonium sulfate and 96 h time point with yeast 55 synthetic medium containing 80 g/L glucose and 5 g/L ammonium sulfate. Introducing MGA2-36 to the engineered strain leads to elevated level of lipid accumulation, suggesting MGA2-36 can be used a lipid enhancer in the rationally engineered lipid production strain. Lipid accumulation characterized with flow cytometry using cells stained with Nile Red on 192 h time point with yeast synthetic medium containing 160 g/L glucose and 0.2 g/L ammonium sulfate. PO1fΔmga2 leu+ showed improved level of lipid accumulation 60 comparing to PO1f leu+ indicating mga2 knockout

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could improve lipid accumulation. Introducing a transmembrane domain truncated MGA2-36 in PO1f could elevate the lipid level inside the cell.

FIG. 18. Gas chromatography characterization of major fatty acid species profile in PO1f, Engineered strain and L36. L36 overproduced C16:1n9 fatty acid which could be linked with the mutant of MGA2 gene, which plays an important function on activating/regulating delta9 desaturase expression.

FIG. 19. Lipid accumulation characterized with flow cytometry using cells stained with Nile Red on 96 h time point with yeast synthetic medium containing 80 g/L glucose and 5 g/L ammonium sulfate. 1st round EMS mutagenesis and floating cell transfer method selected strain E26 and E13 using final engineered strain PO1f Δpepx10,mfe DGA1 leu+ ura+ presented a higher lipid accumulation level comparing to the engineered strain.

FIG. 20. Lipid production (g/L) in bioreaction with 160 g/L glucose and 13.4 g/L YNB with ammonium sulfate without amino acid (set control DO at 50% and pH=3.5) with engineered strain and evolved strain E26.

FIG. 21. List of consensus mutations in strain E26 and E13 identified in open reading frame through next generation sequencing analysis. Among them, YLOSH6; YLIRC20; YLRME1; YLYOX1; YLUGA2 contains missense mutations in annotated protein.

FIG. 22. Summary illustration of 1st and 2nd round of EMS mutagenesis and floating cells transfer selection with final engineered strain PO1f Δpepx10,mfe DGA1 leu+ ura+ as starting strain for evolving and selecting high lipid production strain. Green indicating the final engineering strain, blue indicating the non-EMS treated control stains and red indicating the selected high lipid production strains. Strains were rank ordered based on the value cultured OD600*Nile Red mean fluorescence intensity*10⁻⁴.

FIG. 23. Fermentation profiles of pex10 mfe1 leucine⁺ uracil⁺ DGA1 and PO1f leucine⁺ uracil⁺. Time courses of the 1.5 L scale batch fermentation of the pex10 mfe1 leucine⁺ uracil⁺ DGA1 (a,b) and PO1f leucine⁺ uracil⁺ (c,d) strains in 80 g/L glucose, 6.7 g/L YNB (no amino acids, 1.365 g/L ammonium) are shown, including production of biomass, lipids, and citric acid (left axis a,c), consumption of glucose (right axis a,c), and ammonium level (b,d). (a) During the pex10 mfe1 leucine⁺ uracil⁺ DGA1 fermentation, negligible citric acid was produced, and lipid product accumulated during and after biomass production phases. This fermentation was run three times in identical conditions, reaching final yields of 15.25 g/L lipids and 20.3 g/L biomass (75% lipid content), 14.96 g/L lipids and 20.6 g/L biomass (73% lipid content), and 16.9 g/L lipids and 19.21 g/L biomass (88% lipid content). Most time points show average values from the former two fermentations (75% and 73% final lipid content), while endpoints represent averages from all three final values. Glucose and ammonium substrate were fully consumed after 72 hours, but surprisingly, (b) ammonium level was replenished to a steady state level of ~0.5 g/L, almost 40% of the original starting level. (c) During the PO1f leucine⁺ uracil⁺ fermentation, citric acid accumulated to more than 14 g/L after 72 hours before quickly reducing to 4 g/L. Lipid production did not trend with biomass production, reaching a final yield of only 3 g/L lipids, compared to 30 g/L biomass, and glucose was again consumed within 72 hours. (d) Ammonium was fully consumed after 72 hours with no replenishment as observed in the mutant strain.

DETAILED DESCRIPTION

Our work described herein represents the largest scale engineering effort in an oleaginous organism to date. We

analyzed the effect of nitrogen starvation and carbon level on a wildtype *Y. lipolytica* strain and a strain with two genomic modifications to increase lipid (e.g. triacylglyceride) accumulation. By testing twenty media formulations containing between 10 g/L and 320 g/L glucose and 0.04 g/L and 10 g/L ammonium sulfate, we demonstrated that increasing carbon to nitrogen ratio (C:N ratio) generally induces lipid (e.g. triacylglyceride) accumulation, that carbon level is more important than nitrogen level towards this induction, and that this optimum carbon level is dependent upon genomic background. We further determined that lipid (e.g. triacylglyceride) accumulation could be increased through the addition of certain metallic cofactors in the wildtype background as well as for some *Y. lipolytica* strains already engineered for increased lipid (e.g. triacylglyceride) content. In an effort to rationally engineer *Y. lipolytica* for increased lipid (e.g. triacylglyceride) accumulation while accounting for unpredictable cumulative effects arising from simultaneously altering fatty acid, lipid, and central carbon metabolism, we overexpressed multiple (e.g. five) enzymes implicated in lipid (e.g. triacylglyceride) accumulation in multiple (e.g. four) background strains differentially deficient in fatty acid degradation. These native enzymatic overexpressions were driven by high-strength constitutive promoters, occurred singly or in tandem with a second enzyme overexpression, and alleviated one of two auxotrophies (leucine and uracil). This combinatorial approach generated over 50 distinct genotypes that produced a large range in lipid (e.g. triacylglyceride) accumulation ability, culminating in upwards of 40-fold above control when using Nile-red based fluorescence and nearly 5-fold when using concentration (g/L) or percent lipid by cell mass (% dcw). In the process, we discovered a correlation between the auxotrophic marker used to select for protein overexpression and a strain's capacity to accumulate oleo-content. Specifically, the ability to endogenously produce the amino acid leucine, conferred by a selectable leucine auxotrophic marker, is beneficial (e.g. essential) to enable high lipid titer. We further examined a few (e.g. thirteen) of these strains to determine how C:N ratio and genotype interacted towards producing lipid (e.g. triacylglyceride) content on a larger scale. We observed a strong tendency towards high lipid (e.g. triacylglyceride) levels in most high producers at a single media formulation—cultivated in 80 g/L glucose and 5 g/L ammonium sulfate. We selected a MFE1, PEX10 double knockout strain with no auxotrophies overexpressing the DGA1p lipid synthesis as our final rationally engineered strain, and demonstrated its triacylglyceride accumulation ability on a variety of carbon sources, demonstrating its robust capacity to accumulate triacylglycerides regardless of media composition.

Through our time working with *Y. lipolytica*, we became aware of its surprising capacity to randomly (or forcibly) through the use of an exogenous mutagen such as EMS) generate isolatable sub-strains that reproducibly displayed higher than wildtype triacylglyceride levels. In fact, one such strain, dubbed L36, displayed remarkable accumulation ability. Whole-genome sequencing of this strain pinpointed a mutation in the MGA2 transcriptional regulator as the most likely genomic explanation. Complementation assays of an MGA2p truncation mutant into wildtype background reached 50% of L36 lipid levels. We sought to harness this general capacity for beneficial mutation by subjecting wildtype, L36, and two of our highest producing rationally engineered strains to ethylmethanesulfonate (EMS) mutagenesis and positive selection. By combining large-scale investigations of phenotypic induction, genomic

engineering, and positive random mutations, this work establishes a framework for engineering oleaginous organisms for increased lipid production. In this regard, we have pinpointed specific media formulations, genomic modifications, and genomic mutations that positively effect lipid (e.g. triacylglyceride) biosynthesis. The resultant strains are ideal for direct biodiesel precursor synthesis, lipid synthesis, oleochemical synthesis, lipid precursor synthesis, or for in vivo catalysis of fatty acid reserves to value added chemicals. Lipid accumulation characterized with flow cytometry using cells stained with Nile Red on 192 h time point with yeast synthetic medium containing 160 g/L glucose and 0.2 g/L ammonium sulfate and 96 h time point with yeast synthetic medium containing 80 g/L glucose and 5 g/L ammonium sulfate. Introducing MGA2-36 to the engineered strain leads to elevated level of lipid accumulation, suggesting MGA2-36 can be used a lipid enhancer in the rationally engineered lipid production strain. Lipid accumulation characterized with flow cytometry using cells stained with Nile Red on 192 h time point with yeast synthetic medium containing 160 g/L glucose and 0.2 g/L ammonium sulfate. PO1fΔmga2 leu+ showed improved level of lipid accumulation comparing to PO1f leu+ indicating mga2 knockout could improve lipid accumulation. Introducing a transmembrane domain truncated MGA2-36 in PO1f could elevate the lipid level inside the cell.

I. DEFINITIONS

The term “oleaginous organism” means an organism (e.g. a cell such as a yeast cell) that is capable of producing a lipid, lipid precursor, oleochemical, or oil (or combinations thereof) at a level exceeding the amount required for normal cellular survival and propagation of the organism (e.g. cell, yeast cell), such as for example necessary for structural integrity (e.g. membrane formation and maintenance) and cellular maintenance. Examples of amounts exceeding the amount required for normal cellular survival and propagation include an amount of lipids, oils, lipid precursors, and oleochemicals greater than 20% wt/wt total dry weight (e.g. greater than 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99%). In embodiments, the oleaginous organism is an oleaginous yeast. In some embodiments, the oleaginous yeast is from a genus selected from the group consisting of *Apotrichum*, *Candida*, *Cryptococcus*, *Debaromyces*, *Endomycopsis*, *Geotrichum*, *Hyphopichia*, *Lipomyces*, *Lypomyces*, *Pichia*, *Rodosporidium*, *Rhodotorula*, *Sporobolomyces*, *Starmerella*, *Torulaspora*, *Trichosporon*, *Wickerhamomyces*, *Yarrowia*, and *Zygoascus*. In embodiments, the oleaginous yeast is selected from the group consisting of *Apotrichum curvatum*, *Candida apicola*, *Candida curvata*, *Candida revkaufi*, *Candida pulcherrima*, *Candida tropicalis*, *Candida utilis*, *Cryptococcus curvatus*, *Cryptococcus terricolus*, *Debaromyces hansenii*, *Endomycopsis vernalis*, *Geotrichum carabidarum*, *Geotrichum cucujoidarum*, *Geotrichum histeridarum*, *Geotrichum silvicola*, *Geotrichum vulgare*, *Hyphopichia burtonii*, *Lipomyces lipoferus*, *Lipomyces lipofer*, *Lipomyces orientalis*, *Lipomyces starkeyi*, *Lipomyces tetrasporous*, *Pichia mexicana*, *Rodosporidium sphaeroecarpum*, *Rhosporidium toruloides*, *Rhodotorula aurantiaca*, *Rhodotorula dairenensis*, *Rhodotorula diffuens*, *Rhodotorula glutinus*, *Rhodotorula glutinis* var. *glutinis*, *Rhodotorula gracilis*, *Rhodotorula graminis*, *Rhodotorula*

minuta, *Rhodotorula mucilaginosa*, *Rhodotorula mucilaginosa* *Rhodotorula mucilaginosa*, *Rhodotorula terpenoidalis*, *Rhodotorula toruloides*, *Sporobolomyces alborubescens*, *Starmerella bombicola*, *Torulaspora delbruekii*, *Torulaspora pretoriensis*, *Trichosporon behrend*, *Trichosporon brassicae*, *Trichosporon cutaneum*, *Trichosporon domesticum*, *Trichosporon fermentans*, *Trichosporon laibachii*, *Trichosporon loubieri*, *Trichosporon loubieri* var. *loubieri*, *Trichosporon montevideense*, *Trichosporon pullulans*, *Wickerhamomyces canadensis*, *Yarrowia lipolytica*, and *Zygoascus meyeriae*.

The term “buoyancy” is used according to its plain ordinary meaning and refers to the upward force exerted by a fluid, which opposes the weight of an immersed object (e.g. oleaginous organism or oleaginous yeast cell). Pressure increases with depth, resulting in a net force tending to accelerate object upward, wherein the magnitude of the force is proportional to the difference between the top and bottom of the fluid and is equivalent to the weight of the fluid that would otherwise occupy the space occupied by the object (i.e. the displace fluid). In embodiments, an oleaginous organism or yeast cell is considered “buoyant” when it does not settle (e.g. due to gravitation force alone, due to centrifugal force, due to an applied force, or due to a combination of forces such as centrifugation) to the bottom of a vessel holding a liquid (e.g. media) in which the oleaginous organism or yeast cell resides. For example, a cell may be buoyant if it floats above the bottom of the vessel, at an intermediate position between the bottom level and top level of the liquid, or on top of the upper surface of the liquid. An example of a measurement of the buoyancy of an object (e.g. cell) is the weight of the fluid the object would displace if the object were placed in the fluid. Another example of a measurement of the buoyancy of an object (e.g. cell) is a comparison of the average density of the object and the average density of the liquid to be displaced, taking into account the depth of the liquid in a column of the liquid. The term “buoyant density” is used according to its plain ordinary meaning and refers to a measure of the tendency of a substance to float in some other substance.

The term “carbon substrate” means a carbon source that a microorganism (e.g. oleaginous organism or oleaginous yeast) will metabolize to derive energy (e.g. monosaccharides, oligosaccharides, polysaccharides, alkanes, fatty acids, esters of fatty acids, monoglycerides, carbon dioxide, methanol, formaldehyde, formate or carbon-containing amines). The term “carbon source” refers to a carbon containing composition (e.g. compound, mixture of compounds) that an organism (e.g. oleaginous organism, yeast cell) may metabolize for use by the organism or that may be used for organism viability. A “majority carbon source” refers to a carbon containing composition that accounts for greater than 50% of the available carbon sources for an organism (e.g. in a media, in a growth media, in a defined media for growing yeast cells, or in a defined media for producing lipids by yeast cells) at a specified time (e.g. media when starting a yeast culture, media in a bioreactor when growing yeast, or media when producing lipids from yeast). In embodiments, an oleaginous yeast may be cultured using a medium comprising one or more carbon sources selected from the group consisting of glucose, fructose, sucrose, lactose, galactose, xylose, mannose, rhamnose, arabinose, glycerol, acetate, depolymerized sugar beet pulp, black liquor, corn starch, depolymerized cellulosic material, corn stover, sugar beet pulp, switchgrass, milk whey, molasses, potato, rice, sorghum, sugar cane, wheat, and mixtures thereof (e.g. mixtures of glycerol and glucose, mixtures of

glucose and xylose, mixtures of fructose and glucose, mixtures of sucrose and depolymerized sugar beet pulp, black liquor, corn starch, depolymerized cellulosic material, corn stover, sugar beet pulp, switchgrass, milk whey, molasses, potato, rice, sorghum, sugar cane, and/or wheat). In embodiments, an oleaginous yeast is cultured using a medium comprising one or more carbon sources selected from the group consisting of depolymerized sugar beet pulp, black liquor, corn starch, depolymerized cellulosic material, corn stover, sugar beet pulp, switchgrass, milk whey, molasses, potato, rice, sorghum, sugar cane, thick cane juice, sugar beet juice, and wheat. In embodiments, an oleaginous yeast is cultured using a medium comprising lignocellulosic biomass. In embodiments carbon sources may be monosaccharides (e.g., glucose, fructose), disaccharides (e.g., lactose, sucrose), oligosaccharides, polysaccharides (e.g., starch, cellulose or mixtures thereof), sugar alcohols (e.g., glycerol) or mixtures from renewable feedstocks (e.g., cheese whey permeate, cornsteep liquor, sugar beet molasses, or barley malt). Additionally, carbon sources may include alkanes, fatty acids, esters of fatty acids, monoglycerides, diglycerides, triglycerides, phospholipids, various commercial sources of fatty acids including vegetable oils (e.g., soybean oil) or animal fats.

Nitrogen may be supplied from an inorganic (e.g., $(\text{NH}_4)_2\text{SO}_4$) or organic source (e.g., urea, glutamate). The term “nitrogen source” refers to a nitrogen containing composition (e.g. compound, mixture of compounds, salt) that an organism (e.g. oleaginous organism, yeast cell) may metabolize for use by the organism or that may be used for organism viability. A “majority nitrogen source” refers to a nitrogen containing composition that accounts for greater than 50% of the available nitrogen sources for an organism (e.g. in a media, in a growth media, in a defined media for growing yeast cells, or in a defined media for producing lipids by yeast cells) at a specified time (e.g. media when starting a yeast culture, media in a bioreactor when growing yeast, or media when producing lipids from yeast).

The term “Biomass” refers to material produced by growth and/or propagation of cells. “Lignocellulosic biomass” is used according to its plain ordinary meaning and refers to plant dry matter comprising carbohydrate (e.g. cellulose or hemicellulose) and polymer (e.g. lignin). Lignocellulosic biomass may include agricultural residues (e.g. corn stover or sugarcane bagasse), energy crops (e.g. poplar trees, willow, *Miscanthus purpureum*, *Pennisetum purpureum*, elephant grass, maize, Sudan grass, millet, white sweet clover, rapeseed, giant miscanthus, switchgrass, jatropha, *Miscanthus giganteus*, or sugarcane), wood residues (e.g. sawmill or papermill discard), or municipal paper waste.

The term “Culture”, “cultivate”, and “ferment” are used interchangeably and refer to the intentional growth, propagation, proliferation, and/or enablement of metabolism, catabolism, and/or anabolism of one or more cells (e.g. oleaginous organism or oleaginous yeast). The combination of both growth and propagation may be termed proliferation. Examples include production by an organism of lipids, lipid precursors, and/or oleochemicals or production of a lipid, lipid precursor, and/or oleochemical of interest. Culture does not refer to the growth or propagation of microorganisms in nature or otherwise without human intervention.

The terms “dry weight” and “dry cell weight” are used interchangeably and refer to a weight determined in the relative absence of water. In embodiments, oleaginous yeast biomass comprising a fraction or percentage of a particular component by dry weight means that the fraction or per-

centage is calculated based on the weight of the biomass after substantially all water has been removed.

The term “growth” means an increase in cell size, total cellular contents, and/or cell mass or weight of a cell (e.g. oleaginous organism or oleaginous yeast).

The term “lipid” refers to a class of molecules that are soluble in nonpolar solvents (e.g. ether or chloroform), are relatively or completely insoluble in water, and include one or more hydrocarbon chains which are hydrophobic. In 10 embodiments, a lipid may be a triacylglyceride (i.e. fat), fatty acid (e.g. saturated or unsaturated); glyceride or glycerolipid (e.g. monoglyceride, diglyceride, triglyceride, neutral fat, phosphoglyceride, or glycerophospholipid); sphingolipid; sterol lipid (e.g. cholesterol or a steroid hormone); prenol lipid (e.g. terpenoid); fatty alcohol; wax; polyketide; sugar-linked lipid, glycolipid, or protein-linked lipid.

The term “oil” means a triacylglyceride (or triglyceride oil), produced by an organism (e.g. oleaginous organism, oleaginous yeast, plant, and/or animal). An oil is generally 20 liquid at normal ambient temperatures and pressures. In embodiments, oil may be vegetable or seed oils derived from plants (e.g. soy, rapeseed, canola, palm, palm kernel, coconut, corn, olive, sunflower, cotton seed, *cuphea*, peanut, camelina sativa, mustard seed, cashew nut, oats, lupine, kenaf, *calendula*, hemp, coffee, linseed, hazelnut, *euphorbia*, pumpkin seed, coriander, camellia, sesame, safflower, rice, tung oil tree, cocoa, copra, pium poppy, castor beans, pecan, jojoba, jatropha, *macadamia*, Brazil nuts, avocado, or combinations thereof). An oil may include a plurality of 30 different triacylglycerides. For example, a vegetable or seed oil may include more than one triacylglyceride and use of the name of that vegetable or seed oil (e.g. soy, rapeseed, canola, palm, etc.) when referring to an oil generated by an oleaginous organism will be understood to mean an oil including most (e.g. all) of the triacylglycerides normally in the vegetable or seed oil (e.g. at different ratios relative to each other or the same or similar ratios relative to each other). In other embodiments, an oil may be a plurality of 35 triacylglyceride and other lipid molecules produced by an oleaginous organism.

The term “propagation” refers to an increase in cell number via cell division.

The terms “V/V”, “vol/vol”, or “v/v”, referring to proportions by volume, means the ratio of the volume of one substance in a composition to the volume of the total composition including the substance.

The term “W/W”, “wt/wt”, or “w/w”, referring to proportions by weight, means the ratio of the weight of one substance in a composition to the weight of the total composition including the substance. For example, 5% w/w substance X means that 5% of the composition’s weight is composed of substance X and the remainder of the weight of the composition (i.e. 95%) is composed of other substances.

The term “promoter” or “regulatory element” refers to a 55 region or sequence determinants located upstream or downstream from the start of transcription and which are involved in recognition and binding of RNA polymerase and other proteins to initiate transcription. Promoters need not be of yeast origin, for example, promoters derived from viruses or from other organisms can be used in the compositions or methods described herein.

A polynucleotide sequence is “heterologous to” a second 60 polynucleotide sequence if it originates from a foreign species, or, if from the same species, is modified by human action from its original form. For example, a promoter operably linked to a heterologous coding sequence refers to a coding sequence from a species different from that from

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which the promoter was derived, or, if from the same species, a coding sequence which is different from naturally occurring allelic variants.

The term "recombinant" refers to a human manipulated nucleic acid (e.g. polynucleotide) or a copy or complement of a human manipulated nucleic acid (e.g. polynucleotide), or if in reference to a protein (i.e., a "recombinant protein"), a protein encoded by a recombinant nucleic acid (e.g. polynucleotide). In embodiments, a recombinant expression cassette comprising a promoter operably linked to a second nucleic acid (e.g. polynucleotide) may include a promoter that is heterologous to the second nucleic acid (e.g. polynucleotide) as the result of human manipulation (e.g., by methods described in Sambrook et al., *Molecular Cloning—A Laboratory Manual*, Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y., (1989) or Current Protocols in Molecular Biology Volumes 1-3, John Wiley & Sons, Inc. (1994-1998)). In another example, a recombinant expression cassette may comprise nucleic acids (e.g. polynucleotides) combined in such a way that the nucleic acids (e.g. polynucleotides) are extremely unlikely to be found in nature. For instance, human manipulated restriction sites or plasmid vector sequences may flank or separate the promoter from the second nucleic acid (e.g. polynucleotide). In embodiments, a recombinant nucleic acid is a nucleic acid in an oleaginous organism (e.g. oleaginous yeast) that has been manipulated by a human, for example a recombinant nucleic acid comprising a coding region for a protein that is over-expressed in an oleaginous organism relative to the absence of the recombinant nucleic acid or a recombinant nucleic acid that results in disruption of a coding region or promoter region of an oleaginous organism and reduces or eliminates expression of a protein relative the absence of the recombinant nucleic acid. One of skill will recognize that nucleic acids (e.g. polynucleotides) can be manipulated in many ways and are not limited to the examples above.

"Nucleic acid" or "oligonucleotide" or "polynucleotide" or grammatical equivalents used herein means at least two nucleotides covalently linked together. The term "nucleic acid" includes single-, double-, or multiple-stranded DNA, RNA and analogs (derivatives) thereof. Oligonucleotides are typically from about 5, 6, 7, 8, 9, 10, 12, 15, 25, 30, 40, 50 or more nucleotides in length, up to about 100 nucleotides in length. Nucleic acids and polynucleotides are polymers of any length, including longer lengths, e.g., 200, 300, 500, 1000, 2000, 3000, 5000, 7000, 10,000, etc. In certain embodiments, the nucleic acids herein contain phosphodiester bonds. In other embodiments, nucleic acid analogs are included that may have alternate backbones. The term encompasses nucleic acids containing known analogues of natural nucleotides which have similar or improved binding properties, for the purposes desired, as the reference nucleic acid. A particular nucleic acid sequence also encompasses "splice variants." Similarly, a particular protein encoded by a nucleic acid encompasses any protein encoded by a splice variant of that nucleic acid. "Splice variants," as the name suggests, are products of alternative splicing of a gene. After transcription, an initial nucleic acid transcript may be spliced such that different (alternate) nucleic acid splice products encode different polypeptides. Mechanisms for the production of splice variants vary, but include alternate splicing of exons. Alternate polypeptides derived from the same nucleic acid by read-through transcription are also encompassed by this definition. Any products of a splicing reaction, including recombinant forms of the splice products, are included in this definition. An example of potas-

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sium channel splice variants is discussed in Leicher, et al., *J. Biol. Chem.* 273(52):35095-35101 (1998).

The term "expression cassette" refers to a nucleic acid construct, which when introduced into a host cell, results in transcription and/or translation of a RNA or polypeptide, respectively.

The terms "identical" or percent "identity," in the context of two or more nucleic acids or polypeptide sequences, refer to two or more sequences or subsequences that are the same or have a specified percentage of amino acid residues or nucleotides that are the same (i.e., about 60% identity, preferably 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or higher identity over a specified region when compared and aligned for maximum correspondence over a comparison window or designated region) as measured using a BLAST or BLAST 2.0 sequence comparison algorithms with default parameters described below, or by manual alignment and visual inspection (see, e.g., NCBI web site or the like). Such sequences are then said to be "substantially identical." This definition also refers to, or may be applied to, the compliment of a test sequence. The definition also includes sequences that have deletions and/or additions, as well as those that have substitutions. As described below, the preferred algorithms can account for gaps and the like. Preferably, identity exists over a region that is at least about 10 amino acids or 20 nucleotides in length, or more preferably over a region that is 10-50 amino acids or 20-50 nucleotides in length. As used herein, percent (%) amino acid sequence identity is defined as the percentage of amino acids in a candidate sequence that are identical to the amino acids in a reference sequence, after aligning the sequences and introducing gaps, if necessary, to achieve the maximum percent sequence identity. Alignment for purposes of determining percent sequence identity can be achieved in various ways that are within the skill in the art, for instance, using publicly available computer software such as BLAST, BLAST-2, ALIGN, ALIGN-2 or Megalign (DNASTAR) software. Appropriate parameters for measuring alignment, including any algorithms needed to achieve maximal alignment over the full-length of the sequences being compared can be determined by known methods.

For sequence comparisons, typically one sequence acts as a reference sequence, to which test sequences are compared. When using a sequence comparison algorithm, test and reference sequences are entered into a computer, subsequence coordinates are designated, if necessary, and sequence algorithm program parameters are designated. Preferably, default program parameters can be used, or alternative parameters can be designated. The sequence comparison algorithm then calculates the percent sequence identities for the test sequences relative to the reference sequence, based on the program parameters.

A "comparison window", as used herein, includes reference to a segment of any one of the number of contiguous positions selected from the group consisting of from 10 to 600, usually about 50 to about 200, more usually about 100 to about 150 in which a sequence may be compared to a reference sequence of the same number of contiguous positions after the two sequences are optimally aligned. Methods of alignment of sequences for comparison are well-known in the art. Optimal alignment of sequences for comparison can be conducted, e.g., by the local homology algorithm of Smith & Waterman, *Adv. Appl. Math.* 2:482 (1981), by the homology alignment algorithm of Needleman & Wunsch, J.

Mol. Biol. 48:443 (1970), by the search for similarity method of Pearson & Lipman, *Proc. Nat'l. Acad. Sci. USA* 85:2444 (1988), by computerized implementations of these algorithms (GAP, BESTFIT, FASTA, and TFASTA in the Wisconsin Genetics Software Package, Genetics Computer Group, 575 Science Dr., Madison, Wis.), or by manual alignment and visual inspection (see, e.g., *Current Protocols in Molecular Biology* (Ausubel et al., eds. 1995 supplement)).

One example of algorithm that is suitable for determining percent sequence identity and sequence similarity are the BLAST and BLAST 2.0 algorithms, which are described in Altschul et al. (1977) *Nuc. Acids Res.* 25:3389-3402, and Altschul et al. (1990) *J. Mol. Biol.* 215:403-410, respectively. Software for performing BLAST analyses is publicly available through the National Center for Biotechnology Information (<http://www.ncbi.nlm.nih.gov/>). This algorithm involves first identifying high scoring sequence pairs (HSPs) by identifying short words of length W in the query sequence, which either match or satisfy some positive-valued threshold score T when aligned with a word of the same length in a database sequence. T is referred to as the neighborhood word score threshold (Altschul et al., supra). These initial neighborhood word hits act as seeds for initiating searches to find longer HSPs containing them. The word hits are extended in both directions along each sequence for as far as the cumulative alignment score can be increased. Cumulative scores are calculated using, for nucleotide sequences, the parameters M (reward score for a pair of matching residues; always >0) and N (penalty score for mismatching residues; always <0). For amino acid sequences, a scoring matrix is used to calculate the cumulative score. Extension of the word hits in each direction are halted when: the cumulative alignment score falls off by the quantity X from its maximum achieved value; the cumulative score goes to zero or below, due to the accumulation of one or more negative-scoring residue alignments; or the end of either sequence is reached. The BLAST algorithm parameters W, T, and X determine the sensitivity and speed of the alignment. The BLASTN program (for nucleotide sequences) uses as defaults a wordlength (W) of 11, an expectation (E) or 10, M=5, N=-4 and a comparison of both strands. For amino acid sequences, the BLASTP program uses as defaults a wordlength of 3, and expectation (E) of 10, and the BLOSUM62 scoring matrix (see Henikoff and Henikoff (1989) *Proc. Natl. Acad. Sci. USA* 89:10915) alignments (B) of 50, expectation (E) of 10, M=5, N=-4, and a comparison of both strands.

The BLAST algorithm also performs a statistical analysis of the similarity between two sequences (see, e.g., Karlin and Altschul (1993) *Proc. Natl. Acad. Sci. USA* 90:5873-5877). One measure of similarity provided by the BLAST algorithm is the smallest sum probability (P(N)), which provides an indication of the probability by which a match between two nucleotide or amino acid sequences would occur by chance. For example, a nucleic acid is considered similar to a reference sequence if the smallest sum probability in a comparison of the test nucleic acid to the reference nucleic acid is less than about 0.2, more preferably less than about 0.01.

The phrase "selectively (or specifically) hybridizes to" refers to the binding, duplexing, or hybridizing of a molecule only to a particular nucleotide sequence with a higher affinity, e.g., under more stringent conditions, than to other nucleotide sequences (e.g., total cellular or library DNA or RNA).

The phrase "stringent hybridization conditions" refers to conditions under which a probe will hybridize to its target subsequence, typically in a complex mixture of nucleic acids, but to no other sequences. Stringent conditions are sequence-dependent and will be different in different circumstances. Longer sequences hybridize specifically at higher temperatures. An extensive guide to the hybridization of nucleic acids is found in Tijsse, *Techniques in Biochemistry and Molecular Biology—Hybridization with Nucleic Probes*, "Overview of principles of hybridization and the strategy of nucleic acid assays" (1993). Generally, stringent conditions are selected to be about 5-10° C. lower than the thermal melting point (T_m) for the specific sequence at a defined ionic strength pH. The T_m is the temperature (under defined ionic strength, pH, and nucleic concentration) at

which 50% of the probes complementary to the target hybridize to the target sequence at equilibrium (as the target sequences are present in excess, at T_m , 50% of the probes are occupied at equilibrium). Stringent conditions may also be achieved with the addition of destabilizing agents such as formamide. For selective or specific hybridization, a positive signal is at least two times background, preferably 10 times background hybridization. Exemplary stringent hybridization conditions can be as following: 50% formamide, 5×SSC, and 1% SDS, incubating at 42° C., or, 5×SSC, 1% SDS, incubating at 65° C., with wash in 0.2×SSC, and 0.1% SDS at 65° C.

Nucleic acids that do not hybridize to each other under stringent conditions are still substantially identical if the polypeptides which they encode are substantially identical. This occurs, for example, when a copy of a nucleic acid is created using the maximum codon degeneracy permitted by the genetic code. In such cases, the nucleic acids typically hybridize under moderately stringent hybridization conditions. Exemplary "moderately stringent hybridization conditions" include a hybridization in a buffer of 40% formamide, 1 M NaCl, 1% SDS at 37° C., and a wash in 1×SSC at 45° C. A positive hybridization is at least twice background. Those of ordinary skill will readily recognize that alternative hybridization and wash conditions can be utilized to provide conditions of similar stringency. Additional guidelines for determining hybridization parameters are provided in numerous reference, e.g., and *Current Protocols in Molecular Biology*, ed. Ausubel, et al. One of skill will recognize that these values can be appropriately adjusted to determine corresponding identity of proteins encoded by two nucleotide sequences by taking into account codon degeneracy, amino acid similarity, reading frame positioning and the like. Polypeptides which are "substantially similar" share sequences as noted above except that residue positions which are not identical may differ by conservative amino acid changes. Conservative amino acid substitutions refer to the interchangeability of residues having similar side chains. For example, a group of amino acids having aliphatic side chains is glycine, alanine, valine, leucine, and isoleucine; a group of amino acids having aliphatic-hydroxyl side chains is serine and threonine; a group of amino acids having amide-containing side chains is asparagine and glutamine; a group of amino acids having aromatic side chains is phenylalanine, tyrosine, and tryptophan; a group of amino acids having basic side chains is lysine, arginine, and histidine; and a group of amino acids having sulfur-containing side chains is cysteine and methionine. Exemplary conservative amino acids substitution groups are: valine-leucine-isoleucine, phenylalanine-tyrosine, lysine-arginine, alanine-valine, aspartic acid-glutamic acid, and asparagine-glutamine.

The term "modulator" refers to a composition that increases or decreases the level of a target molecule or the level of activity or function of a target molecule or the physical state of the target of the molecule. In embodiments a modulator is a recombinant nucleic acid that is capable of increasing or decreasing the amount of a protein in a cell or the level of activity of a protein in a cell or transcription of a second nucleic acid in a cell. In embodiments, a modulator increases or decreases the level of activity of a protein or the amount of the protein in a cell. The term "modulate" is used in accordance with its plain ordinary meaning and refers to the act of changing or varying one or more properties. "Modulation" refers to the process of changing or varying one or more properties. For example, as applied to the effects of a modulator on a target protein, to modulate means to change by increasing or decreasing a property or function of the target molecule or the amount of the target molecule. In embodiments, a recombinant nucleic acid that modulates the level of activity of a protein may increase the activity or amount of the protein relative the absence of the recombinant nucleic acid. In embodiments, an increase in the activity or amount of a protein may include overexpression of the protein. "Overexpression" is used in accordance with its plain meaning and refers to an increased level of expression of a protein relative to a control (e.g. cell or expression system not including a recombinant nucleic acid that contributes to the overexpression of a protein). In embodiments, a decrease in the activity or amount of a protein may include a mutation (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid; all/any of which may be in the coding region for a protein or in an operably linked region (e.g. promoter)) of the protein. The term "increased" refers to a detectable increase compared to a control. In some embodiments, the increase is by about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 250, 300, 350, 400, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, 10000, 20000, 30000, 40000, 50000, 60000, 70000, 80000, 90000, 100000%, or more compared to the control. In embodiments, the increase is by 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 250, 300, 350, 400, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, 100000%, or more compared to the control. In embodiments, the increase is by at least 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 250, 300, 350, 400, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, 10000, 20000, 30000, 40000, 50000, 60000, 70000, 80000, 90000, 100000%, or more compared to the control.

30000, 40000, 50000, 60000, 70000, 80000, 90000, 100000%, compared to the control. Similarly, the term “decreased” refers to a measurable decrease compared to a control. In some embodiments, the decrease is by about 1, 2, 5 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 10 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100%, or more compared to the control. In embodiments, the decrease is by 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 15 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100%, or more compared to the control. In embodiments, the decrease is by at least 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 20 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100%, compared to the control. One of ordinary skill will be able to identify a relevant control.

Nucleic acid is "operably linked" when it is placed into a functional relationship with another nucleic acid sequence.

30 For example, DNA for a presequence or secretory leader is operably linked to DNA for a polypeptide if it is expressed as a preprotein that participates in the secretion of the polypeptide; a promoter or enhancer is operably linked to a coding sequence if it affects the transcription of the sequence; or a ribosome binding site is operably linked to a coding sequence if it is positioned so as to facilitate translation. Generally, "operably linked" means that the DNA sequences being linked are near each other, and, in the case of a secretory leader, contiguous and in reading phase.

40 However, operably linked nucleic acids (e.g. enhancers and coding sequences) do not have to be contiguous. Linking is accomplished by ligation at convenient restriction sites. If such sites do not exist, the synthetic oligonucleotide adaptors or linkers are used in accordance with conventional practice. In embodiments, a promoter is operably linked with a coding sequence when it is capable of affecting (e.g., modulating relative to the absence of the promoter) the expression of a protein from that coding sequence (i.e., the coding sequence is under the transcriptional control of the promoter).

"Transformation" refers to the transfer of a nucleic acid molecule into a host organism (e.g. oleaginous organism or oleaginous yeast). In embodiments, the nucleic acid molecule may be a plasmid that replicates autonomously or it 55 may integrate into the genome of the host organism (e.g. oleaginous organism or oleaginous yeast). Host organisms containing the transformed nucleic acid molecule may be referred to as "transgenic" or "recombinant" or "transformed" organisms (e.g. oleaginous organism or oleaginous yeast). A "genetically modified" organism (e.g. genetically modified yeast cell) is an organism (e.g. yeast cell) that includes a nucleic acid that has been modified by human intervention. Examples of a nucleic acid that has been modified by human intervention include, but are not limited 60 to, insertions, deletions, mutations, expression nucleic acid constructs (e.g. over-expression or expression from a non-natural promoter or control sequence or an operably linked

promoter and gene nucleic acid distinct from a naturally occurring promoter and gene nucleic acid in an organism), extra-chromosomal nucleic acids, and genetically contained modified nucleic acids. Genetically modified organisms may be made by rational modification of a nucleic acid or may be made by use of a mutagen or mutagenesis protocol that results in a mutation that was not identified (e.g. intended or targeted) prior to the use of the mutagen or mutagenesis protocol (e.g. UV exposure, EMS exposure, mutagen exposure, random genomic mutagenesis, transformation of a library of different nucleic acid constructs). Genetically modified organisms that include a modification (e.g. modification, insertion, deletion, mutation) not previously known or intended prior to making of the genetically modified organism may be identified through screening a plurality of organism including one or more genetically modified organisms by using a selection criteria that identifies the genetically modified organism of interest (e.g. an increased level of lipids, lipid precursors, and/or oleochemicals; floats above an organism not including the same genetic modification). In embodiments, a genetically modified organism includes a recombinant nucleic acid.

Methods for synthesizing sequences and bringing sequences together are well established and known to those of skill in the art. For example, in vitro mutagenesis and selection, site-directed mutagenesis, error prone PCR (Melnikov et al., Nucleic Acids Research, 27(4):1056-1062 (Feb. 15, 1999)), “gene shuffling” or other means can be employed to obtain mutations of naturally occurring genes.

Mutagenesis (e.g. chemical mutagenesis or site directed mutagenesis) may be used to modulate lipid production or storage in an oleaginous organism (e.g. oleaginous yeast). For example, a mutant construct or mutagen is transformed into an oleaginous yeast cell and the ability of the resulting transformed oleaginous yeast cell to produce or store one or more lipids is assayed and compared to the control cell. In some embodiments, it may be useful to disrupt or inactivate a host organism's native gene to modulate lipid production or storage. For example, a recombinant DNA fragment (e.g. a selectable marker gene) may be inserted into the gene to be disrupted in order to interrupt its coding sequence and the resulting recombinant nucleic acid then transformed into a host cell. Another example of a method of gene disruption is the use of transposable elements or transposons, which is well known to those of skill in the art.

In general, means for the purification of lipids, may include extraction with organic solvents, sonication, supercritical fluid extraction, saponification physical means such as presses, extraction, treatment with urea, fractional crystallization, HPLC, fractional distillation, silica gel chromatography, high-speed centrifugation or distillation, or combinations of these techniques.

In embodiments, the protein AMP Deaminase (AMPD) is a protein able to be translated from the nucleic acid corresponding to YALI0E11495 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene AMP Deaminase (AMPD) is the nucleic acid or gene corresponding to YALI0E11495 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, AMP Deaminase (AMPD) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0E11495 of *Yarrowia lipolytica* as described above. In embodiments, AMP Deaminase (AMPD) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0E11495 of *Yarrowia lipolytica* as described above. In embodiments,

the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Leucine Biosynthesis Gene (LEU2), also known as 3-isopropylmalate dehydrogenase, is a protein able to be translated from the nucleic acid corresponding to GenBank AF260230 or YALI0C00407 g of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Leucine Biosynthesis Gene (LEU2) is the nucleic acid or gene corresponding to GenBank AF260230 or YALI0C00407 g of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Leucine Biosynthesis Gene (LEU2) is a protein or nucleic acid/gene of a yeast strain corresponding to AF260230 of *Yarrowia lipolytica* as described above. In embodiments, Leucine Biosynthesis Gene (LEU2) is a protein or nucleic acid/gene of an oleaginous organism corresponding to AF260230 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Uracil Biosynthesis gene (URA3), also known as Orotidine 5'-phosphate decarboxylase, is a protein able to be translated from the nucleic acid corresponding to GenBank YLU40564 or YALI0E26741 g of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Uracil Biosynthesis gene (URA3) is the nucleic acid or gene corresponding to GenBank YLU40564 or YALI0E26741 g of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Uracil Biosynthesis gene (URA3) is a protein or nucleic acid/gene of a yeast strain corresponding to YLU40564 of *Yarrowia lipolytica* as described above. In embodiments, Uracil Biosynthesis gene (URA3) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YLU40564 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein ATP-Citrate Lyase (ACL) is a protein including the protein ACL1, also called ATP-Citrate Lyase 1, able to be translated from the nucleic acid corresponding to YALI0E34793 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene ATP-Citrate Lyase (ACL) includes the nucleic acid or gene ACL1 corresponding to YALI0E34793 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, the protein ATP-Citrate Lyase (ACL) is a protein including the protein ACL2, also called ATP-Citrate Lyase 2, able to be translated from the nucleic acid corresponding to YALI0D24431 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, the

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nucleic acid or gene ATP-Citrate Lyase (ACL) includes the nucleic acid or gene ACL2 corresponding to YALI0D24431 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, ATP-Citrate Lyase (ACL) includes a protein or nucleic acid/gene of a yeast strain corresponding to YALI0D24431 of *Yarrowia lipolytica* as described above. In embodiments, ATP-Citrate Lyase (ACL) includes a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0D24431 of *Yarrowia lipolytica* as described above. In embodiments, the protein ATP-Citrate Lyase (ACL) is a protein including the protein ACL1 able to be translated from the nucleic acid corresponding to YALI0E34793 of the Genolevures database and the protein ACL2 able to be translated from the nucleic acid corresponding to YALI0D24431 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, the nucleic acid or gene ATP-Citrate Lyase (ACL) includes the nucleic acid or gene ACL1 corresponding to YALI0E34793 and the nucleic acid or gene ACL2 corresponding to YALI0D24431 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, ATP-Citrate Lyase (ACL) includes proteins or nucleic acids/genes of a yeast strain corresponding to YALI0E34793 and YALI0D24431 of *Yarrowia lipolytica* as described above. In embodiments, ATP-Citrate Lyase (ACL) includes proteins or nucleic acids/genes of an oleaginous organism corresponding to YALI0E34793 and YALI0D24431 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Malic Enzyme (MAE, MEA, MEA1) is a protein able to be translated from the nucleic acid corresponding to YALI0E18634 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C, D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Malic Enzyme (MAE, MEA, MEA1) is the nucleic acid or gene corresponding to YALI0E18634 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Malic Enzyme (MAE, MEA, MEA1) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0E18634 of *Yarrowia lipolytica* as described above. In embodiments, Malic Enzyme (MAE, MEA, MEA1) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0E18634 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein acyl-CoA: diacylglycerol acyltransferase (DGA1), also called acyl-CoA:diacylglycerol acyltransferase 1 is a protein able to be translated from the nucleic acid corresponding to YALI0E32769 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C, D,E,F specifies chromosome. In embodiments, the nucleic acid or gene acyl-CoA:diacylglycerol acyltransferase (DGA1) is the nucleic acid or gene corresponding to YALI0E32769 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, acyl-CoA: diacylglycerol acyltransferase (DGA1) is a protein or

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nucleic acid/gene of a yeast strain corresponding to YALI0E32769 of *Yarrowia lipolytica* as described above. In embodiments, acyl-CoA:diacylglycerol acyltransferase (DGA1) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0E32769 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein acyl-CoA: diacylglycerol acyltransferase (DGA2), also called acyl-CoA:diacylglycerol acyltransferase 2, is a protein able to be translated from the nucleic acid corresponding to YALI0D07986 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C, D,E,F specifies chromosome. In embodiments, the nucleic acid or gene acyl-CoA:diacylglycerol acyltransferases (DGA2) is the nucleic acid or gene corresponding to YALI0D07986 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, acyl-CoA: diacylglycerol acyltransferases (DGA2) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0D07986 of *Yarrowia lipolytica* as described above. In embodiments, acyl-CoA:diacylglycerol acyltransferases (DGA2) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0D07986 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Lipid synthesis regulator (MGA2) is a protein able to be translated from the nucleic acid corresponding to YALI0B12342 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C, D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Lipid synthesis regulator (MGA2) is the nucleic acid or gene corresponding to YALI0B12342 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Lipid synthesis regulator (MGA2) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0B12342 of *Yarrowia lipolytica* as described above. In embodiments, Lipid synthesis regulator (MGA2) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0B12342 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Chromatin assembly gene (RLF2 subunit p90) is a protein able to be translated from the nucleic acid corresponding to YALI0F21637 g of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Chromatin assembly gene (RLF2 subunit p90) is the nucleic acid or gene corresponding to YALI0F21637 g of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Chromatin assembly gene (RLF2 subunit p90) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0F21637

g of *Yarrowia lipolytica* as described above. In embodiments, Chromatin assembly gene (RLF2 subunit p90) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0F21637 g of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Mitochondrial 2' O-ribose methyltransferase (MRM2) is a protein able to be translated from the nucleic acid corresponding to YALI0E31933 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Mitochondrial 2' O-ribose methyltransferase (MRM2) is the nucleic acid or gene corresponding to YALI0E31933 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Mitochondrial 2' O-ribose methyltransferase (MRM2) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0E31933 of *Yarrowia lipolytica* as described above. In embodiments, Mitochondrial 2' O-ribose methyltransferase (MRM2) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0E31933 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Transcription Factor (PEX10) is a protein able to be translated from the nucleic acid corresponding to YALI0C01023 g of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C, D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Transcription Factor (PEX10) is the nucleic acid or gene corresponding to YALI0C01023 g of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Transcription Factor (PEX10) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0C01023 g of *Yarrowia lipolytica* as described above. In embodiments, Transcription Factor (PEX10) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0C01023 g of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein multifunctional enzyme (MFE1) is a protein able to be translated from the nucleic acid corresponding to YALI0E15378 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C, D,E,F specifies chromosome. In embodiments, the nucleic acid or gene multifunctional enzyme (MFE1) is the nucleic acid or gene corresponding to YALI0E15378 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, multifunctional enzyme (MFE1) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0E15378 of *Yarrowia lipolytica* as described above. In embodiments, multifunctional enzyme (MFE1) is a protein or nucleic acid/gene of an oleaginous organism correspond-

ing to YALI0E15378 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein O-6-methylguanine-DNA methyltransferase (MGMT, O6M) is a protein able to be translated from the nucleic acid corresponding to YALI0C10010p of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene O-6-methylguanine-DNA methyltransferase (MGMT, O6M) is the nucleic acid or gene corresponding to YALI0C10010p of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, O-6-methylguanine-DNA methyltransferase (MGMT, O6M) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0C10010p of *Yarrowia lipolytica* as described above. In embodiments, O-6-methylguanine-DNA methyltransferase (MGMT, O6M) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0C10010p of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Aconitase (ACO1) is a protein able to be translated from the nucleic acid corresponding to YALI0D09361 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Aconitase (ACO1) is the nucleic acid or gene corresponding to YALI0D09361 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Aconitase (ACO1) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0D09361 of *Yarrowia lipolytica* as described above. In embodiments, O Aconitase (ACO1) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0D09361 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Citrate Synthase (CIT1) is a protein able to be translated from the nucleic acid corresponding to YALI0E02684 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Citrate Synthase (CIT1) is the nucleic acid or gene corresponding to YALI0E02684 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Citrate Synthase (CIT1) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0E02684 of *Yarrowia lipolytica* as described above. In embodiments, Citrate Synthase (CIT1) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0E02684 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein Acetyl-CoA Carboxylase (ACC) is a protein able to be translated from the nucleic acid corresponding to YALI0C11407 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene Acetyl-CoA Carboxylase (ACC) is the nucleic acid or gene corresponding to YALI0C11407 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, Acetyl-CoA Carboxylase (ACC) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0C11407 of *Yarrowia lipolytica* as described above. In embodiments, Acetyl-CoA Carboxylase (ACC) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0C11407 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein RME1 zinc-finger transcription factor (RME1) is a protein able to be translated from the nucleic acid corresponding to YALI0E17215 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene RME1 zinc-finger transcription factor (RME1) is the nucleic acid or gene corresponding to YALI0E17215 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, RME1 zinc-finger transcription factor (RME1) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0E17215 of *Yarrowia lipolytica* as described above. In embodiments, RME1 zinc-finger transcription factor (RME1) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0E17215 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein YOX1 homeodomain protein (YOX1) is a protein able to be translated from the nucleic acid corresponding to YALI0E20449 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene YOX1 homeodomain protein (YOX1) is the nucleic acid or gene corresponding to YALI0E20449 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, YOX1 homeodomain protein (YOX1) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0E20449 of *Yarrowia lipolytica* as described above. In embodiments, YOX1 homeodomain protein (YOX1) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0E20449 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid.

acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein UGA2 succinate semialdehyde dehydrogenase (UGA2) is a protein able to be translated from the nucleic acid corresponding to YALI0F26191 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene UGA2 succinate semialdehyde dehydrogenase (UGA2) is the nucleic acid or gene corresponding to YALI0F26191 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, UGA2 succinate semialdehyde dehydrogenase (UGA2) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0F26191 of *Yarrowia lipolytica* as described above. In embodiments, UGA2 succinate semialdehyde dehydrogenase (UGA2) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0F26191 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein OSH6 oxysterol-binding protein homolog 6 (OSH6) is a protein able to be translated from the nucleic acid corresponding to YALI0A02354 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene OSH6 oxysterol-binding protein homolog 6 (OSH6) is the nucleic acid or gene corresponding to YALI0A02354 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, OSH6 oxysterol-binding protein homolog 6 (OSH6) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0A02354 of *Yarrowia lipolytica* as described above. In embodiments, OSH6 oxysterol-binding protein homolog 6 (OSH6) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0A02354 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

In embodiments, the protein IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) is a protein able to be translated from the nucleic acid corresponding to YALI0C07150 of the Genolevures database (i.e. found at <http://www.genolevures.org/>), wherein YALI0 stands for *Yarrowia lipolytica* and A,B,C,D,E,F specifies chromosome. In embodiments, the nucleic acid or gene IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) is the nucleic acid or gene corresponding to YALI0C07150 of the Genolevures database (i.e. found at <http://www.genolevures.org/>). In embodiments, IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) is a protein or nucleic acid/gene of a yeast strain corresponding to YALI0C07150 of *Yarrowia lipolytica* as described above. In embodiments, IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) is a protein or nucleic acid/gene of an oleaginous organism corresponding to YALI0C07150 of *Yarrowia lipolytica* as described above. In embodiments, the protein and/or nucleic acid is a wildtype version of the protein or nucleic acid. In embodiments, the protein and/or nucleic acid is a mutant form of the protein and/or nucleic acid.

acid is a mutant form of the protein and/or nucleic acid (e.g. point mutant, loss of function mutation, missense mutation, deletion, or insertion of heterologous nucleic acid).

As used to describe a protein or nucleic acid of another organism in comparison to a protein or nucleic acid of *Yarrowia lipolytica*, the term “corresponds” or “corresponding” is used according to its ordinary meaning and refers to a protein or nucleic acid/gene that includes similar or identical sequence of amino acid or nucleotides respectively and/or performs a similar or identical function and/or has a similar or identical activity as the protein or nucleic acid/gene in *Yarrowia lipolytica* as described above. In some embodiments, a protein or nucleic acid corresponding to a protein or nucleic acid from *Yarrowia lipolytica* is a homolog. In embodiments, the protein and/or nucleic acid of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Aconitase (ACO1), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), or IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) includes an amino acid and/or nucleotide sequence included in the protein and/or nucleic acid sequence for Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Aconitase (ACO1), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), or IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) respectively, described herein (e.g. Examples section and/or sequence listing). In embodiments, the protein and/or nucleic acid of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Aconitase (ACO1), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), or IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) is the amino acid and/or nucleo-

tide sequence of the protein and/or nucleic acid sequence for Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Aconitase (ACO1), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), or IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) respectively, described herein (e.g. Examples section and/or sequence listing).

The term “wildtype” as used herein when referring to an oleaginous organism (e.g. yeast strain or *Yarrowia lipolytica* strain) means an organism that has not been genetically modified to improve production of a lipid (e.g. increase yield of a lipid, alter the structure of a lipid produced by the organism, reduce production of one lipid to improve production of a second lipid, or modulate the production of a lipid). In embodiments, a wildtype yeast strain may be auxotrophic for one or more compounds (e.g. leucine and/or uracil). In embodiments, a wildtype *Yarrowia lipolytica* strain is PO1f (ATCC #MYA-2613), a leucine and uracil auxotroph devoid of any secreted protease activity (Madzak et al., 2000).

The term “oleochemical” is used herein in accordance with its well known meaning and refers to chemicals or compounds derived from lipids or fats. In embodiments, an oleochemical is a lipid or fat derived from a different lipid or fat. In embodiments an oleochemical is a chemical or compound produced by an oleaginous organism. In embodiments, an oleochemical is a chemical or compound derived from a lipid or lipid precursor produced by an oleaginous organism (e.g., fatty acid esters such as methyl esters, ethyl esters, propyl esters, or butyl esters that are derived from a fatty acid produced by an oleaginous organism by transesterification). In embodiments, an oleochemical may include further *in vivo* or *in vitro* modification of a lipid or lipid precursor enabled by endogenous or heterologous modifying enzymes or chemical reactions.

The term “lipid precursor” is used in accordance with its well known meaning and refers to a pathway intermediate (e.g., acetyl-CoA or malonyl-CoA) in the biosynthesis of a lipid. In embodiments, a lipid precursor may be any molecule along the biosynthetic pathway making triglycerides including free citrate, acetyl-CoA, free fatty acids, pyruvate, citric acid cycle intermediates, diacylglycerides, and/or triacylglycerides.

The term “micronutrient” is used in accordance with its well known meaning and refers to nutrients used by an organism (e.g. oleaginous organisms, yeast, oleaginous yeast) for growth, proliferation, propagation, survival, one or more essential biological functions, production of a lipid, lipid precursor, or oleochemical, which are required for such functions in small quantities. Examples of micronutrients include, but are not limited to, minerals, vitamins, and elements (e.g. cobalt, iron, magnesium, potassium, zinc, nickel, molybdenum, manganese, copper, and/or boron).

II. OLEAGINOUS ORGANISMS

In a first aspect is provided a genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell,

Yarrowia lipolytica, algae, or plant cell) wherein the dry weight of the oleaginous organism includes greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).

79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94,
95, 96, 97, 98, or 99% lipids, lipid precursors, and/or
oleochemicals (e.g., lipid, lipids, lipid precursors, lipid pre-
cursor, oleochemicals, or oleochemical) in dry weight. In
5 embodiments, the oleaginous organism (e.g. yeast cell,
oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant
cell) includes greater than 20, 21, 22, 23, 24, 25, 26, 27, 28,
29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44,
45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60,
10 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76,
77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92,
93, 94, 95, 96, 97, 98, or 99% lipids, lipid precursors, and/or
oleochemicals (e.g., lipid, lipids, lipid precursors, lipid pre-
cursor, oleochemicals, or oleochemical) in dry weight.
15 In embodiments, the oleaginous organism (e.g. yeast cell,
oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant
cell) produces about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13,
14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29,
20 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45,
46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61,
62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77,
78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93,
94, 95, 96, 97, 98, 99, or 100 g/L culture (e.g. in a bioreactor)
of lipids, lipid precursors, and/or oleochemicals (e.g., lipid,
lipids, lipid precursors, lipid precursor, oleochemicals, or
oleochemical). In embodiments, the oleaginous organism
25 (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*,
algae, or plant cell) produces at least 1, 2, 3, 4, 5, 6, 7, 8, 9,
10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25,
30 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41,
42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57,
58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73,
74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89,
35 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, or 100 g/L culture (e.g.
in a bioreactor) of lipids, lipid precursors, and/or oleochemi-
cals (e.g., lipid, lipids, lipid precursors, lipid precursor,
oleochemicals, or oleochemical). In embodiments, the ole-
aginous organism (e.g. yeast cell, oleaginous yeast cell,
40 *Yarrowia lipolytica*, algae, or plant cell) produces 1, 2, 3, 4,
5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21,
22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37,
38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53,
45 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69,
70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85,
86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, or 100
g/L culture (e.g. in a bioreactor) of lipids, lipid precursors,
and/or oleochemicals (e.g., lipid, lipids, lipid precursors,
lipid precursor, oleochemicals, or oleochemical).
In embodiments, the oleaginous organism is a yeast cell.
50 In embodiments, the oleaginous organism is an oleaginous
yeast cell. In embodiments, the yeast cell is selected from the
group consisting of the genera *Yarrowia*, *Candida*, *Rhodoto-*
rula, *Rhodosporidium*, *Cryptococcus*, *Trichosporon* and
Lipomyces. In embodiments, the yeast cell is selected from
55 the group consisting of *Rhodosporidium toruloides*, *Lipo-*
myes starkeyii, *Lipomyces lipoferus*, *Apotrichum curva-*
tum, *Candida curvata*, *Cryptococcus curvatus*, *Trichos-*
poron fermentans, *Candida revkaufi*, *Candida pulcherrima*,
Candida tropicalis, *Candida utilis*, *Trichosporon pullans*,
60 *Trichosporon cutaneum*, *Rhodotorula glutinis*, *Rhodotorula*
graminis and *Yarrowia lipolytica*. In embodiments, the yeast
cell is selected from the group consisting of *Lipomyces*
starkeyii, *Rhodosporidium toruloides*, *Apotrichum curva-*
tum, *Candida curvata*, *Cryptococcus curvatus*, *Trichos-*
65 *poron fermentans*, *Rhodotorula glutinis*, and *Yarrowia*
lipolytica. In embodiments, the yeast cell is *Yarrowia*
lipolytica.

In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is buoyant in an aqueous medium. In embodiments, the yeast cell includes a greater buoyancy (i.e. greater tendency to float, lower density) than a yeast cell that includes less than about 99% lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) by dry weight (e.g. less than about 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight). In embodiments, the yeast cell includes a greater buoyancy (i.e. greater tendency to float, lower density) than a yeast cell that includes less than 99% lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) by dry weight (e.g. less than 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) does not sediment to the bottom of a column of liquid (e.g. water, buffer, growth media, minimal media) that is about 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1.0, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 96, 97, 98, or 99 mm tall due to gravitation force alone. The term "about" when used in connection with a defined amount refers to an amount up to and including greater than and/or less than 10% of the associated value and includes the associated value. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) does not sediment to the bottom of a column of liquid (e.g. water, buffer, growth media, minimal media) that is 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1.0, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 96, 97, 98, or 99 mm tall due to gravitation force alone. In embodiments, the yeast cell includes a greater buoyancy (i.e. greater tendency to float, lower density) than a yeast cell that does not include the same recombinant nucleic acid or combination of recombinant nucleic acids as the buoyant yeast cell. In embodiments, the yeast cell is buoyant following centrifugation (e.g. at about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 250, 300, 350, 400, 450, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 2500, 3000, 3500, 4000, 4500, 5000, 5500, 6000, 6500, 7000, 7500, 8000, 8500, 9000, 9500, or 10000×g).

In embodiments of the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or

plant cell) including more than about 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight (e.g. more than 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight), included are lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) selected from the group consisting of a fatty acid, wax, sterol, vitamin, mono-glyceride, diglyceride, triglyceride, phospholipid, glycerolipid, glycerophospholipid, sphingolipid, saccharolipid, polyketide, sterol lipid, triacylglyceride, wax ester, fatty acid ethyl ester, fatty acid methyl ester, component of biodiesel, saturated hydrocarbon, unsaturated hydrocarbon, branched hydrocarbon, and a prenol lipid. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a fatty acid. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a wax. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a sterol. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a vitamin. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a monoglyceride. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a diglyceride. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a triglyceride. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a phospholipid. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a glycerophospholipid. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a sphingolipid. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a saccharolipid. In embodiments, the majority lipid, lipid precursor, or oleochemical in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a polyketide.

yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C20:2 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C20:3 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C21:0 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C21:1 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C21:2 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C21:3 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C22:0 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C22:1 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C22:2 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C22:3 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C23:0 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C23:1 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C23:2 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces C23:3 fatty acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces a fatty acid described herein above at a greater level (e.g. 1.1, 1.2, 1.3, 1.4, 1.5, 1.6, 1.7, 1.8, 1.9, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 200, 300, 400, 500, 600, 700, 800, 900, 1000, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, 10000, 100000, 1000000 fold) compared to the same oleaginous organism lacking the genetic modification. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces a lipid including a fatty acid selected from the group consisting of C5:0, C5:1, C5:2, C5:3, C6:0, C6:1, C6:2, C6:3, C7:0, C7:1, C7:2, C7:3, C8:0, C8:1, C8:2, C8:3, C9:0, C9:1, C9:2, C9:3, C10:0, C10:1, C10:2, C10:3, C11:0, C11:1, C11:2, C11:3, C12:0, C12:1, C12:2, C12:3, C13:0, C13:1, C13:2, C13:3, C14:0, C14:1, C14:2, C14:3, C15:0, C15:1, C15:2, C15:3, C16:0, C16:1, C16:2, C16:3, C17:0, C17:1, C17:2, C17:3, C18:0, C18:1, C18:2, C18:3, C19:0, C19:1, C19:2, C19:3, C20:0, C20:1, C20:2, C20:3, C21:0, C21:1, C21:2, C21:3, C22:0, C22:1, C22:2, C22:3, C23:0, C23:1, C23:2, and C23:3. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces a lipid derived from an endogenously produced fatty acid selected from the group consisting of C5:0, C5:1, C5:2, C5:3, C6:0, C6:1, C6:2, C6:3, C7:0, C7:1, C7:2, C7:3, C8:0, C8:1, C8:2, C8:3, C9:0, C9:1, C9:2, C9:3, C10:0, C10:1, C10:2, C10:3, C11:0, C11:1, C11:2, C11:3,

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C12:0, C12:1, C12:2, C12:3, C13:0, C13:1, C13:2, C13:3, C14:0, C14:1, C14:2, C14:3, C15:0, C15:1, C15:2, C15:3, C16:0, C16:1, C16:2, C16:3, C17:0, C17:1, C17:2, C17:3, C18:0, C18:1, C18:2, C18:3, C19:0, C19:1, C19:2, C19:3, C20:0, C20:1, C20:2, C20:3, C21:0, C21:1, C21:2, C21:3, C22:0, C22:1, C22:2, C22:3, C23:0, C23:1, C23:2, and C23:3. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces a lipid, lipid precursor, or oleochemical (e.g. fatty acid) described herein at a greater level (e.g. 1.1, 1.2, 1.3, 1.4, 1.5, 1.6, 1.7, 1.8, 1.9, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 200, 300, 400, 500, 600, 700, 800, 900, 1000, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, 10000, 100000, 1000000 fold) compared to the same oleaginous organism lacking the genetic modification.

In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a recombinant nucleic acid, wherein the recombinant nucleic acid modulates the level of activity of a protein in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) relative to the absence of the recombinant nucleic acid. In embodiments, the protein is selected from the group consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA:diacylglycerol acyltransferase (DGA1), acyl-CoA:diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Aconitase (ACO1), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), and IRC20 E3 ubiquitin-protein ligase and helicase (IRC20). In embodiments, the protein is Leucine Biosynthesis Gene (LEU2). In embodiments, the protein is Uracil Biosynthesis gene (URA3). In embodiments, the protein is multifunctional enzyme (MFE1). In embodiments, the protein is Transcription Factor (PEX10). In embodiments, the protein is AMP Deaminase (AMPD). In embodiments, the protein is ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2). In embodiments, the protein is Malic Enzyme (MAE). In embodiments, the protein is Acetyl-CoA Carboxylase (ACC). In embodiments, the protein is acyl-CoA: diacylglycerol acyltransferase (DGA1). In embodiments, the protein is acyl-CoA:diacylglycerol acyltransferases (DGA2). In embodiments, the protein is Mitochondrial 2' O-ribose methyltransferase (MRM2). In embodiments, the protein is Lipid synthesis regulator (MGA2). In embodiments, the protein is Chromatin assembly gene (RLF2 subunit p90). In embodiments, the protein is O-6-methylguanine-DNA methyltransferase (MGMT). In embodiments, the protein is Aconitase (ACO1). In embodiments, the protein is Citrate Synthase (CIT1). In embodiments, the protein is RME1 zinc-finger transcription factor (RME1). In embodiments, the protein is YOX1 homeodo-

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main protein (YOX1). In embodiments, the protein is UGA2 succinate semialdehyde dehydrogenase (UGA2). In embodiments, the protein is OSH6 oxysterol-binding protein homolog 6 (OSH6). In embodiments, the protein is IRC20 E3 ubiquitin-protein ligase and helicase (IRC20). In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is modulating the function of the protein. In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is modulating the amount of the protein. In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is modulating the transcription of the mRNA encoding the protein. In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is modulating the translation of the protein. In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is modulating the coding sequence of the gene encoding the protein (e.g. mutating (e.g. point mutant or missense mutant), truncating, inserting into, or deleting). In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is modulating the regulatory elements (e.g. promoter) of the gene encoding the protein. In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is modulating the stability of the protein. In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is modulating the stability of the transcript encoding the protein. In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is reducing the level of activity of the protein. In embodiments, modulating the level of activity of a protein in an oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is increasing the level of activity of the protein. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein in the citric acid cycle in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in the Kennedy Pathway in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in fatty acid synthesis in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modifica-

tion (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in fatty acid storage (e.g. accumulation) in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in lipid synthesis in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in lipid storage (e.g. accumulation) in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in triacylglyceride storage (e.g. accumulation) in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in triacylglyceride synthesis in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in the beta-oxidation cycle in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in fatty acid degradation in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in lipid degradation in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in triacylglyceride degradation in the oleaginous organism relative to the

absence of the genetic modification (e.g. recombinant nucleic acid). In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a genetic modification (e.g. recombinant nucleic acid) that modulates (e.g. reduces or increases) the level of activity of a protein involved in central carbon metabolism in the oleaginous organism relative to the absence of the genetic modification (e.g. recombinant nucleic acid).

In embodiments, the recombinant nucleic acid increases the level of activity of a protein in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the protein is selected from the group consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA:diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), and IRC20 E3 ubiquitin-protein ligase and helicase (IRC20). In embodiments, the protein is selected from the group consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA:diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), and Citrate Synthase (CIT1). In embodiments, the protein is Leucine Biosynthesis Gene (LEU2). In embodiments, the protein is Uracil Biosynthesis gene (URA3). In embodiments, the protein is AMP Deaminase (AMPD). In embodiments, the protein is ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2). In embodiments, the protein is Malic Enzyme (MAE). In embodiments, the protein is Acetyl-CoA Carboxylase (ACC). In embodiments, the protein is acyl-CoA:diacylglycerol acyltransferase (DGA1). In embodiments, the protein is acyl-CoA:diacylglycerol acyltransferases (DGA2). In embodiments, the protein is Mitochondrial 2' O-ribose methyltransferase (MRM2). In embodiments, the protein is Lipid synthesis regulator (MGA2). In embodiments, the protein is Chromatin assembly gene (RLF2 subunit p90). In embodiments, the protein is O-6-methylguanine-DNA methyltransferase (MGMT). In embodiments, the protein is Citrate Synthase (CIT1). In embodiments, the protein is RME1 zinc-finger transcription factor (RME1). In embodiments, the protein is YOX1 homeodomain protein (YOX1). In embodiments, the protein is UGA2 succinate semialdehyde dehydrogenase (UGA2). In embodiments, the protein is OSH6 oxysterol-binding protein homolog 6 (OSH6). In embodiments, the protein is IRC20 E3 ubiquitin-protein ligase and helicase (IRC20). In embodiments, the protein is selected from the group consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), Malic Enzyme (MAE), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid

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synthesis regulator (MGA2), and O-6-methylguanine-DNA methyltransferase (MGMT) or said nucleic acid decreases the level of activity of Lipid synthesis regulator (MGA2).

In embodiments, the genetic modification (e.g. recombinant nucleic acid) decreases the level of activity of a protein in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the protein is selected from the group consisting of multifunctional enzyme (MFE1), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), Transcription Factor (PEX10), and Aconitase (ACO1). In embodiments, the protein is multifunctional enzyme (MFE1). In embodiments, the protein is Lipid synthesis regulator (MGA2). In embodiments, the protein is Chromatin assembly gene (RLF2 subunit p90). In embodiments, the protein is Transcription Factor (PEX10). In embodiments, the protein is Aconitase (ACO1). In embodiments, the protein is RME1 zinc-finger transcription factor (RME1). In embodiments, the protein is YOX1 homeodomain protein (YOX1). In embodiments, the protein is UGA2 succinate semialdehyde dehydrogenase (UGA2). In embodiments, the protein is OSH6 oxysterol-binding protein homolog 6 (OSH6). In embodiments, the protein is IRC20 E3 ubiquitin-protein ligase and helicase (IRC20).

In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a plurality of genetic modifications (e.g. recombinant nucleic acids) that collectively modulate one, two, three, four, five, six, seven, eight, nine, ten, or more of the group of proteins consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Aconitase (ACO1), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), and IRC20 E3 ubiquitin-protein ligase and helicase (IRC20).

In embodiments, the recombinant nucleic acid encodes a protein comprising a mutation relative to the wildtype protein. In embodiments, the mutation is a point mutation. In embodiments, the mutation is a deletion. In embodiments, the mutation is an insertion. In embodiments, the mutation is a fusion with a second protein. In embodiments, the recombinant nucleic acid encodes a mutant of a protein selected from the group consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Aconitase (ACO1), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase

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(UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), or IRC20 E3 ubiquitin-protein ligase and helicase (IRC20).

In embodiments, the recombinant nucleic acid encodes a mutant of a protein selected from the group consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Aconitase (ACO1), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), or IRC20 E3 ubiquitin-protein ligase and helicase (IRC20).

In embodiments, the recombinant nucleic acid is an AMP Deaminase (AMPD) having the nucleotide sequence of SEQ ID NO.:33. In embodiments, the recombinant nucleic acid is an AMP Deaminase (AMPD) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, or the entire sequence) with SEQ ID NO.:33, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Leucine Biosynthesis Gene (LEU2) having the nucleotide sequence of SEQ ID NO.:35. In embodiments, the recombinant nucleic acid is a Leucine Biosynthesis Gene (LEU2) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, or the entire sequence) with SEQ ID NO.:35, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Uracil Biosynthesis gene (URA3) having the nucleotide sequence of SEQ ID NO.:37. In embodiments, the recombinant nucleic acid is a Uracil Biosynthesis gene (URA3) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, or the entire sequence) with SEQ ID NO.:37, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is an ATP-Citrate Lyase (ACL) (subunit 1) having the nucleotide sequence of SEQ ID NO.:39. In embodiments, the recombinant nucleic acid is an ATP-Citrate Lyase (ACL) (subunit 1) having at least 60% identity (e.g. at least 61%, 62%, 63%,

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64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, or the entire sequence) with SEQ ID NO.:39, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is an ATP-Citrate Lyase (ACL) (subunit 2) having the nucleotide sequence of SEQ ID NO.:41. In embodiments, the recombinant nucleic acid is an ATP-Citrate Lyase (ACL) (subunit 2) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, or the entire sequence) with SEQ ID NO.:41, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Malic Enzyme (MEA, MAE, MEA1) having the nucleotide sequence of SEQ ID NO.:43. In embodiments, the recombinant nucleic acid is a Malic Enzyme (MEA, MAE, MEA1) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, or the entire sequence) with SEQ ID NO.:43, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a acyl-CoA:diacylglycerol acyltransferase (DGA1) having the nucleotide sequence of SEQ ID NO.:45. In embodiments, the recombinant nucleic acid is a acyl-CoA: diacylglycerol acyltransferase (DGA1) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, or the entire sequence) with SEQ ID NO.:45, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a acyl-CoA:diacylglycerol acyltransferase (DGA2) having the nucleotide sequence of SEQ ID NO.:47. In embodiments, the recombinant nucleic acid is a acyl-CoA:diacylglycerol acyltransferase (DGA2) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, or the entire sequence) with SEQ ID NO.:47, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Lipid synthesis regulator (MGA2) having the nucleotide sequence

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of SEQ ID NO.:49. In embodiments, the recombinant nucleic acid is a Lipid synthesis regulator (MGA2) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 2100, 2200, 2300, 2400, 2500, 2600, 2700, 2800, 2900, 3000, 3100, 3200, 3300, 3400, 3500, 3600, 3700, 3800, or the entire sequence) with SEQ ID NO.:49, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a mutant Lipid synthesis regulator (MGA2-L36 mutant) having the nucleotide sequence of SEQ ID NO.:51. In embodiments, the recombinant nucleic acid is a mutant Lipid synthesis regulator (MGA2-L36 mutant) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 2100, 2200, 2300, 2400, 2500, 2600, 2700, 2800, 2900, 3000, 3100, 3200, 3300, 3400, 3500, 3600, 3700, 3800, or the entire sequence) with SEQ ID NO.:51, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a truncated Lipid synthesis regulator (MGA2-truncated) having the nucleotide sequence of SEQ ID NO.:53. In embodiments, the recombinant nucleic acid is a truncated Lipid synthesis regulator (MGA2-truncated) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 2100, 2200, 2300, 2400, 2500, 2600, 2700, 2800, 2900, 3000, 3100, 3200, 3300, 3400, 3500, 3600, 3700, 3800, or the entire sequence) with SEQ ID NO.:53, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Chromatin assembly gene (RLF2 subunit p90) having the nucleotide sequence of SEQ ID NO.:58. In embodiments, the recombinant nucleic acid is a Chromatin assembly gene (RLF2 subunit p90) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, or the entire sequence) with SEQ ID NO.:58, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Mitochondrial 2' O-ribose methyltransferase (MRM2) having the nucleotide sequence of SEQ ID NO.:63. In embodiments, the recombinant nucleic acid is a Mitochondrial 2' O-ribose methyltransferase (MRM2) having at least 60% identity

(e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, or the entire sequence) with SEQ ID NO.:63, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Citrate Synthase (CIT1) having the nucleotide sequence of SEQ ID NO.:67. In embodiments, the recombinant nucleic acid is a Citrate Synthase (CIT1) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, or the entire sequence) with SEQ ID NO.:67, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Acetyl-CoA Carboxylase (ACC) having the nucleotide sequence of SEQ ID NO.:69. In embodiments, the recombinant nucleic acid is a Acetyl-CoA Carboxylase (ACC) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, or the entire sequence) with SEQ ID NO.:69, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Transcription Factor (PEX10) having the nucleotide sequence of SEQ ID NO.:71. In embodiments, the recombinant nucleic acid is a Transcription Factor (PEX10) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 3000, 4000, 5000, 6000, 7000, or the entire sequence) with SEQ ID NO.:71, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a multifunctional enzyme (MFE1) having the nucleotide sequence of SEQ ID NO.:73. In embodiments, the recombinant nucleic acid is a multifunctional enzyme (MFE1) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 2100, 2200, 2300, or the entire sequence) with SEQ ID NO.:73, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a Aconitase (ACO1) having the nucleotide sequence of SEQ ID NO.:75. In embodiments, the recombinant nucleic acid is a Aconitase (ACO1) having at least 60% identity (e.g. at least

61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 2100, 2200, 2300, 2400, 2500, 2600, or the entire sequence) with SEQ ID NO.:75, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a YOX1 homeodomain protein (YOX1) having the nucleotide sequence of SEQ ID NO.:77. In embodiments, the recombinant nucleic acid is a YOX1 homeodomain protein (YOX1) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 2100, 2200, 2300, 2400, 2500, 2600, or the entire sequence) with SEQ ID NO.:77, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a UGA2 succinate semialdehyde dehydrogenase (UGA2) having the nucleotide sequence of SEQ ID NO.:78. In embodiments, the recombinant nucleic acid is a UGA2 succinate semialdehyde dehydrogenase (UGA2) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, or the entire sequence) with SEQ ID NO.:78, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a RME1 zinc-finger transcription factor (RME1) having the nucleotide sequence of SEQ ID NO.:79. In embodiments, the recombinant nucleic acid is a RME1 zinc-finger transcription factor (RME1) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, or the entire sequence) with SEQ ID NO.:79, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a OSH6 oxysterol-binding protein homolog 6 (OSH6) having the nucleotide sequence of SEQ ID NO.:80. In embodiments, the recombinant nucleic acid is a OSH6 oxysterol-binding protein homolog 6 (OSH6) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, or the entire sequence) with SEQ ID NO.:80, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) having the

nucleotide sequence of SEQ ID NO.:81. In embodiments, the recombinant nucleic acid is a IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 3000, 4000, 5000, or the entire sequence) with SEQ ID NO.:81, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the recombinant nucleic acid is a O-6-methylguanine-DNA methyltransferase (MGMT) having the nucleotide sequence of SEQ ID NO.:65. In embodiments, the recombinant nucleic acid is a O-6-methylguanine-DNA methyltransferase (MGMT) having at least 60% identity (e.g. at least 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identity) over a region of at least 100 nucleotides (e.g. at least 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, or the entire sequence) with SEQ ID NO.:65, (e.g. using the same length of nucleotides for comparison or the entirety of both nucleic acids).

In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a recombinant nucleic acid that decreases the level of activity of multifunctional enzyme (MFE1) protein and Transcription Factor (PEX10) protein, increases the level of activity of acyl-CoA: diacylglycerol acyltransferase (DGA1) protein, or increases the level of activity of Leucine Biosynthesis Gene (LEU2) protein relative to a oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) that does not include the recombinant nucleic acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes recombinant nucleic acids that decrease the level of activity of multifunctional enzyme (MFE1) protein and Transcription Factor (PEX10) protein, increase the level of activity of acyl-CoA:diacylglycerol acyltransferase (DGA1) protein, and increase the level of activity of Leucine Biosynthesis Gene (LEU2) protein relative to a oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) that does not include the recombinant nucleic acids. In embodiments, the level of activity is the level of expression of the protein.

In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes an extra-chromosomal recombinant nucleic acid. In embodiments, the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes a recombinant nucleic acid integrated into the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) genome. In embodiments, the extra-chromosomal recombinant nucleic acid includes a gene that is also included in the genome of the yeast cell oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) (e.g. Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), IRC20 E3 ubiquitin-protein ligase and helicase (IRC20), a wildtype version thereof, or a mutant version thereof). In embodiments, the extra-chromosomal recombinant nucleic acid includes a gene that is also included in the genome of the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) (e.g. Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL) (e.g. ACL subunit 1, ACL subunit 2, or subunit 1 and 2), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), O-6-methylguanine-DNA methyltransferase (MGMT), Citrate Synthase (CIT1), RME1 zinc-finger transcription factor (RME1), YOX1 homeodomain protein (YOX1), UGA2 succinate semialdehyde dehydrogenase (UGA2), OSH6 oxysterol-binding protein homolog 6 (OSH6), IRC20 E3 ubiquitin-protein ligase and helicase (IRC20), a wildtype version thereof, or a mutant version thereof). In embodiments, a recombinant nucleic acid integrated into the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) genome replaces (e.g. partially or completely) a promoter included in the oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) genome prior to integration of the recombinant nucleic acid.

In embodiments, the yeast cell is a yeast cell including one or more genetic modifications (e.g. recombinant nucleic acids), as described herein (including in the Examples section below, the tables, the figures, and the claims herein). In embodiments, the yeast cell is a yeast cell described herein, including in an example, table, figure, or claim. In embodiments, the oleaginous yeast cell is L36 as described herein (e.g. examples, tables, and figures). In embodiments, the oleaginous yeast cell is derived from L36 as described herein (e.g. examples, tables, and figures). In embodiments, the oleaginous yeast cell is E26 as described herein (e.g. examples, tables, and figures). In embodiments, the oleaginous yeast cell is E13 as described herein (e.g. examples, tables, and figures). In embodiments, the oleaginous yeast cell is derived from E26 or E13.

In embodiments, the dry weight of the genetically modified yeast cell described herein includes greater than about 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., greater than about 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99%; greater than 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88,

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89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99%; of lipid; lipids; lipid precursors; lipid precursor, oleochemical, and/or oleochemicals).

In embodiments, the genetically modified yeast cell described herein includes a recombinant Leucine Biosynthesis Gene (LEU2). In embodiments, the genetic modification increases the level of activity of the Leucine Biosynthesis Gene (LEU2) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein is capable of de novo synthesis of leucine (e.g. at sufficient levels to meet the leucine requirements of the yeast cell). In embodiments, the genetically modified yeast cell described herein is capable of de novo synthesis of leucine independent of the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant Uracil Biosynthesis gene (URA3). In embodiments, the genetic modification increases the level of activity of the Uracil Biosynthesis gene (URA3) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein is capable of de novo synthesis of uracil (e.g. at sufficient levels to meet the uracil requirements of the yeast cell). In embodiments, the genetically modified yeast cell described herein is capable of de novo synthesis of uracil independent of the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a genetically modified multifunctional enzyme (MFE1) gene. In embodiments, the genetic modification decreases the level of activity of the multifunctional enzyme (MFE1) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant AMP Deaminase (AMPD) protein. In embodiments, the genetic modification increases the level of activity of the AMP Deaminase (AMPD) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant ATP-Citrate Lyase 1 (ACL1) protein. In embodiments, the genetic modification increases the level of activity of the ATP-Citrate Lyase 1 (ACL1) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant ATP-Citrate Lyase 2 (ACL2) protein. In embodiments, the genetic modification increases the level of activity of the ATP-Citrate Lyase 2 (ACL2) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant ATP-Citrate Lyase 1 (ACL1) protein and ATP-Citrate Lyase 2 (ACL2) protein. In embodiments, the genetic modification increases the level of activity of the ATP-Citrate Lyase 1 (ACL1) protein and ATP-Citrate Lyase 2 (ACL2) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant Malic Enzyme (MAE) protein. In embodiments, the genetic modification increases the level of activity of the Malic Enzyme (MAE) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments,

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the genetically modified yeast cell described herein includes a recombinant Acetyl-CoA Carboxylase (ACC) protein. In embodiments, the genetic modification increases the level of activity of the Acetyl-CoA Carboxylase (ACC) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant acyl-CoA:diacylglycerol acyltransferase 1 (DGA1) protein. In embodiments, the genetic modification increases the level of activity of the acyl-CoA:diacylglycerol acyltransferase 1 (DGA1) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant acyl-CoA:diacylglycerol acyltransferase 2 (DGA2) protein. In embodiments, the genetic modification increases the level of activity of the acyl-CoA:diacylglycerol acyltransferase 2 (DGA2) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant Mitochondrial 2' O-ribose methyltransferase (MRM2) protein. In embodiments, the genetic modification increases the level of activity of the Mitochondrial 2' O-ribose methyltransferase (MRM2) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant Lipid synthesis regulator (MGA2) protein. In embodiments, the genetically modified yeast cell described herein includes a genetically modified Lipid synthesis regulator (MGA2) gene. In embodiments, the genetically modified yeast cell described herein includes at least one nucleotide deletion in the genomic Lipid synthesis regulator (MGA2) gene and expression of a Lipid synthesis regulator (MGA2) protein including a mutation corresponding to G643R in *Yarrowia lipolytica* Lipid synthesis regulator (MGA2). In embodiments, the genetic modification decreases the level of activity of the Lipid synthesis regulator (MGA2) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a genetically modified Chromatin assembly gene (RLF2 subunit p90) gene. In embodiments, the genetic modification decreases the level of activity of the Chromatin assembly gene (RLF2 subunit p90) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant O-6-methylguanine-DNA methyltransferase (MGMT) protein. In embodiments, the genetic modification increases the level of activity of the O-6-methylguanine-DNA methyltransferase (MGMT) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a genetically modified Aconitase (ACO1) gene. In embodiments, the genetic modification decreases the level of activity of the Aconitase (ACO1) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant Citrate Synthase (CIT1) gene. In embodiments, the genetic modification increases the level of activity of the Citrate Synthase (CIT1) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a recombinant RME1 zinc-finger transcription factor (RME1) gene. In embodiments, the genetic modification decreases the level of activity of the RME1 zinc-finger transcription factor (RME1) protein relative to an otherwise

identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a genetically modified YOX1 homeodomain protein (YOX1) gene. In embodiments, the genetic modification decreases the level of activity of the YOX1 homeodomain protein (YOX1) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a genetically modified UGA2 succinate semialdehyde dehydrogenase (UGA2) gene. In embodiments, the genetic modification decreases the level of activity of the UGA2 succinate semialdehyde dehydrogenase (UGA2) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a genetically modified OSH6 oxysterol-binding protein homolog 6 (OSH6) gene. In embodiments, the genetic modification decreases the level of activity of the OSH6 oxysterol-binding protein homolog 6 (OSH6) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the genetically modified yeast cell described herein includes a genetically modified IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) gene. In embodiments, the genetic modification decreases the level of activity of the IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) protein relative to an otherwise identical yeast cell lacking the genetic modification. In embodiments, the gene or protein described herein is a *Yarrowia lipolytica* gene or protein. In embodiments, the gene or protein is a yeast gene or protein corresponding to the *Yarrowia lipolytica* gene or protein. In embodiments, the gene or protein is a gene or protein from an oleaginous organism corresponding to the *Yarrowia lipolytica* gene or protein. In embodiments, the gene or protein is the *Yarrowia lipolytica* gene or protein identified by sequence herein. In embodiments, the gene or protein is a mutant gene or protein of a wildtype gene or protein corresponding to the *Yarrowia lipolytica* gene or protein. In embodiments, the gene or protein is a mutant gene or protein of a wildtype yeast gene or protein corresponding to the *Yarrowia lipolytica* gene or protein. In embodiments, the gene or protein is a homolog of the *Yarrowia lipolytica* gene or protein. In embodiments, the gene or protein is a homolog of the *Yarrowia lipolytica* gene or protein identified by sequence herein. In embodiments, the gene or protein is a mutant of the *Yarrowia lipolytica* gene or protein. In embodiments, the gene or protein described in this paragraph is LEU2, URA3, MFE1, PEX10, AMPD, ACL, ACL1, ACL2, MAE, ACC, DGA, DGA1, DGA2, MRM2, MGA2, RLF2 subunit p90, MGMT, ACO1, CIT1, RME1, YOX1, UGA2, OSH6, or IRC20). In embodiments, the gene or protein described in this paragraph is LEU2, URA3, MFE1, PEX10, AMPD, ACL, ACL1, ACL2, MAE, ACC, DGA, DGA1, DGA2, MRM2, MGA2, RLF2 subunit p90, MGMT, ACO1, CIT1, RME1, YOX1, UGA2, OSH6, or IRC20), having the sequence identified herein.

In embodiments, the genetic modification modulates the level of activity of a component of a lipid biosynthetic pathway. In embodiments, the genetic modification modulates the level of activity of a component of a lipid precursor biosynthetic pathway. In embodiments, the genetic modification modulates the level of activity of a component of an oleochemical biosynthetic pathway. In embodiments, the genetic modification modulates the level of activity of a component of a pathway incorporating Acetyl-CoA into a lipid, lipid precursor, or oleochemical. In embodiments, the genetic modification modulates the level of activity of a component of a pathway incorporating malonyl-CoA into a

lipid, lipid precursor, or oleochemical. In embodiments, the genetic modification increases the level of activity of a component of a lipid biosynthetic pathway. In embodiments, the genetic modification increases the level of activity of a component of a lipid precursor biosynthetic pathway. In embodiments, the genetic modification increases the level of activity of a component of an oleochemical biosynthetic pathway. In embodiments, the genetic modification increases the level of activity of a component of a pathway incorporating acetyl-CoA into a lipid, lipid precursor, or oleochemical. In embodiments, the genetic modification increases the level of activity of a component of a pathway incorporating malonyl-CoA into a lipid, lipid precursor, or oleochemical. In embodiments, the genetic modification modulates the level of activity of a component of a lipid, or lipid precursor, metabolic pathway. In embodiments, the genetic modification decreases the level of activity of a component of a lipid, or lipid precursor, metabolic pathway. In embodiments, the genetic modification decreases the level of activity of a component of a lipid, or lipid precursor, metabolic pathway. In embodiments, the genetic modification increases the level of acetyl-CoA in the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) relative to a genetically unmodified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the genetic modification increases the level of malonyl-CoA in the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) relative to a genetically unmodified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the genetic modification increases the level of triglyceride production in the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) relative to a genetically unmodified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) that is otherwise identical (e.g. genetically) to the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the genetic modification decreases the level of beta-oxidation activity in the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) relative to a genetically unmodified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) that is otherwise identical (e.g. genetically) to the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the genetic modification decreases the level of fatty acid catabolism in the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) relative to a genetically unmodified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) that is otherwise identical (e.g. genetically) to the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the genetic modification decreases the level of peroxisome biogenesis activity in the genetically modified

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oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) relative to a genetically unmodified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) that is otherwise identical (e.g. genetically) to the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the genetic modification produces a lipid, lipid precursor, or oleochemical at a higher level than by a genetically unmodified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) that is otherwise identical to the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) produces a lipid, lipid precursor, or oleochemical at a higher level than by a genetically unmodified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) that is otherwise identical (e.g. genetically) to the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the genetic modification modulates the level of activity of a component of the citric acid cycle. In embodiments, the genetic modification modulates the level of activity of a component of the TCA cycle. In embodiments, the genetic modification modulates the level of activity of a component of the Kennedy pathway. In embodiments, the genetic modification reduces the level of activity of the TCA cycle. In embodiments, the genetic modification increases the level of activity of the Kennedy pathway.

In embodiments, the lipid, lipid precursor, or oleochemical produced at a higher level by the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) is a fatty acid, wax, sterol, vitamin, monoglyceride, diglyceride, triglyceride, phospholipid, glycerolipid, glycerophospholipid, sphingolipid, saccharolipid, polyketide, sterol lipid, triacylglyceride, prenol lipid, fatty acid ester, fatty acid methyl ester, fatty acid ethyl ester, fatty acid propyl ester, fatty acid butyl ester, fatty alcohol, fatty amine, glycerol, alcohol ethoxylate, alcohol sulfate, or alcohol ether sulfate. In embodiments, the genetic modification includes a mutation relative to the wild type gene. In embodiments, the genetic modification includes a deletion of a portion of a gene. In embodiments, the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) includes an increased level of a fatty acid selected from the group consisting of C5:0, C5:1, C5:2, C5:3, C6:0, C6:1, C6:2, C6:3, C7:0, C7:1, C7:2, C7:3, C8:0, C8:1, C8:2, C8:3, C9:0, C9:1, C9:2, C9:3, C10:0, C10:1, C10:2, C10:3, C11:0, C11:1, C11:2, C11:3, C12:0, C12:1, C12:2, C12:3, C13:0, C13:1, C13:2, C13:3, C14:0, C14:1, C14:2, C14:3, C15:0, C15:1, C15:2, C15:3, C16:0, C16:1, C16:2, C16:3, C17:0, C17:1, C17:2, C17:3, C18:0, C18:1, C18:2, C18:3, C19:0, C19:1, C19:2, C19:3, C20:0, C20:1, C20:2, C20:3, C21:0, C21:1, C21:2, C21:3, C22:0, C22:1, C22:2, C22:3, C23:0, C23:1, C23:2, and C23:3, relative to a genetically unmodified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell) that is otherwise identical (e.g. genetically) to the genetically modified oleaginous organism (e.g. yeast cell, oleaginous yeast cell, *Yarrowia lipolytica*, algae, or plant cell). In embodiments, the fatty acid is C17:0 C17:1. In embodiments, the fatty acid is C16:1n9.

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In embodiments, the genetic modification is an engineered genetic modification. In embodiments, the engineered genetic modification includes modulated expression of a protein. In embodiments, the engineered genetic modification includes increased expression of a protein. In embodiments, the engineered genetic modification includes decreased expression of a protein. In embodiments, the genetic modification is associated with exposure to a mutagen. In embodiments, the genetic modification includes modulated expression of a protein in a lipid, or lipid precursor, or oleochemical biosynthetic pathway.

III. METHODS OF MAKING AND PURIFYING LIPIDS, LIPID PRECURSORS, AND/OR OLEOCHEMICALS

Lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) produced by cells of the invention can be harvested, or otherwise collected, by any convenient method (e.g. centrifugation of extracellular secreted lipids, exposure to solvent, whole cell extraction (e.g. cell disruption and collection), hydrophobic solvent extraction (e.g. hexane), liquefaction, supercritical carbon dioxide extraction, freeze drying, mechanical pulverization, secretion (e.g. by addition of effective exporter proteins), or combinations thereof).

In embodiments, reduced nitrogen conditions promote accumulation of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical). In embodiments, cells (e.g. oleaginous organisms or oleaginous yeast) are first cultured in standard conditions and then cultured in low nitrogen conditions where harvesting is desired. In embodiments, oleaginous yeast species are grown in a medium including a carbon substrate and/or nitrogen source, optionally in the absence of light, optionally in an aerobic environment. In embodiments, media for culturing oleaginous yeast may include a carbon substrate, a fixed nitrogen source, trace elements, a buffer for pH maintenance, phosphate, or a combination thereof.

In embodiments, the carbon substrate may be selected from the group consisting of monosaccharides, oligosaccharides, polysaccharides, alkanes, fatty acids, esters of fatty acids, monoglycerides, carbon dioxide, methanol, formaldehyde, formate, carbon-containing amines, glucose, fructose, sucrose, lactose, galactose, xylose, mannose, rhamnose, arabinose, glycerol, acetate, depolymerized sugar beet pulp, black liquor, corn starch, depolymerized cellulosic material, corn stover, sugar beet pulp, switchgrass, milk whey, molasses, potato, rice, sorghum, sugar cane, wheat, thick cane juice, sugar beet juice, wheat, lignocellulosic biomass, and combinations thereof.

Examples of cellulosic material that may be depolymerized and used as a carbon substrate (e.g. fixed carbon source) include sugarcane bagasse, rice hulls, corn fiber (including stalks, leaves, husks, and cobs), wheat straw, rice straw, sugar beet pulp, citrus pulp, citrus peels; hardwood and softwood thinnings; hardwood and softwood residues; saw mill wastes (wood chips, sawdust) and pulp mill waste; paper fractions of municipal solid waste; municipal grass clippings; wood construction waste; and cellulosic crops such as switchgrass, hybrid poplar wood, and *miscanthus*, fiber cane, and fiber sorghum.

Oleaginous yeast cultures may yield oleaginous yeast biomass in fermentation media. To extract lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical)

from the biomass, the biomass may be harvested, concentrated, dewatered (i.e. separation of the biomass from the liquid medium) (e.g. through centrifugation, filtration, use of mechanical pressure, simple sedimentation, or sedimentation), or combinations thereof. Centrifugation does not always remove significant amounts of intracellular water from the oleaginous yeast and so is often considered a dewatering, not a drying, step. The biomass can optionally be dried (oven dried, lyophilized, and the like) and conditioned prior to cell disruption (lysis).

In a second aspect is provided a method of producing a lipid, lipid precursor, or oleochemical (e.g., lipid, lipid precursor, oleochemical) including: 1) culturing a yeast cell as described herein (including embodiments or as described in the examples, tables, figures, and/or claims) in a growth medium; and 2) isolating the lipid, lipid precursor, or oleochemical (e.g., lipid, lipid precursor, oleochemical) (e.g. from the medium or yeast cell).

In embodiments, the lipid, lipid precursor, or oleochemical (e.g., lipid, lipid precursor, oleochemical) is isolated from the yeast cell. In embodiments, the lipid, lipid precursor, or oleochemical (e.g., lipid, lipid precursor, oleochemical) is isolated from the medium. In embodiments, the growth medium includes a majority carbon source selected from the group consisting of glucose, glycerol, xylose, fructose, mannose, ribose, sucrose, and lignocellulosic biomass. In embodiments, the majority carbon source is glucose. In embodiments, the majority carbon source is glycerol. In embodiments, the majority carbon source is xylose. In embodiments, the majority carbon source is fructose. In embodiments, the majority carbon source is mannose. In embodiments, the majority carbon source is ribose. In embodiments, the majority carbon source is sucrose. In embodiments, the majority carbon source is lignocellulosic biomass. In embodiments, the carbon source is glucose. In embodiments, the carbon source is glycerol. In embodiments, the carbon source is xylose. In embodiments, the carbon source is fructose. In embodiments, the carbon source is mannose. In embodiments, the carbon source is ribose. In embodiments, the carbon source is sucrose. In embodiments, the carbon source is lignocellulosic biomass. In embodiments, the majority carbon source is not glucose. In embodiments, the majority nitrogen source is ammonium sulfate ($(\text{NH}_4)_2\text{SO}_4$).

In embodiments, the growth medium includes a carbon source and a nitrogen source wherein the carbon source is at a concentration at least 2-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 3-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 4-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 5-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 6-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 7-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 8-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 9-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 10-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 11-fold greater than the concentration of the nitrogen source. In embodiments, the carbon

source is at a concentration at least 12-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 13-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 14-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 15-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 16-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 17-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 18-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 19-fold greater than the concentration of the nitrogen source. In embodiments, the carbon source is at a concentration at least 20-fold greater than the concentration of the nitrogen source. In embodiments, the ratio of the carbon source to the nitrogen source (wt/wt) is about 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40. In embodiments, the ratio of the carbon source to the nitrogen source (wt/wt) is 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40. In embodiments, the ratio of the carbon source to the nitrogen source (wt/wt) is about 0.03125, 0.0625, 0.125, 0.25, 0.5, 1.0, 2.0, 4.0, 8.0, 16, 32, 64, 128, 256, 512, 1024, 1600, 2048, 4096, 8192, or 16284. In embodiments, the ratio of the carbon source to the nitrogen source (wt/wt) is about 0.001, 0.002, 0.003, 0.004, 0.005, 0.006, 0.007, 0.008, 0.009, 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1.0, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 250, 300, 350, 400, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, or 10000. In embodiments, the ratio of the carbon source to the nitrogen source (wt/wt) is 0.001, 0.002, 0.003, 0.004, 0.005, 0.006, 0.007, 0.008, 0.009, 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1.0, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 250, 300, 350, 400, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, or 10000. In embodiments, the ratio of the carbon source to the nitrogen source (wt/wt) is at least 0.001, 0.002, 0.003, 0.004, 0.005, 0.006, 0.007, 0.008, 0.009, 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1.0, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 250, 300, 350, 400, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000, 2000, 3000, 4000, 5000, 6000, 7000, 8000, 9000, or 10000. In embodiments, the carbon source to nitrogen source ratio corresponds to a ratio calculated from one or more of the ratios described above when the ratios described above are for a carbon source of glucose (g/L) and a nitrogen source of ammonium sulfate (g/L) for a carbon source that may not be glucose and a nitrogen source that may not be ammonium sulfate. In embodiments, the ratio of the concentration of the carbon source to the concentration of the nitrogen source is as described herein, including in embodiments, examples, tables, figures, and claims. In embodiments, the amount and

ratio of the carbon source to the nitrogen source (wt/wt) is equivalent to 160:0.2 glucose:ammonium sulfate. In embodiments, the amount and ratio of the carbon source to the nitrogen source (wt/wt) is equivalent to 80:5 glucose: ammonium sulfate.

In embodiments, the carbon source is at a concentration (g/L) of about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 210, 220, 230, 240, 250, 260, 270, 280, 290, 300, 310, 320, 330, 340, 350, 360, 370, 380, 390, 100, 410, 420, 430, 440, 450, 460, 470, 480, 490, or 500. In embodiments, the carbon source is at a concentration (g/L) of 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 210, 220, 230, 240, 250, 260, 270, 280, 290, 300, 310, 320, 330, 340, 350, 360, 370, 380, 390, 100, 410, 420, 430, 440, 450, 460, 470, 480, 490, or 500. In embodiments, the carbon source, which is optionally not glucose, is at a concentration for the carbon source that would provide an equal amount of carbon as one of the amounts described above where the amount described above is for glucose.

In embodiments, the nitrogen source is at a concentration (g/L) of about 0.001, 0.002, 0.003, 0.004, 0.005, 0.006, 0.007, 0.008, 0.009, 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100. In embodiments, the nitrogen source is at a concentration (g/L) of 0.001, 0.002, 0.003, 0.004, 0.005, 0.006, 0.007, 0.008, 0.009, 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100. In embodiments, the nitrogen source, which is optionally not ammonium sulfate, is at a concentration for the nitrogen source that would provide an equal amount of nitrogen as one of the amounts described above where the amount described above is for ammonium sulfate.

In embodiments, the growth medium includes a micronutrient. In embodiments, the growth medium includes a plurality of micronutrients. In embodiments, the growth medium includes cobalt, iron, magnesium, potassium, zinc, nickel, molybdenum, manganese, copper, and/or boron. In embodiments, the growth medium includes iron and copper or molybdenum. In embodiments, the growth medium includes copper and nickel. In embodiments, the growth medium includes copper, iron, and either molybdenum or nickel. In embodiments, the growth medium includes copper, iron, molybdenum, and nickel. In embodiments, the

growth medium includes cobalt. In embodiments, the growth medium includes iron. In embodiments, the growth medium includes magnesium. In embodiments, the growth medium includes potassium. In embodiments, the growth medium includes zinc. In embodiments, the growth medium includes nickel. In embodiments, the growth medium includes molybdenum. In embodiments, the growth medium includes manganese. In embodiments, the growth medium includes copper. In embodiments, the growth medium includes boron. In embodiments, the growth medium is supplemented with cobalt, iron, magnesium, potassium, zinc, nickel, molybdenum, manganese, copper, and/or boron. In embodiments, the growth medium is supplemented with iron and copper or molybdenum. In embodiments, the growth medium is supplemented with copper and nickel. In embodiments, the growth medium is supplemented with copper, iron, and either molybdenum or nickel. In embodiments, the growth medium is supplemented with copper, iron, molybdenum, and nickel. In embodiments, the growth medium is supplemented with cobalt. In embodiments, the growth medium is supplemented with iron. In embodiments, the growth medium is supplemented with magnesium. In embodiments, the growth medium is supplemented with potassium. In embodiments, the growth medium is supplemented with zinc. In embodiments, the growth medium is supplemented with nickel. In embodiments, the growth medium is supplemented with molybdenum. In embodiments, the growth medium is supplemented with manganese. In embodiments, the growth medium is supplemented with copper. In embodiments, the growth medium is supplemented with boron. In embodiments, the growth medium includes CoCl_2 at a concentration of about 15 mg/L. In embodiments, the growth medium includes MgSO_4 at a concentration of about 250 mg/L. In embodiments, the growth medium includes KI at a concentration of about 15 mg/L. In embodiments, the growth medium includes $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ at a concentration of about 20 mg/L. In embodiments, the growth medium includes $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ at a concentration of about 12.5 mg/L. In embodiments, the growth medium includes Boric acid at a concentration of about 12.5 mg/L. In embodiments, the growth medium includes $(\text{NH}_4)_2\text{Mo} \cdot 4\text{H}_2\text{O}$ at a concentration of about 15 mg/L. In embodiments, the growth medium includes $\text{NiSO}_4 \cdot 6\text{H}_2\text{O}$ at a concentration of about 12.5 mg/L. In embodiments, the growth medium includes $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ at a concentration of about 20 mg/L. In embodiments, the growth medium includes CuSO_4 at a concentration of about 15 mg/L. In embodiments, the growth medium includes CoCl_2 at a concentration of 15 mg/L. In embodiments, the growth medium includes MgSO_4 at a concentration of 250 mg/L. In embodiments, the growth medium includes KI at a concentration of 15 mg/L. In embodiments, the growth medium includes $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ at a concentration of 20 mg/L. In embodiments, the growth medium includes $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ at a concentration of 12.5 mg/L. In embodiments, the growth medium includes Boric acid at a concentration of 12.5 mg/L. In embodiments, the growth medium includes $(\text{NH}_4)_2\text{Mo} \cdot 4\text{H}_2\text{O}$ at a concentration of 15 mg/L. In embodiments, the growth medium includes $\text{NiSO}_4 \cdot 6\text{H}_2\text{O}$ at a concentration of 12.5 mg/L. In embodiments, the growth medium includes $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ at a concentration of 20 mg/L. In embodiments, the growth medium includes CuSO_4 at a concentration of 15 mg/L. In embodiments, the growth medium is supplemented with CoCl_2 at a concentration of about 15 mg/L. In embodiments, the growth medium is supplemented with MgSO_4 at a concentration of about 250 mg/L. In embodiments, the growth medium is supplemented

with KI at a concentration of about 15 mg/L. In embodiments, the growth medium is supplemented with $ZnSO_4 \cdot 7H_2O$ at a concentration of about 20 mg/L. In embodiments, the growth medium is supplemented with $MnSO_4 \cdot H_2O$ at a concentration of about 12.5 mg/L. In embodiments, the growth medium is supplemented with Boric acid at a concentration of about 12.5 mg/L. In embodiments, the growth medium is supplemented with $(NH_4)_2Mo \cdot 4H_2O$ at a concentration of about 15 mg/L. In embodiments, the growth medium is supplemented with $NiSO_4 \cdot 6H_2O$ at a concentration of about 12.5 mg/L. In embodiments, the growth medium is supplemented with $FeSO_4 \cdot 7H_2O$ at a concentration of about 20 mg/L. In embodiments, the growth medium is supplemented with $CuSO_4$ at a concentration of about 15 mg/L. In embodiments, the growth medium is supplemented with $CoCl_2$ at a concentration of 15 mg/L. In embodiments, the growth medium is supplemented with $MgSO_4$ at a concentration of 250 mg/L. In embodiments, the growth medium is supplemented with KI at a concentration of 15 mg/L. In embodiments, the growth medium is supplemented with $ZnSO_4 \cdot 7H_2O$ at a concentration of 20 mg/L. In embodiments, the growth medium is supplemented with $MnSO_4 \cdot H_2O$ at a concentration of 12.5 mg/L. In embodiments, the growth medium is supplemented with Boric acid at a concentration of 12.5 mg/L. In embodiments, the growth medium is supplemented with $(NH_4)_2Mo \cdot 4H_2O$ at a concentration of 15 mg/L. In embodiments, the growth medium is supplemented with $NiSO_4 \cdot 6H_2O$ at a concentration of 12.5 mg/L. In embodiments, the growth medium is supplemented with $FeSO_4 \cdot 7H_2O$ at a concentration of 20 mg/L. In embodiments, the growth medium is supplemented with $CuSO_4$ at a concentration of 15 mg/L. In embodiments, the growth medium includes $CoCl_2$ at a concentration of about 7.5 to 22.5 mg/L. In embodiments, the growth medium includes $MgSO_4$ at a concentration of about 125 to 375 mg/L. In embodiments, the growth medium includes KI at a concentration of about 7.5 to 22.5 mg/L. In embodiments, the growth medium includes $ZnSO_4 \cdot 7H_2O$ at a concentration of about 10 to 30 mg/L. In embodiments, the growth medium includes $MnSO_4 \cdot H_2O$ at a concentration of about 6 to 18 mg/L. In embodiments, the growth medium includes Boric acid at a concentration of about 6 to 18 mg/L. In embodiments, the growth medium includes $(NH_4)_2Mo \cdot 4H_2O$ at a concentration of about 7.5 to 22.5 mg/L. In embodiments, the growth medium includes $NiSO_4 \cdot 6H_2O$ at a concentration of about 6 to 18 mg/L. In embodiments, the growth medium includes $FeSO_4 \cdot 7H_2O$ at a concentration of about 10 to 30 mg/L. In embodiments, the growth medium includes $CuSO_4$ at a concentration of about 7.5 to 22.5 mg/L. In embodiments, the growth medium includes $CoCl_2$ at a concentration of 7.5 to 22.5 mg/L. In embodiments, the growth medium includes $MgSO_4$ at a concentration of 125 to 375 mg/L. In embodiments, the growth medium includes KI at a concentration of 7.5 to 22.5 mg/L. In embodiments, the growth medium includes $ZnSO_4 \cdot 7H_2O$ at a concentration of 10 to 30 mg/L. In embodiments, the growth medium includes $MnSO_4 \cdot H_2O$ at a concentration of 6 to 18 mg/L. In embodiments, the growth medium includes Boric acid at a concentration of 6 to 18 mg/L. In embodiments, the growth medium includes $(NH_4)_2Mo \cdot 4H_2O$ at a concentration of 7.5 to 22.5 mg/L. In embodiments, the growth medium includes $NiSO_4 \cdot 6H_2O$ at a concentration of 6 to 18 mg/L. In embodiments, the growth medium includes $FeSO_4 \cdot 7H_2O$ at a concentration of 10 to 30 mg/L. In embodiments, the growth medium includes $CuSO_4$ at a concentration of 7.5 to 22.5 mg/L. In embodiments, the growth medium is supplemented

with CoCl_2 at a concentration of about 7.5 to 22.5 mg/L. In embodiments, the growth medium is supplemented with MgSO_4 at a concentration of about 125 to 375 mg/L. In embodiments, the growth medium is supplemented with KI at a concentration of about 7.5 to 22.5 mg/L. In embodiments, the growth medium is supplemented with $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ at a concentration of about 10 to 30 mg/L. In embodiments, the growth medium is supplemented with $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ at a concentration of about 6 to 18 mg/L. In embodiments, the growth medium is supplemented with Boric acid at a concentration of about 6 to 18 mg/L. In embodiments, the growth medium is supplemented with $(\text{NH}_4)_2\text{Mo.4H}_2\text{O}$ at a concentration of about 7.5 to 22.5 mg/L. In embodiments, the growth medium is supplemented with $\text{NiSO}_4 \cdot 6\text{H}_2\text{O}$ at a concentration of about 6 to 18 mg/L. In embodiments, the growth medium is supplemented with $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ at a concentration of about 10 to 30 mg/L. In embodiments, the growth medium is supplemented with CuSO_4 at a concentration of about 7.5 to 22.5 mg/L. In embodiments, the growth medium is supplemented with CoCl_2 at a concentration of 7.5 to 22.5 mg/L. In embodiments, the growth medium is supplemented with MgSO_4 at a concentration of 125 to 375 mg/L. In embodiments, the growth medium is supplemented with KI at a concentration of 7.5 to 22.5 mg/L. In embodiments, the growth medium is supplemented with $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ at a concentration of 10 to 30 mg/L. In embodiments, the growth medium is supplemented with $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ at a concentration of 6 to 18 mg/L. In embodiments, the growth medium is supplemented with Boric acid at a concentration of 6 to 18 mg/L. In embodiments, the growth medium is supplemented with $(\text{NH}_4)_2\text{Mo.4H}_2\text{O}$ at a concentration of 7.5 to 22.5 mg/L. In embodiments, the growth medium is supplemented with $\text{NiSO}_4 \cdot 6\text{H}_2\text{O}$ at a concentration of 6 to 18 mg/L. In embodiments, the growth medium is supplemented with $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ at a concentration of 10 to 30 mg/L. In embodiments, the growth medium is supplemented with CuSO_4 at a concentration of 7.5 to 22.5 mg/L.

In embodiments, the method does not include nitrogen starvation of the oleaginous organism (e.g. oleaginous yeast cell).

In embodiments, the oleaginous yeast is cultured for about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 250, 300, 350, 400, or 500 hours. In embodiments, the oleaginous yeast is cultured for 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 250, 300, 350, 400, or 500 hours. In embodiments, the oleaginous yeast is cultured for about 48, 96, 144, or 192 hours. In embodiments, the oleaginous yeast is cultured for 48, 96, 144, or 192 hours. In embodiments, the oleaginous yeast is cultured for about 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5, 5, 5.5, 6, 6.5, 7, 7.5, 8, 8.5, 9, 9.5, or 10 days. In embodiments, the oleaginous yeast is cultured for 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5, 5, 5.5, 6, 6.5, 7, 7.5, 8, 8.5, 9, 9.5, or 10 days.

In an aspect is provided a method of producing a lipid, lipid precursor, or oleochemical including culturing a yeast cell described herein in a growth medium; and isolating the lipid, lipid precursor, or oleochemical.

In embodiments, the lipid, lipid precursor, or oleochemical is isolated from the yeast cell. In embodiments, the lipid, lipid precursor, or oleochemical is isolated from the growth medium. In embodiments, the growth medium includes a majority carbon source selected from the group consisting of glucose, glycerol, xylose, fructose, mannose, ribose, sucrose, and lignocellulosic biomass. In embodiments, the growth medium includes lignocellulosic biomass as the majority carbon source. In embodiments, the growth medium includes a carbon source and a nitrogen source wherein the carbon source is at a concentration at least 10-fold greater than the concentration of the nitrogen source (wt/wt). In embodiments, the growth medium includes a carbon source and a nitrogen source wherein the carbon source is at a concentration at least 16-fold greater than the concentration of the nitrogen source (wt/wt). In embodiments, the growth medium includes a carbon source and a nitrogen source wherein the carbon source is at a concentration at least 320-fold greater than the concentration of the nitrogen source (wt/wt).

In embodiments, the growth medium includes cobalt, iron, magnesium, potassium, zinc, nickel, molybdenum, manganese, copper, or boron. In embodiments, the growth medium includes any combination of two or more of cobalt, iron, magnesium, potassium, zinc, nickel, molybdenum, manganese, copper, or boron. In embodiments, the growth medium includes cobalt in an amount equivalent to 7.5 to 22.5 mg/L CoCl₂, magnesium in an amount equivalent to 125 to 375 mg/L MgSO₄, potassium in an amount equivalent to 7.5 to 22.5 mg/L KI, zinc in an amount equivalent to 10 to 30 mg/L ZnSO₄·7H₂O, manganese in an amount equivalent to 6 to 18 mg/L MnSO₄·H₂O, boron in an amount equivalent to 6 to 18 mg/L Boric acid, molybdenum in an amount equivalent to 7.5 to 22.5 mg/L (NH₄)₂Mo·4H₂O, nickel in an amount equivalent to 6 to 18 mg/L NiSO₄·6H₂O, iron in an amount equivalent to 10 to 30 mg/L FeSO₄·7H₂O, or copper in an amount equivalent to 7.5 to 22.5 mg/L CuSO₄. In embodiments, the growth medium includes about 5.77×10⁻⁵ M to 1.73×10⁻⁴ M cobalt, about 0.001 M to 0.003 M magnesium, about 4.52×10⁻⁵ M to 1.35×10⁻⁴ M potassium, about 4.05×10⁻⁵ M to 1.22×10⁻⁴ M zinc, about 3.55×10⁻⁵ to 1.06×10⁻⁴ M manganese, about 9.07×10⁻⁵ M to 2.91×10⁻⁴ M boron, about 3.76×10⁻⁵ M to 1.10×10⁻⁴ M molybdenum, about 2.28×10⁻⁵ M to 6.84×10⁻⁵ M nickel, about 3.60×10⁻⁵ M to 1.08×10⁻⁴ M iron, or about 4.70×10⁻⁵ M to 1.41×10⁻⁴ M copper. In embodiments, the growth medium includes 5.77×10⁻⁵ M to 1.73×10⁻⁴ M cobalt, 0.001 M to 0.003 M magnesium, 4.52×10⁻⁵ M to 1.35×10⁻⁴ M potassium, 4.05×10⁻⁵ M to 1.22×10⁻⁴ M zinc, 3.55×10⁻⁵ to 1.06×10⁻⁴ M manganese, 9.07×10⁻⁵ M to 2.91×10⁻⁴ M boron, 3.76×10⁻⁵ M to 1.10×10⁻⁴ M molybdenum, 2.28×10⁻⁵ M to 6.84×10⁻⁵ M nickel, 3.60×10⁻⁵ M to 1.08×10⁻⁴ M iron, or 4.70×10⁻⁵ M to 1.41×10⁻⁴ M copper. In embodiments, the growth medium includes about 5.77×10⁻⁵ M to 1.73×10⁻⁴ M cobalt. In embodiments, the growth medium includes about 0.001 M to 0.003 M magnesium. In embodiments, the growth medium includes about 4.52×10⁻⁵ M to 1.35×10⁻⁴ M potassium. In embodiments, the growth medium includes about 4.05×10⁻⁵ M to 1.22×10⁻⁴ M zinc. In embodiments, the growth medium includes about 3.55×10⁻⁵ to 1.06×10⁻⁴ M manganese. In embodiments, the growth medium includes about 9.07×10⁻⁵ M to 2.91×10⁻⁴ M boron. In embodiments, the growth medium includes about

3.76×10⁻⁵ M to 1.10×10⁻⁴ M molybdenum. In embodiments, the growth medium includes about 2.28×10⁻⁵ M to 6.84×10⁻⁵ M nickel. In embodiments, the growth medium includes about 3.60×10⁻⁵ M to 1.08×10⁻⁴ M iron. In embodiments, the growth medium includes about 4.70×10⁻⁵ M to 1.41×10⁻⁴ M copper. In embodiments, the growth medium includes 5.77×10⁻⁵ M to 1.73×10⁻⁴ M cobalt. In embodiments, the growth medium includes 0.001 M to 0.003 M magnesium. In embodiments, the growth medium includes about 4.52×10⁻⁵ M to 1.35×10⁻⁴ M potassium. In embodiments, the growth medium includes 4.05×10⁻⁵ M to 1.22×10⁻⁴ M zinc. In embodiments, the growth medium includes 3.55×10⁻⁵ to 1.06×10⁻⁴ M manganese. In embodiments, the growth medium includes 9.07×10⁻⁵ M to 2.91×10⁻⁴ M boron. In embodiments, the growth medium includes 3.76×10⁻⁵ M to 1.10×10⁻⁴ M molybdenum. In embodiments, the growth medium includes 2.28×10⁻⁵ M to 6.84×10⁻⁵ M nickel. In embodiments, the growth medium includes 3.60×10⁻⁵ M to 1.08×10⁻⁴ M iron. In embodiments, the growth medium includes 4.70×10⁻⁵ M to 1.41×10⁻⁴ M copper. In embodiments, the growth medium includes iron, copper, and molybdenum. In embodiments, the growth medium includes molybdenum in an amount equivalent to 7.5 to 22.5 mg/L (NH₄)₂Mo·4H₂O, iron in an amount equivalent to 10 to 30 mg/L FeSO₄·7H₂O, or copper in an amount equivalent to 7.5 to 22.5 mg/L CuSO₄. In embodiments, the growth medium includes 3.76×10⁻⁵ M to 1.10×10⁻⁴ M molybdenum, 3.60×10⁻⁵ M to 1.08×10⁻⁴ M iron, or 4.70×10⁻⁵ M to 1.41×10⁻⁴ M copper. In embodiments, the growth medium includes copper and nickel. In embodiments, the growth medium includes nickel in an amount equivalent to 6 to 18 mg/L NiSO₄·6H₂O or copper in an amount equivalent to 7.5 to 22.5 mg/L CuSO₄. In embodiments, the growth medium includes 2.28×10⁻⁵ M to 6.84×10⁻⁵ M nickel or 4.70×10⁻⁵ M to 1.41×10⁻⁴ M copper. In embodiments, the growth medium includes copper, iron, and either molybdenum or nickel. In embodiments, the growth medium includes molybdenum in an amount equivalent to 7.5 to 22.5 mg/L (NH₄)₂Mo·4H₂O, nickel in an amount equivalent to 6 to 18 mg/L NiSO₄·6H₂O, iron in an amount equivalent to 10 to 30 mg/L FeSO₄·7H₂O, or copper in an amount equivalent to 7.5 to 22.5 mg/L CuSO₄. In embodiments, the growth medium includes 3.76×10⁻⁵ M to 1.10×10⁻⁴ M molybdenum, 2.28×10⁻⁵ M to 6.84×10⁻⁵ M nickel, 3.60×10⁻⁵ M to 1.08×10⁻⁴ M iron, or 4.70×10⁻⁵ M to 1.41×10⁻⁴ M copper. In embodiments, the growth medium includes copper, iron, molybdenum, and nickel.

In another aspect is provided a method of isolating a yeast cell including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight from a plurality of yeast cells, including allowing a yeast cell including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) to separate from a population of yeast cells within the plurality of yeast cells by floating above the population of yeast cells within an aqueous medium thereby isolating the yeast cell including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical), wherein the population of yeast cells includes a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) than the yeast cell including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors,

lipid precursor, oleochemicals, or oleochemical). In another aspect is provided a method of isolating a genetically modified yeast cell from a plurality of yeast cells including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight, including allowing a genetically modified yeast cell to separate from a population of yeast cells within the plurality of yeast cells by floating above the population of yeast cells within an aqueous medium thereby isolating the genetically modified yeast cell, wherein the population of yeast cells includes a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) than the genetically modified yeast cell.

In embodiments is a method of isolating a yeast cell (e.g. genetically modified yeast cell), including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight, from a plurality of yeast cells, including allowing a yeast cell (e.g. genetically modified yeast cell) to separate from a population of yeast cells within the plurality of yeast cells by floating above the population of yeast cells within an aqueous medium thereby isolating the yeast cell (e.g. genetically modified yeast cell), wherein the population of yeast cells includes a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) than the genetically modified yeast cell.

In embodiments, the yeast cell (e.g., genetically modified yeast cell) includes greater than 30% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight. In embodiments, the yeast cell (e.g., genetically modified yeast cell) includes greater than 40% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight. In embodiments, the yeast cell (e.g., genetically modified yeast cell) includes greater than 50% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight. In embodiments, the yeast cell (e.g., genetically modified yeast cell) includes greater than 60% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight. In embodiments, the yeast cell (e.g., genetically modified yeast cell) includes greater than 70% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight. In embodiments, the yeast cell (e.g., genetically modified yeast cell) includes greater than 80% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight. In embodiments, the yeast cell (e.g., genetically modified yeast cell) includes greater than 90% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight. In embodiments, the yeast cell (e.g., genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight (e.g., greater than 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66,

67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82,
83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98,
or 99% lipids, lipid precursors, and/or oleochemicals (e.g.,
lipid, lipids, lipid precursors, lipid precursor, oleochemicals,
5 or oleochemical) in dry weight) is floating on the top surface
of the aqueous medium. In embodiments, the yeast cell (e.g.,
genetically modified yeast cell) including greater than 20%
wt/wt lipids, lipid precursors, and/or oleochemicals (e.g.,
lipid, lipids, lipid precursors, lipid precursor, oleochemicals,
10 or oleochemical) in dry weight (e.g. greater than 20, 21, 22,
23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38,
39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54,
15 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70,
71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86,
20 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% lipids,
lipid precursors, and/or oleochemicals (e.g., lipid, lipids,
lipid precursors, lipid precursor, oleochemicals, or oleo-
chemical) in dry weight) is above the bottom of a vessel
containing the aqueous medium. In embodiments, the yeast
25 cell (e.g. genetically modified yeast cell) including greater
than 20% wt/wt lipids, lipid precursors, and/or oleochemicals
(e.g., lipid, lipids, lipid precursors, lipid precursor,
oleochemicals, or oleochemical) in dry weight (e.g. greater
than 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34,
30 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50,
51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66,
67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82,
83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98,
35 or 99% lipids, lipid precursors, and/or oleochemicals (e.g.,
lipid, lipids, lipid precursors, lipid precursor, oleochemicals,
or oleochemical) in dry weight) is floating above the popu-
lation of yeast cells including a lower percentage wt/wt of
lipids, lipid precursors, and/or oleochemicals (e.g., lipid,
lipids, lipid precursors, lipid precursor, oleochemicals, or
30 oleochemical) than the yeast cell (e.g. genetically modified
yeast cell) including greater than 20% wt/wt lipids, lipid
precursors, and/or oleochemicals (e.g., lipid, lipids, lipid
precursors, lipid precursor, oleochemicals, or oleochemical)
in dry weight by about 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8,
35 0.9, 1.0, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17,
18, 19, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85,
90, 95, 96, 97, 98, or 99 mm in the aqueous medium. In
40 embodiments, the yeast cell (e.g. genetically modified yeast
cell) including greater than 20% wt/wt lipids, lipid pre-
cursors, and/or oleochemicals (e.g., lipid, lipids, lipid pre-
cursors, lipid precursor, oleochemicals, or oleochemical) in dry
45 weight (e.g. greater than 20, 21, 22, 23, 24, 25, 26, 27, 28,
29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44,
45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60,
50 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76,
77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92,
93, 94, 95, 96, 97, 98, or 99% lipids, lipid precursors, and/or
oleochemicals (e.g., lipid, lipids, lipid precursors, lipid pre-
55 cursor, oleochemicals, or oleochemical) in dry weight) is
floating above the population of yeast cells including a lower
percentage wt/wt of lipids, lipid precursors, and/or oleo-
chemicals (e.g., lipid, lipids, lipid precursors, lipid pre-
cursor, oleochemicals, or oleochemical) than the yeast cell (e.g.
genetically modified yeast cell) including greater than 20%
60 wt/wt lipids, lipid precursors, and/or oleochemicals (e.g.,
lipid, lipids, lipid precursors, lipid precursor, oleochemicals,
or oleochemical) in dry weight by at least 0.1, 0.2, 0.3, 0.4,
0.5, 0.6, 0.7, 0.8, 0.9, 1.0, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12,
65 13, 14, 15, 16, 17, 18, 19, 20, 25, 30, 35, 40, 45, 50, 55, 60,
65, 70, 75, 80, 85, 90, 95, 96, 97, 98, or 99 mm in the
aqueous medium. In embodiments, the genetically modified
yeast cell including greater than 20% wt/wt lipids, lipid

precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight (e.g. greater than 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight floating above the population of yeast cells including a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) than the yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight has a buoyant density greater than the buoyant density of the population of yeast cells including a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) than yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight by about 0.001, 0.002, 0.003, 0.004, 0.005, 0.006, 0.007, 0.008, 0.009, 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, g/mL. In embodiments, the yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight (e.g. greater than 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight floating above the population of yeast cells including a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) than the yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight has a buoyant density greater than the buoyant density of the population of yeast cells including a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) than yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight by 0.001, 0.002, 0.003, 0.004, 0.005, 0.006, 0.007, 0.008, 0.009, 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, g/mL. In embodiments, the yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical), includes a mutation created by natural genetic drift.

In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of about

0.5 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 1.0 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 2.0 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 2.5 vvm (volume per volume per minute). In 10 embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 3.0 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 4.0 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of 0.5 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of 1.0 15 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of 2.0 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of 2.5 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of 3.0 vvm (volume per volume per minute). In embodiments of the method, the plurality of yeast cells are in a bioreactor with agitation and aeration rates of 4.0 vvm (volume per volume per minute).

In embodiments of the method, the aqueous medium includes a yeast growth medium, minimal media, complete supplement media, or greater than 50 g/L carbon source (e.g. glucose) and less than 5 g/L of a nitrogen source (e.g. ammonium sulfate). In embodiments of the method, the aqueous medium includes a yeast growth medium. In embodiments of the method, the aqueous medium includes a minimal media. In embodiments of the method, the aqueous medium includes a complete supplement media. In 35 embodiments of the method, the aqueous medium includes greater than 50 g/L carbon source (e.g. glucose) and less than 5 g/L of a nitrogen source (e.g. ammonium sulfate). In embodiments of the method, the aqueous medium is a yeast growth medium. In embodiments of the method, the aqueous medium is a minimal media. In embodiments of the method, the aqueous medium is a complete supplement media.

In embodiments of the method of isolating a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight, including allowing a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight to separate from a population of yeast cells within the plurality of yeast cells by floating above the population of yeast cells within an aqueous medium, the allowing is performed by centrifugation or simple sedimentation. In embodiments of the method of isolating a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight, including allowing a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight,

lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight to separate from a population of yeast cells within the plurality of yeast cells by floating above the population of yeast cells within an aqueous medium, the allowing is performed by centrifugation. In embodiments of the method of isolating a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight, including allowing a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight to separate from a population of yeast cells within the plurality of yeast cells by floating above the population of yeast cells within an aqueous medium, the allowing is performed by simple sedimentation. In embodiments of the method of isolating a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight, including allowing a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight to separate from a population of yeast cells within the plurality of yeast cells by floating above the population of yeast cells within an aqueous medium, the allowing is performed by sedimentation. In embodiments of the method of isolating a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight, including allowing a yeast cell (e.g. genetically modified yeast cell) including greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight to separate from a population of yeast cells within the plurality of yeast cells by floating above the population of yeast cells within an aqueous medium, the allowing is performed by sedimentation due to gravity.

In embodiments of the method, the genetically modified yeast cell is formed by transforming a yeast cell with a recombinant nucleic acid (e.g. as described herein, including in embodiments, examples, tables, figures, and/or claims). In embodiments, the genetically modified yeast cell is formed by mutagenizing a yeast cell. In embodiments, the yeast cell (e.g. genetically modified yeast cell includes a mutation created by natural genetic drift.

In embodiments, the method is a method described herein, including in embodiments, examples, tables, figures, and claims.

IV. ADDITIONAL EMBODIMENTS

- 1p. A genetically modified yeast cell wherein the dry weight of said yeast cell comprises greater than 20% wt/wt lipid.
- 2p. The yeast cell of embodiment 1p comprising greater than 30% wt/wt lipid.
- 3p. The yeast cell of embodiment 1p comprising greater than 40% wt/wt lipid.
- 4p. The yeast cell of embodiment 1p comprising greater than 50% wt/wt lipid.
- 5p. The yeast cell of embodiment 1p comprising greater than 60% wt/wt lipid.

- 6p. The yeast cell of embodiment 1p comprising greater than 70% wt/wt lipid.
- 7p. The yeast cell of embodiment 1p comprising greater than 80% wt/wt lipid.
- 8p. The yeast cell of embodiment 1p comprising greater than 90% wt/wt lipid.
- 9p. The yeast cell of any one of embodiments 1p to 8p, selected from the group consisting of the genera *Yarrowia*, *Candida*, *Rhodotorula*, *Rhodosporidium*, *Cryptococcus*, *Trichosporon* and *Lipomyces*.
- 10p. The yeast cell of any one of embodiments 1p to 8p, selected from the group consisting of *Rhodosporidium toruloides*, *Lipomyces starkeyii*, *Lipomyces lipoferus*, *Apotrichum curvatum*, *Candida curvata*, *Cryptococcus curvatus*, *Trichosporon fermentans*, *Candida revkaufi*, *Candida pulcherrima*, *Candida tropicalis*, *Candida utilis*, *Trichosporon pullans*, *Trichosporon cutaneum*, *Rhodotorula glutinis*, *Rhodotorula graminis* and *Yarrowia lipolytica*.
- 11p. The yeast cell of any one of embodiments 1p to 8p, selected from the group consisting of *Lipomyces starkeyii*, *Rhodosporidium toruloides*, *Apotrichum curvatum*, *Candida curvata*, *Cryptococcus curvatus*, *Trichosporon fermentans*, *Rhodotorula glutinis*, and *Yarrowia lipolytica*.
- 12p. The yeast cell of any one of embodiments 1p to 8p, wherein said yeast cell is *Yarrowia lipolytica*.
- 13p. The yeast cell of any one of embodiments 1p to 12p, wherein said yeast cell is buoyant in an aqueous medium.
- 14p. The yeast cell of any one of embodiments 1p to 13p, wherein said lipid is selected from the group consisting of a fatty acid, wax, sterol, vitamin, monoglyceride, diglyceride, triglyceride, phospholipid, glycerolipid, glycerophospholipid, sphingolipid, saccharolipid, polyketide, sterol lipid, triacylglyceride, and a prenol lipid.
- 15p. A yeast cell comprising a recombinant nucleic acid, wherein said recombinant nucleic acid modulates the level of activity of a protein in said yeast cell relative to the absence of the recombinant nucleic acid, and wherein said protein is selected from the group consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), multifunctional enzyme (MFE1), Transcription Factor (PEX10), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA:diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), and O-6-methylguanine-DNA methyltransferase (MGMT).
- 16p. The yeast cell of embodiment 15p, wherein said recombinant nucleic acid increases the level of activity of a protein in said yeast cell selected from the group consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), AMP Deaminase (AMPD), ATP-Citrate Lyase (ACL), Malic Enzyme (MAE), Acetyl-CoA Carboxylase (ACC), acyl-CoA: diacylglycerol acyltransferase (DGA1), acyl-CoA: diacylglycerol acyltransferases (DGA2), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), and O-6-methylguanine-DNA methyltransferase (MGMT).
- 17p. The yeast cell of any one of embodiments 15p to 16p, wherein said recombinant nucleic acid decreases the level of activity of a protein in said yeast cell selected from the group consisting of multifunctional enzyme (MFE1), Lipid synthesis regulator (MGA2), Chromatin assembly gene (RLF2 subunit p90), and Transcription Factor (PEX10).

18p. The yeast cell of any one of embodiments 15p to 17p, wherein said recombinant nucleic acid increases the level of activity of a protein in said yeast cell selected from the group consisting of Leucine Biosynthesis Gene (LEU2), Uracil Biosynthesis gene (URA3), Malic Enzyme (MAE), Mitochondrial 2' O-ribose methyltransferase (MRM2), Lipid synthesis regulator (MGA2), and O-6-methylguanine-DNA methyltransferase (MGMT) or said nucleic acid decrease the level of activity of Lipid synthesis regulator (MGA2).

19p. The yeast cell of any one of embodiments 15p to 18p, wherein said recombinant nucleic acid encodes a protein comprising a mutation relative to the wildtype protein.

20p. The yeast cell of any one of embodiments 15p to 18p, wherein said nucleic acid modulates the level of expression of a protein.

21p. The yeast cell of embodiment 15p, wherein said yeast cell comprises a recombinant nucleic acid that decreases the level of activity of multifunctional enzyme (MFE1) protein and Transcription Factor (PEX10) protein, increases the level of activity of acyl-CoA:diacylglycerol acyltransferase (DGA1) protein, or increases the level of activity of Leucine Biosynthesis Gene (LEU2) protein relative to a yeast cell that does not comprise said recombinant nucleic acids.

22p. The yeast cell of any one of embodiments 1p to 21p, wherein said yeast cell comprises a fatty acid selected from the group consisting of C5:0, C5:1, C5:2, C5:3, C6:0, C6:1, C6:2, C6:3, C7:0, C7:1, C7:2, C7:3, C8:0, C8:1, C8:2, C8:3, C9:0, C9:1, C9:2, C9:3, C10:0, C10:1, C10:2, C10:3, C11:0, C11:1, C11:2, C11:3, C12:0, C12:1, C12:2, C12:3, C13:0, C13:1, C13:2, C13:3, C14:0, C14:1, C14:2, C14:3, C15:0, C15:1, C15:2, C15:3, C16:0, C16:1, C16:2, C16:3, C17:0, C17:1, C17:2, C17:3, C18:0, C18:1, C18:2, C18:3, C19:0, C19:1, C19:2, C19:3, C20:0, C20:1, C20:2, C20:3, C21:0, C21:1, C21:2, C21:3, C22:0, C22:1, C22:2, C22:3, C23:0, C23:1, C23:2, and C23:3.

23p. The yeast cell of any one of embodiments 1p to 21p, wherein said yeast cell comprises a fatty acid selected from the group consisting of C17:0 and C17:1.

24p. A method of producing a lipid comprising:

- 1) culturing a yeast cell of any one of embodiments 1p to 23p in a growth medium;
- 2) isolating said lipid.

25p. The method of embodiment 24p, wherein said lipid is isolated from said yeast cell.

26p. The method of embodiment 24p, wherein said lipid is isolated from the medium.

27p. The method of any one of embodiments 24p to 26p, wherein said growth medium comprises a majority carbon source selected from the group consisting of glucose, glycerol, xylose, fructose, mannose, ribose, sucrose, and lignocellulosic biomass.

28p. The method of any one of embodiments 24p to 26p, wherein said growth medium comprises lignocellulosic biomass as the majority carbon source.

29p. The method of any one of embodiments 24p to 28p, wherein said growth medium comprises a carbon source and a nitrogen source wherein said carbon source is at a concentration at least 10-fold greater than the concentration of the nitrogen source.

30p. The method of any one of embodiments 24p to 29p, wherein said growth medium comprises cobalt, iron, magnesium, potassium, zinc, nickel, molybdenum, manganese, copper, or boron.

31p. The method of embodiment 30p, wherein the growth medium comprises iron, copper, and molybdenum.

32p. The method of embodiment 30p, wherein the growth medium comprises copper and nickel.

33p. The method of embodiment 30p, wherein the growth medium comprises copper, iron, and either molybdenum or nickel.

34p. The method of embodiment 30p, wherein the growth medium comprises copper, iron, molybdenum, and nickel.

35p. A method of isolating a genetically modified yeast cell from a plurality of yeast cells comprising greater than 20% wt/wt lipids in dry weight, comprising allowing a genetically modified yeast cell to separate from a population of yeast cells within said plurality of yeast cells by floating above said population of yeast cells within an aqueous medium thereby isolating said genetically modified yeast cell, wherein said population of yeast cells comprises a lower percentage wt/wt of lipids than said genetically modified yeast cell.

36p. The method of any embodiment 35p, wherein said genetically modified yeast cell comprises greater than 30% wt/wt lipids in dry weight.

37p. The method of embodiment 35p, wherein said genetically modified yeast cell comprises greater than 40% wt/wt lipids in dry weight.

38p. The method of embodiment 35p, wherein said genetically modified yeast cell comprises greater than 50% wt/wt lipids in dry weight.

39p. The method of embodiment 35p, wherein said genetically modified yeast cell comprises greater than 60% wt/wt lipids in dry weight.

40p. The method of any one of embodiments 35p to 39p, wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 0.5 vvm (volume per volume per minute).

41p. The method of any one of embodiments 35p to 39p, wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 1.0 vvm (volume per volume per minute).

42p. The method of any one of embodiments 35p to 39p, wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 2.0 vvm (volume per volume per minute).

43p. The method of any one of embodiments 35p to 39p, wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 3.0 vvm (volume per volume per minute).

44p. The method of any one of embodiments 35p to 39p, wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 4.0 vvm (volume per volume per minute).

45p. The method of any one of embodiments 35p to 44p, wherein said aqueous medium comprises a yeast growth medium, minimal media, complete supplement media, or greater than 50 g/L glucose and less than 5 g/L of a nitrogen source.

46p. The method of any one of embodiments 35p to 45p, wherein said allowing is performed by centrifugation or simple sedimentation.

47p. The method of any one of embodiments 35p to 46p, wherein said genetically modified yeast cell was formed by transforming a yeast cell with a recombinant nucleic acid.

48p. The method of any one of embodiments 35p to 47p, wherein said genetically modified yeast cell was formed by mutagenizing a yeast cell.

1. A genetically modified yeast cell wherein the dry weight of said yeast cell comprises greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).

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2. The genetically modified yeast cell of embodiment 1 comprising greater than 30% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).
3. The genetically modified yeast cell of embodiment 1 comprising greater than 40% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).
4. The genetically modified yeast cell of embodiment 1 comprising greater than 50% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).
5. The genetically modified yeast cell of embodiment 1 comprising greater than 60% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).
6. The genetically modified yeast cell of embodiment 1 comprising greater than 70% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).
7. The genetically modified yeast cell of embodiment 1 comprising greater than 80% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).
8. The genetically modified yeast cell of embodiment 1 comprising greater than 90% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).
9. The genetically modified yeast cell of any one of embodiments 1 to 8, selected from the group consisting of the genera *Yarrowia*, *Candida*, *Rhodotorula*, *Rhodosporidium*, *Cryptococcus*, *Trichosporon* and *Lipomyces*.
10. The genetically modified yeast cell of any one of embodiments 1 to 8, selected from the group consisting of *Rhodosporidium toruloides*, *Lipomyces starkeyii*, *Lipomyces lipoferus*, *Apotrichum curvatum*, *Candida curvata*, *Cryptococcus curvatus*, *Trichosporon fermentans*, *Candida revkaufti*, *Candida pulcherrima*, *Candida tropicalis*, *Candida utilis*, *Trichosporon pullans*, *Trichosporon cutaneum*, *Rhodotorula glutinis*, *Rhodotorula graminis* and *Yarrowia lipolytica*.
11. The genetically modified yeast cell of any one of embodiments 1 to 8, selected from the group consisting of *Lipomyces starkeyii*, *Rhodosporidium toruloides*, *Apotrichum curvatum*, *Candida curvata*, *Cryptococcus curvatus*, *Trichosporon fermentans*, *Rhodotorula glutinis*, and *Yarrowia lipolytica*.
12. The genetically modified yeast cell of any one of embodiments 1 to 8, wherein said yeast cell is *Yarrowia lipolytica*.
13. The genetically modified yeast cell of any one of embodiments 1 to 12, wherein said yeast cell is buoyant in an aqueous medium.
14. The genetically modified yeast cell of one of embodiments 1 to 13, comprising a recombinant Leucine Biosynthesis Gene (LEU2).
15. The genetically modified yeast cell of one of embodiments 1 to 13, wherein said genetic modification increases the level of activity of the Leucine Biosynthesis Gene (LEU2) protein relative to an otherwise identical yeast cell lacking said genetic modification.
16. The genetically modified yeast cell of one of embodiments 1 to 15, comprising a recombinant Uracil Biosynthesis gene (URA3).
17. The genetically modified yeast cell of one of embodiments 1 to 15, wherein said genetic modification increases

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- the level of activity of the Uracil Biosynthesis gene (URA3) protein relative to an otherwise identical yeast cell lacking said genetic modification.
18. The genetically modified yeast cell of one of embodiments 1 to 17, comprising a genetically modified multifunctional enzyme (MFE1) gene.
19. The genetically modified yeast cell of one of embodiments 1 to 17, wherein said genetic modification decreases the level of activity of the multifunctional enzyme (MFE1) protein relative to an otherwise identical yeast cell lacking said genetic modification.
20. The genetically modified yeast cell of one of embodiments 1 to 19, comprising a genetically modified PEX10 Transcription Factor (PEX10) gene.
21. The genetically modified yeast cell of one of embodiments 1 to 19, wherein said genetic modification decreases the level of activity of the PEX10 Transcription Factor (PEX10) protein relative to an otherwise identical yeast cell lacking said genetic modification.
22. The genetically modified yeast cell of one of embodiments 1 to 21, comprising a recombinant AMP Deaminase (AMPD) protein.
23. The genetically modified yeast cell of one of embodiments 1 to 21, wherein said genetic modification increases the level of activity of the AMP Deaminase (AMPD) protein relative to an otherwise identical yeast cell lacking said genetic modification.
24. The genetically modified yeast cell of one of embodiments 1 to 23, comprising a recombinant ATP-Citrate Lyase (ACL1) protein.
25. The genetically modified yeast cell of one of embodiments 1 to 23, wherein said genetic modification increases the level of activity of the ATP-Citrate Lyase (ACL1) protein relative to an otherwise identical yeast cell lacking said genetic modification.
26. The genetically modified yeast cell of one of embodiments 1 to 25, comprising a recombinant ATP-Citrate Lyase (ACL2) protein.
27. The genetically modified yeast cell of one of embodiments 1 to 25, wherein said genetic modification increases the level of activity of the ATP-Citrate Lyase (ACL2) protein relative to an otherwise identical yeast cell lacking said genetic modification.
28. The genetically modified yeast cell of one of embodiments 1 to 27, comprising a recombinant Malic Enzyme (MAE) protein.
29. The genetically modified yeast cell of one of embodiments 1 to 27, wherein said genetic modification increases the level of activity of the Malic Enzyme (MAE) protein relative to an otherwise identical yeast cell lacking said genetic modification.
30. The genetically modified yeast cell of one of embodiments 1 to 29, comprising a recombinant Acetyl-CoA Carboxylase (ACC) protein.
31. The genetically modified yeast cell of one of embodiments 1 to 29, wherein said genetic modification increases the level of activity of the Acetyl-CoA Carboxylase (ACC) protein relative to an otherwise identical yeast cell lacking said genetic modification.
32. The genetically modified yeast cell of one of embodiments 1 to 31, comprising a recombinant acyl-CoA: diacylglycerol acyltransferase 1 (DGA1) protein.
33. The genetically modified yeast cell of one of embodiments 1 to 31, wherein said genetic modification increases the level of activity of the acyl-CoA: diacylglycerol acyltransferase 1 (DGA1) protein relative to an otherwise identical yeast cell lacking said genetic modification.

34. The genetically modified yeast cell of one of embodiments 1 to 33, comprising a recombinant acyl-CoA: diacylglycerol acyltransferase 2 (DGA2) protein.
35. The genetically modified yeast cell of one of embodiments 1 to 33, wherein said genetic modification increases the level of activity of the acyl-CoA:diacylglycerol acyltransferase 2 (DGA2) protein relative to an otherwise identical yeast cell lacking said genetic modification.
36. The genetically modified yeast cell of one of embodiments 1 to 35, comprising a recombinant Mitochondrial 2' O-ribose methyltransferase (MRM2) protein.
37. The genetically modified yeast cell of one of embodiments 1 to 35, wherein said genetic modification increases the level of activity of the Mitochondrial 2' O-ribose methyltransferase (MRM2) protein relative to an otherwise identical yeast cell lacking said genetic modification.
38. The genetically modified yeast cell of one of embodiments 1 to 37, comprising a recombinant Lipid synthesis regulator (MGA2) protein.
39. The genetically modified yeast cell of one of embodiments 1 to 37, comprising a genetically modified Lipid synthesis regulator (MGA2) gene.
40. The genetically modified yeast cell of one of embodiments 1 to 37, comprising at least one nucleotide deletion in the genomic Lipid synthesis regulator (MGA2) gene and expression of a Lipid synthesis regulator (MGA2) protein comprising a mutation corresponding to G643R in *Yarrowia lipolytica*. Lipid synthesis regulator (MGA2)
41. The genetically modified yeast cell of one of embodiments 1 to 37, wherein said genetic modification decreases the level of activity of the Lipid synthesis regulator (MGA2) protein relative to an otherwise identical yeast cell lacking said genetic modification.
42. The genetically modified yeast cell of one of embodiments 1 to 41, comprising a genetically modified Chromatin assembly gene (RLF2 subunit p90) gene.
43. The genetically modified yeast cell of one of embodiments 1 to 41, wherein said genetic modification decreases the level of activity of the Chromatin assembly gene (RLF2 subunit p90) protein relative to an otherwise identical yeast cell lacking said genetic modification.
44. The genetically modified yeast cell of one of embodiments 1 to 43, comprising a recombinant O-6-methylguanine-DNA methyltransferase (MGMT) protein.
45. The genetically modified yeast cell of one of embodiments 1 to 43, wherein said genetic modification increases the level of activity of the O-6-methylguanine-DNA methyltransferase (MGMT) protein relative to an otherwise identical yeast cell lacking said genetic modification.
46. The genetically modified yeast cell of one of embodiments 1 to 45, comprising a genetically modified Aconitase (ACO1) gene.
47. The genetically modified yeast cell of one of embodiments 1 to 45, wherein said genetic modification decreases the level of activity of the Aconitase (ACO1) protein relative to an otherwise identical yeast cell lacking said genetic modification.
48. The genetically modified yeast cell of one of embodiments 1 to 47, comprising a recombinant Citrate Synthase (CIT1) gene.
49. The genetically modified yeast cell of one of embodiments 1 to 47, wherein said genetic modification increases the level of activity of the Citrate Synthase (CIT1) protein relative to an otherwise identical yeast cell lacking said genetic modification.

50. The genetically modified yeast cell of one of embodiments 1 to 49, comprising a genetically modified RME1 zinc-finger transcription factor (RME1) gene.
51. The genetically modified yeast cell of one of embodiments 1 to 49, wherein said genetic modification decreases the level of activity of the RME1 zinc-finger transcription factor (RME1) protein relative to an otherwise identical yeast cell lacking said genetic modification.
52. The genetically modified yeast cell of one of embodiments 1 to 51, comprising a genetically modified YOX1 homeodomain protein (YOX1) gene.
53. The genetically modified yeast cell of one of embodiments 1 to 51, wherein said genetic modification decreases the level of activity of the YOX1 homeodomain protein (YOX1) protein relative to an otherwise identical yeast cell lacking said genetic modification.
54. The genetically modified yeast cell of one of embodiments 1 to 53, comprising a genetically modified UGA2 succinate semialdehyde dehydrogenase (UGA2) gene.
55. The genetically modified yeast cell of one of embodiments 1 to 53, wherein said genetic modification decreases the level of activity of the UGA2 succinate semialdehyde dehydrogenase (UGA2) protein relative to an otherwise identical yeast cell lacking said genetic modification.
56. The genetically modified yeast cell of one of embodiments 1 to 55, comprising a genetically modified OSH6 oxysterol-binding protein homolog 6 (OSH6) gene.
57. The genetically modified yeast cell of one of embodiments 1 to 55, wherein said genetic modification decreases the level of activity of the OSH6 oxysterol-binding protein homolog 6 (OSH6) protein relative to an otherwise identical yeast cell lacking said genetic modification.
58. The genetically modified yeast cell of one of embodiments 1 to 57, comprising a genetically modified IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) gene.
59. The genetically modified yeast cell of one of embodiments 1 to 57, wherein said genetic modification decreases the level of activity of the IRC20 E3 ubiquitin-protein ligase and helicase (IRC20) protein relative to an otherwise identical yeast cell lacking said genetic modification.
60. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification modulates the level of activity of a component of a lipid biosynthetic pathway.
61. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification modulates the level of activity of a component of a pathway incorporating Acetyl-CoA into a lipid, lipid precursor, or oleochemical.
62. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification modulates the level of activity of a component of a pathway incorporating malonyl-CoA into a lipid, lipid precursor, or oleochemical.
63. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification increases the level of activity of a component of a lipid biosynthetic pathway.
64. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification increases the level of activity of a component of a pathway incorporating acetyl-CoA into a lipid, lipid precursor, or oleochemical.
65. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification

increases the level of activity of a component of a pathway incorporating malonyl-CoA into a lipid, lipid precursor, or oleochemical.

66. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification modulates the level of activity of a component of a lipid, lipid precursor, or oleochemical, metabolic pathway.

67. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification decreases the level of activity of a component of a lipid, lipid precursor, or oleochemical, metabolic pathway.

68. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification decreases the level of activity of a component of a lipid, lipid precursor, or oleochemical, metabolic pathway.

69. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification increases the level of acetyl-CoA in the genetically modified yeast cell relative to a genetically unmodified yeast cell that is otherwise identical to said genetically modified yeast cell.

70. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification increases the level of malonyl-CoA in the genetically modified yeast cell relative to a genetically unmodified yeast cell that is otherwise identical to said genetically modified yeast cell.

71. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification increases the level of triglyceride production in the genetically modified yeast cell relative to a genetically unmodified yeast cell that is otherwise identical to said genetically modified yeast cell.

72. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification decreases the level of beta-oxidation activity in the genetically modified yeast cell relative to a genetically unmodified yeast cell that is otherwise identical to said genetically modified yeast cell.

73. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification decreases the level of fatty acid catabolism in the genetically modified yeast cell relative to a genetically unmodified yeast cell that is otherwise identical to said genetically modified yeast cell.

74. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification decreases the level of peroxisome biogenesis activity in the genetically modified yeast cell relative to a genetically unmodified yeast cell that is otherwise identical to said genetically modified yeast cell.

75. The genetically modified yeast cell of any one of embodiments 1 to 59, wherein said genetic modification produces a lipid, lipid precursor, or oleochemical at a higher level than by a genetically unmodified yeast cell that is otherwise identical to said genetically modified yeast cell.

76. The genetically modified yeast cell of embodiment 75, wherein said lipid, lipid precursor, or oleochemical produced at a higher level by said genetically modified yeast cell is a fatty acid, wax, sterol, vitamin, monoglyceride, diglyceride, triglyceride, phospholipid, glycerolipid, glycerophospholipid, sphingolipid, saccharolipid, polyketide, sterol lipid, triacylglyceride, prenol lipid, fatty acid ester, fatty acid methyl ester, fatty acid ethyl ester, fatty acid propyl ester, fatty acid butyl ester, fatty alcohol, fatty amine, glycerol, alcohol ethoxylate, alcohol sulfate, or alcohol ether sulfate.

77. The genetically modified yeast cell of any one of embodiments 1 to 76, wherein said genetic modification comprises a mutation relative to the wild type gene.

78. The genetically modified yeast cell of any one of embodiments 1 to 76, wherein said genetic modification comprises a deletion of a portion of a gene.

79. The genetically modified yeast cell of one of embodiments 1 to 78, wherein said yeast cell comprises an increased level of a fatty acid selected from the group consisting of C5:0, C5:1, C5:2, C5:3, C6:0, C6:1, C6:2, C6:3, C7:0, C7:1, C7:2, C7:3, C8:0, C8:1, C8:2, C8:3, C9:0, C9:1, C9:2, C9:3, C10:0, C10:1, C10:2, C10:3, C11:0, C11:1, C11:2, C11:3, C12:0, C12:1, C12:2, C12:3, C13:0, C13:1, C13:2, C13:3, C14:0, C14:1, C14:2, C14:3, C15:0, C15:1, C15:2, C15:3, C16:0, C16:1, C16:2, C16:3, C17:0, C17:1, C17:2, C17:3, C18:0, C18:1, C18:2, C18:3, C19:0, C19:1, C19:2, C19:3, C20:0, C20:1, C20:2, C20:3, C21:0, C21:1, C21:2, C21:3, C22:0, C22:1, C22:2, C22:3, C23:0, C23:1, C23:2, and C23:3, relative to a genetically unmodified yeast cell that is otherwise identical to said genetically modified yeast cell.

80. The genetically modified yeast cell of embodiment 79, wherein said fatty acid is C17:0 C17:1.

81. The genetically modified yeast cell of embodiment 79, wherein said fatty acid is C16:1n9.

82. The genetically modified yeast cell of one of embodiments 1 to 81, wherein said genetic modification is an engineered genetic modification.

83. The genetically modified yeast cell of embodiment 82, wherein said engineered genetic modification comprises modulated expression of a protein.

84. The genetically modified yeast cell of embodiment 82, wherein said engineered genetic modification comprises increased expression of a protein.

85. The genetically modified yeast cell of embodiment 82, wherein said engineered genetic modification comprises decreased expression of a protein.

86. The genetically modified yeast cell of one of embodiments 1 to 81, wherein said genetic modification is associated with exposure to a mutagen.

87. The genetically modified yeast cell of one of embodiments 1 to 86, wherein said genetic modification comprises modulated expression of a protein in a lipid, or lipid precursor, biosynthetic pathway.

88. A method of producing a lipid, lipid precursor, or oleochemical comprising:

1) culturing a yeast cell of any one of embodiments 1 to 87 in a growth medium; and

2) isolating said lipid, lipid precursor, or oleochemical.

89. The method of embodiment 88, wherein said lipid, lipid precursor, or oleochemical is isolated from said yeast cell.

90. The method of embodiment 88, wherein said lipid, lipid precursor, or oleochemical is isolated from the growth medium.

91. The method of any one of embodiments 88 to 90, wherein said growth medium comprises a majority carbon source selected from the group consisting of glucose, glycerol, xylose, fructose, mannose, ribose, sucrose, and lignocellulosic biomass.

92. The method of any one of embodiments 88 to 90, wherein said growth medium comprises lignocellulosic biomass as the majority carbon source.

93. The method of any one of embodiments 88 to 92, wherein said growth medium comprises a carbon source and a nitrogen source wherein said carbon source is at a concentration at least 10-fold greater than the concentration of the nitrogen source (wt/wt).

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94. The method of any one of embodiments 88 to 92, wherein said growth medium comprises a carbon source and a nitrogen source wherein said carbon source is at a concentration at least 16-fold greater than the concentration of the nitrogen source (wt/wt).

95. The method of any one of embodiments 88 to 92, wherein said growth medium comprises a carbon source and a nitrogen source wherein said carbon source is at a concentration at least 320-fold greater than the concentration of the nitrogen source (wt/wt).

96. The method of any one of embodiments 88 to 95, wherein said growth medium comprises micronutrients (e.g. cobalt, iron, magnesium, potassium, zinc, nickel, molybdenum, manganese, copper, or boron).

97. The method of any one of embodiments 88 to 95, wherein said growth medium comprises cobalt in an amount equivalent to 7.5 to 22.5 mg/L CoCl_2 , magnesium in an amount equivalent to 125 to 375 mg/L MgSO_4 , potassium in an amount equivalent to 7.5 to 22.5 mg/L KI , zinc in an amount equivalent to 10 to 30 mg/L $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$, manganese in an amount equivalent to 6 to 18 mg/L $\text{MnSO}_4 \cdot \text{H}_2\text{O}$, boron in an amount equivalent to 6 to 18 mg/L Boric acid, molybdenum in an amount equivalent to 7.5 to 22.5 mg/L $(\text{NH}_4)_2\text{Mo.4H}_2\text{O}$, nickel in an amount equivalent to 6 to 18 mg/L $\text{NiSO}_4 \cdot 6\text{H}_2\text{O}$, iron in an amount equivalent to 10 to 30 mg/L $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, or copper in an amount equivalent to 7.5 to 22.5 mg/L CuSO_4 .

98. The method of any one of embodiments 88 to 95, wherein said growth medium comprises 5.77×10^{-5} M to 1.73×10^{-4} M cobalt, 0.001 M to 0.003 M magnesium, 4.52×10^{-5} M to 1.35×10^{-4} M potassium, 4.05×10^{-5} M to 1.22×10^{-4} M zinc, 3.55×10^{-5} M to 1.06×10^{-4} M manganese, 9.07×10^{-5} M to 2.91×10^{-4} M boron, 3.76×10^{-5} M to 1.10×10^{-4} M molybdenum, 2.28×10^{-5} M to 6.84×10^{-5} M nickel, 3.60×10^{-5} M to 1.08×10^{-4} M iron, or 4.70×10^{-5} M to 1.41×10^{-4} M copper.

99. The method of any one of embodiments 88 to 95, wherein the growth medium comprises iron, copper, and molybdenum.

100. The method of any one of embodiments 88 to 95, wherein said growth medium comprises molybdenum in an amount equivalent to 7.5 to 22.5 mg/L $(\text{NH}_4)_2\text{Mo.4H}_2\text{O}$, iron in an amount equivalent to 10 to 30 mg/L $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, or copper in an amount equivalent to 7.5 to 22.5 mg/L CuSO_4 .

101. The method of any one of embodiments 88 to 95, wherein said growth medium comprises 3.76×10^{-5} M to 1.10×10^{-4} M molybdenum, 3.60×10^{-5} M to 1.08×10^{-4} M iron, or 4.70×10^{-5} M to 1.41×10^{-4} M copper.

102. The method of any one of embodiments 88 to 95, wherein the growth medium comprises copper and nickel.

103. The method of any one of embodiments 88 to 95, wherein said growth medium comprises nickel in an amount equivalent to 6 to 18 mg/L $\text{NiSO}_4 \cdot 6\text{H}_2\text{O}$ or copper in an amount equivalent to 7.5 to 22.5 mg/L CuSO_4 .

104. The method of any one of embodiments 88 to 95, wherein said growth medium comprises 2.28×10^{-5} M to 6.84×10^{-5} M nickel or 4.70×10^{-5} M to 1.41×10^{-4} M copper.

105. The method of any one of embodiments 88 to 95, wherein the growth medium comprises copper, iron, and either molybdenum or nickel.

106. The method of any one of embodiments 88 to 95, wherein said growth medium comprises molybdenum in an amount equivalent to 7.5 to 22.5 mg/L $(\text{NH}_4)_2\text{Mo.4H}_2\text{O}$, nickel in an amount equivalent to 6 to 18 mg/L

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$\text{NiSO}_4 \cdot 6\text{H}_2\text{O}$, iron in an amount equivalent to 10 to 30 mg/L $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, or copper in an amount equivalent to 7.5 to 22.5 mg/L CuSO_4 .

107. The method of any one of embodiments 88 to 95, wherein said growth medium comprises 3.76×10^{-5} M to 1.10×10^{-4} M molybdenum, 2.28×10^{-5} M to 6.84×10^{-5} M nickel, 3.60×10^{-5} M to 1.08×10^{-4} M iron, or 4.70×10^{-5} M to 1.41×10^{-4} M copper.

108. The method of any one of embodiments 88 to 95, wherein the growth medium comprises copper, iron, molybdenum, and nickel.

109. A method of isolating a genetically modified yeast cell from a plurality of yeast cells, comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., 15 lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight, comprising allowing a genetically modified yeast cell to separate from a population of yeast cells within said plurality of yeast cells by floating above said population of yeast cells within an aqueous 20 medium thereby isolating said genetically modified yeast cell, wherein said population of yeast cells comprises a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemicals, or oleochemical) than said genetically modified yeast cell.

110. The method of embodiment 109, wherein said genetically modified yeast cell comprises greater than 30% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, 30 lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

111. The method of embodiment 109, wherein said genetically modified yeast cell comprises greater than 40% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, 35 lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

112. The method of embodiment 109, wherein said genetically modified yeast cell comprises greater than 50% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

113. The method of embodiment 109, wherein said genetically modified yeast cell comprises greater than 60% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

114. The method of any one of embodiments 109 to 113, wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 0.5 vvm (volume per volume per minute).

115. The method of any one of embodiments 109 to 113, wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 1.0 vvm (volume per volume per minute).

116. The method of any one of embodiments 109 to 113, 55 wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 2.0 vvm (volume per volume per minute).

117. The method of any one of embodiments 109 to 113, wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 3.0 vvm (volume per volume per minute).

118. The method of any one of embodiments 109 to 113, wherein said plurality of yeast cells are in a bioreactor with agitation and aeration rates of about 4.0 vvm (volume per volume per minute).

119. The method of any one of embodiments 109 to 118, 65 wherein said aqueous medium comprises a yeast growth

medium, minimal media, complete supplement media, or greater than 50 g/L glucose and less than 5 g/L of a nitrogen source.

120. The method of any one of embodiments 109 to 119, wherein said allowing is performed by centrifugation or simple sedimentation.

121. The method of any one of embodiments 109 to 120, wherein said genetically modified yeast cell was formed by transforming a yeast cell with a recombinant nucleic acid.

122. The method of any one of embodiments 109 to 120, wherein said genetically modified yeast cell was formed by mutagenizing a yeast cell.

123. The method of any one of embodiments 109 to 120, wherein said genetically modified yeast cell is created by first exposing a yeast cell to a mutagen (e.g. a chemical mutagen, radiation, UV, or a biological mutagen).

124. The method of any one of embodiments 109 to 120, wherein said genetically modified yeast cell was formed by mutagenizing a yeast cell.

125. A method of isolating a yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight from a plurality of yeast cells, comprising allowing a yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) to separate from a population of yeast cells within said plurality of yeast cells by floating above said population of yeast cells within an aqueous medium thereby isolating said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical), wherein said population of yeast cells comprises a lower percentage wt/wt of lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) than said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical).

126. The method of embodiment 125, wherein said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) comprises greater than 30% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

127. The method of embodiment 125, wherein said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) comprises greater than 40% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

128. The method of embodiment 125, wherein said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) comprises greater than 50% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

129. The method of embodiment 125, wherein said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) comprises greater than 60% wt/wt lipids, lipid precursors, and/or

oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

130. The method of embodiment 125, wherein said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) comprises greater than 70% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

131. The method of embodiment 125, wherein said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) comprises greater than 80% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

132. The method of embodiment 125, wherein said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) comprises greater than 90% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) in dry weight.

133. The method of one of embodiments 125 to 132, wherein said yeast cell comprising greater than 20% wt/wt lipids, lipid precursors, and/or oleochemicals (e.g., lipid, lipids, lipid precursors, lipid precursor, oleochemicals, or oleochemical) comprises a mutation created by natural genetic drift.

134. The method of any one of embodiments 88 to 95, wherein said growth medium comprises cobalt.

135. The method of any one of embodiments 88 to 95 and 134, wherein said growth medium comprises iron.

136. The method of any one of embodiments 88 to 95 and 134 to 135, wherein said growth medium comprises magnesium.

137. The method of any one of embodiments 88 to 95 and 134 to 136, wherein said growth medium comprises potassium.

138. The method of any one of embodiments 88 to 95 and 134 to 137, wherein said growth medium comprises zinc.

139. The method of any one of embodiments 88 to 95 and 134 to 138, wherein said growth medium comprises nickel.

140. The method of any one of embodiments 88 to 95 and 134 to 139, wherein said growth medium comprises molybdenum.

141. The method of any one of embodiments 88 to 95 and 134 to 140, wherein said growth medium comprises manganese.

142. The method of any one of embodiments 88 to 95 and 134 to 141, wherein said growth medium comprises copper.

143. The method of any one of embodiments 88 to 95 and 134 to 142, wherein said growth medium comprises boron.

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V. EXAMPLES

The following examples are meant to illustrate certain embodiments of the invention and not to limit the scope of the invention described herein.

A. MATERIALS AND METHODS

Base Strains and Media.

E. coli strain DH10B was used for cloning and plasmid propagation. DH10B was grown at 37° C. with constant shaking in Luria-Bertani Broth (Teknova) supplemented with 50 µg/ml of ampicillin for plasmid propagation. Yar-

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Yarrowia lipolytica strain PO1f (ATCC #MYA-2613), a leucine and uracil auxotroph devoid of any secreted protease activity (Madzak et al., 2000), was used as the base strain for all studies. Table 1 contains a list of PO1f derivatives produced in this study. *Y. lipolytica* was cultivated at 30° C. with constant agitation. 2 mL cultures of *Y. lipolytica* used in large-scale screens were grown in a rotary drum (CT-7, New Brunswick Scientific) at speed seven, and larger culture volumes were shaken in flasks at 225 rpm.

YSC media consisted of 20 g/L glucose (Fisher Scientific), 0.79 g/L CSM supplement (MP Biomedicals), and 6.7 g/L Yeast Nitrogen Base w/o amino acids (Becton, Dickinson, and Company). YSC-URA, YSC-LEU, and YSC-LEU-URA media contained 0.77 g/L CSM-Uracil, 0.69 g/L CSM-Leucine, or 0.67 g/L CSM-Leucine-Uracil in place of CSM, respectively. YPD media contained 10 g/L yeast extract (Fisher Scientific), 20 g/L peptone (Fisher Scientific) and 20 g/L glucose, and was often supplemented with 300 µg/ml Hygromycin B (Invitrogen) for knockout selection. Lipid accumulation response towards media formulation was investigated by cultivation in varying concentrations of glucose and nitrogen. These media formulations contained 0.79 g/L CSM, 1.7 g/L Yeast Nitrogen Base w/o amino acid and w/o (NH₄)₂SO₄ (Becton, Dickinson, and Company), between 10 g/L and 320 g/L glucose, and between 0.04 g/L and 10 g/L ammonium sulfate—(NH₄)₂SO₄ (Fisher Scientific). These media are routinely referred to by their ratio of carbon content (glucose) to nitrogen content (ammonium sulfate). For instance, media containing 80 g/L glucose and 5 g/L ammonium sulfate is called C₈₀:N₅ media. When utilizing alternative carbon sources, glucose was replaced by 80 g/L arabinose, 80 g/L fructose, 80 g/L galactose, 80 g/L glycerol (Fisher Scientific), 80 g/L mannose, 80 g/L maltose 80 g/L ribose, 80 g/L sucrose (Acros Organics), 80 g/L Xylose, or 80 g/L of a saccharide mix resembling the composition of lignocellulosic biomass (57% Glucose, 32% Xylose, 5% Arabinose, 3% Mannose, and 3% Galactose by weight). Solid media for *E. coli* and *Yarrowia lipolytica* was prepared by adding 20 g/L agar (Teknova) to liquid media formulations.

When analyzing the effect of micronutrient supplementation, CoCl₂ (15 mg/L), MgSO₄ (250 mg/L), KI (15 mg/L), ZnSO₄·7H₂O (20 mg/L), MnSO₄·H₂O (12.5 mg/L), Boric acid (12.5 mg/L), (NH₄)₂Mo·4H₂O (15 mg/L), NiSO₄·6H₂O (12.5 mg/L), FeSO₄·7H₂O (20 mg/L), or CuSO₄ (15 mg/L) were added to the stated media formulation. Concentrations given are the final concentrations of the metal ion.

Initial Optimization of Media Formulation for Wildtype and Engineered Strains.

Nitrogen starvation is the accepted impetus for effecting a state of lipid accumulation in oleaginous organisms (Rattledge 2002). As a preliminary analysis of this induction potential, we selected seven media variations wildly variant in their ratios' of carbon content (glucose) to nitrogen content (ammonium sulfate) to assay for their ability to induce lipid accumulation. These media formulations are routinely referred to by this carbon to nitrogen ratio (C:N ratio), i.e., media containing 160 g/L glucose and 0.2 g/L ammonium sulfate is called C₁₆₀:N_{0.2} media. We cultivated wildtype *Y. lipolytica* strain PO1f in these seven media formulations and assayed for relative lipid (e.g. triacylglyceride) accumulation using nile red fluorescence flow cytometry after 2, 4, 6, and 8 days. We observed a strong correlation between increasing carbon to nitrogen ratio and increased lipid (e.g. triacylglyceride) accumulation that spanned a 10-fold range, and we were able to increase nile red fluorescence levels by three-fold compared to levels

induced in standard minimal (YSC) media. Thus, we confirmed the beneficial effect of increasing C:N ratio towards lipid (e.g. triacylglyceride) accumulation in non-engineered *Y. lipolytica*, so we sought to further improve oleo-content with additional media supplementation. In particular, FeSO₄ supplementation has been implicated in enabling increased citric acid accumulation in *Y. lipolytica* (Kamzolova et al. 2003), specifically under oxygen limiting conditions. Citric acid and fatty acid accumulation are closely linked in *Y. lipolytica*, so we hypothesized that this iron-responsive citric acid accumulation could also increase downstream lipid (e.g. triacylglyceride) accumulation. To fully analyze the potential benefits of micronutrient addition towards lipid (e.g. triacylglyceride) accumulation (Song et al. 2012; Zhao et al. 2008), we cultivated PO1f in minimal media supplemented with cobalt, magnesium, potassium, zinc, manganese, boric acid, molybdenum, nickel, iron, and copper (FIG. 1), and saw increased lipid (e.g. triacylglyceride) accumulation with iron, nickel, copper, molybdenum, and zinc. We performed a combinatorial screening of iron, nickel, copper, and molybdenum supplementation to detect cumulative beneficial effects towards increasing cellular lipid content. Triple supplementation with copper, nickel, and iron increased lipid accumulation levels to the highest observed at that time (FIG. 2).

Thus, manipulating media formulation effectively increased lipid formulation in a wildtype strain, however, the relationship between strain genotype and this effect has yet to be explored. We sought to determine if a strain rationally engineered for increased lipid accumulation would benefit in the same manner from increasing C:N ratio. In our initial attempts to engineer a *Y. lipolytica* strain for increased lipid accumulation, we overexpressed the AMPDp in a ΔPEX10 background to create a strain with a 17-fold increase in nile red fluorescence levels. To determine if genomic modifications could affect differential responses towards media-induced lipid accumulation, we cultivated unmodified PO1f and our engineered high lipid producer in twenty media formulations that varied in carbon and nitrogen levels (Table 3) and analyzed for lipid content with nile red fluorescence flow cytometry after two days, four days, and eight days. Two days was insufficient time to induce lipid accumulation, while lipid accumulation is evident a majority of media formulation for the PO1f ΔPEX10 AMPDp overexpression strain after eight days. Heat graphs of relative fluorescent values illustrate that the PO1f ΔPEX10 AMPDp overexpression strain accumulates lipids efficiently at an optimum value of 80 g/L glucose after 4 days, while PO1f is only slight induced in any condition, most noticeably after six to eight days in C₁₆₀N_{0.2} media. In general, the 320 g/L glucose condition is too high to induce lipid accumulation effectively, most likely because the high sugar content prevents cell growth. Likewise, formulations 0.04 and 0.2 g/L ammonium sulfate tend to poorly induce lipid accumulation, especially within four days or less. Finally, an optimum C:N ratio of ~10 to 40 can be observed when discounting these highest glucose and lowest ammonium sulfate.

B. CLONING AND TRANSFORMATION PROCEDURES

All restriction enzymes were purchased from New England Biolabs and all digestions were performed according to standard protocols. PCR reactions were set up with recommended conditions using Phusion high fidelity DNA polymerase (Finnzymes), or LongAmp Taq DNA polymerase (New England Biolabs). Ligation reactions were performed

overnight at room temperature using T4 DNA Ligase (Fermentas). Gel extractions were performed using the Fermentas GeneJET extraction kit purchased from Fisher Thermo-Scientific. *E. coli* minipreps were performed using the Zippy Plasmid Miniprep Kit (Zymo Research Corporation). *E. coli* maxipreps were performed using the Qiagen HiSpeed Plasmid Maxi Kit. Transformation of *E. coli* strains was performed using standard electroporator protocols (Sambrook and Russell, 2001). Large amounts of linearized DNA (>20n), necessary for *Y. lipolytica* PO1f transformation were cleaned and precipitated using a standard phenol:chloroform extraction followed by an ethanol precipitation (Kirby, 1956).

Genomic DNA (gDNA) was extracted from *Y. lipolytica* using the Wizard Genomic DNA Purification kit (Promega). Transformation of *Y. lipolytica* with replicative plasmids was performed using the Zymogen Frozen EZ Yeast Transformation Kit II (Zymo Research Corporation), with plating on YSC-LEU plates. Transformation of *Y. lipolytica* PO1f with linearized cassettes was performed as described previously (Blazeck et al. 2013a), with selection on appropriate plates. All auxotrophic or antibiotic selection markers were flanked with LoxP sites to allow for retrieval of integrated markers the pMCS-UAS1B₁₆-TEF-Cre replicative vector (Blazeck et al. 2013a).

Plasmid Construction.

Primer sequences can be found in the Table 2. All *Y. lipolytica* episomal plasmids were centromeric, replicative vectors derived from plasmid pS116-Cen1-1(227) (Yamane et al. 2008) after it had been modified to include a multi-cloning site, a hrGFP green fluorescent reporter gene (pIRES-hrGFP, Agilent) driven by the strong UAS1B₁₆-TEF promoter (Blazeck et al. 2011), and a cycl terminator (Mumberg et al. 1995) to create plasmid pMCS-UAS1B₁₆-TEF-hrGFP. Integrative plasmids were derived from plasmids pUC-S1-UAS1B₁₆-Leum or pUC-S1-UAS1B₁₆-TEF (Blazeck et al. 2013a) that contained 5' and 3' rDNA integrative sequences surrounding the following elements—(from 5' to 3') a uracil section marker surrounded by LoxP sites for marker retrieval, the strong UAS1B₁₆-Leum or UAS1B₁₆-TEF promoter, Ascl and Pad restriction enzyme sites for gene insertion, and a XPR2 minimal terminator. These integrative plasmids were also designed to contain two identical NotI restriction enzyme sites directly outside of the rDNA regions so that plasmid linearization would simultaneously remove *E. coli* pUC19-based DNA. All plasmids containing expression cassettes were sequenced confirmed before transformation into *Y. lipolytica*.

Construction of Episomal Expression Cassettes:

The following genes were PCR amplified from *Y. lipolytica* PO1f gDNA and inserted into vector pMCS-UAS1B₁₆-TEF-hrGFP in place of hrGFP with an Ascl/PacI digest: AMPD, ACL subunit 1 (ACL1), ACL subunit 2 (ACL2), MEA1, DGA1, DGA2, the Tup1 general transcriptional repressor (Morin et al. 2011), and the HAC1 basic leucine zipper transcription factor involved in unfolded protein response (Morin et al. 2011) with primers, respectively. This formed plasmids pMCS-UAS1B₁₆-TEF-AMPD, pMCS-UAS1B₁₆-TEF-ACL1, pMCS-UAS1B₁₆-TEF-ACL2, pMCS-UAS1B₁₆-TEF-MEA, pMCS-UAS1B₁₆-TEF-DGA1, pMCS-UAS1B₁₆-TEF-DGA2, pMCS-UAS1B₁₆-TEF-TUP1, and pMCS-UAS1B₁₆-TEF-HAC1.

Construction of Integrative Expression Cassettes:

The following genes were gel extracted from the previously constructed episomal expression vectors and inserted into vector pUC-S1-UAS1B₁₆-TEF with an Ascl/PacI digest: AMPD, ACL subunit 1 (ACL1), ACL subunit 2

(ACL2), MEA1, DGA1, and DGA2. This formed plasmids pUC-S1-UAS1B₁₆-TEF-AMPD, pUC-S1-UAS1B₁₆-TEF-ACL1, pUC-S1-UAS1B₁₆-TEF-ACL2, pUC-S1-UAS1B₁₆-TEF-MEA1, and pUC-S1-UAS1B₁₆-TEF-DGA1, and pUC-S1-UAS1B₁₆-TEF-DGA2. The loxP-surrounded uracil marker of these integrative plasmids was replaced with a loxP-surrounded leucine marker to enable integrative selection with leucine auxotrophy and co-expression of two enzymes without marker retrieval. These leucine marker

integrative plasmids were dubbed plasmids pUC-S2-UAS1B₁₆-TEF-AMPD, pUC-S2-UAS1B₁₆-TEF-ACL1, pUC-S2-UAS1B₁₆-TEF-ACL2, pUC-S2-UAS1B₁₆-TEF-MEA1, and pUC-S2-UAS1B₁₆-TEF-DGA1, and pUC-S2-UAS1B₁₆-TEF-DGA2.

10 ACL1 and ACL2 were similarly inserted into pUC-S1-UAS1B₁₆-Leum with primers, respectively, to form plasmids pUC-S1-UAS1B₁₆-Leum-ACL1 and pUC-S1-UAS1B₁₆-Leum-ACL2.

Strain Construction.

15 All strains were confirmed through gDNA extraction and PCR confirmation and are listed in Table 1. We previously constructed two markerless single-gene deletion strains in the *Y. lipolytica* PO1f background, PO1f-Δmfe1 and PO1f-Δpex10, deficient in their β-oxidation and peroxisomal biogenesis capacity, respectively (Blazeck et al. 2013a). Following our previous protocol, the PEX10 gene was deleted from strain PO1f-Δmfe1 to form the markerless double mutant PO1f-Δmfe1-Δpex10. These four strains, PO1f, PO1f-Δmfe1, PO1f-Δpex10, and PO1f-Δmfe1-Δpex10 were utilized as backgrounds for single and double overexpression of the AMPD, ACL1, ACL2, MEA, DGA1, and DGA2 genes, including variation in selective marker utilized, i.e., leucine (S2 integrative cassette or pMCS episomal cassette) vs. uracil (S1 integrative cassette). S2 and S1 integrative cassettes were linearized, transformed into our four background strains, and selected for on appropriate dropout plates. Table 1 contains a list of rationally engineered strains derived in this manner. ORF-less plasmids pUC-S1-UAS1B₁₆-TEF and pUC-S1-UAS1B₁₆-TEF were utilized to create strains lacking leucine, uracil, or both leucine and uracil auxotrophies, dubbed S1-Ø, S2-Ø, and S1-S2-Ø (Table 1).

Combinatorial Genome Engineering.

Prior engineering efforts have successfully increased lipid 20 accumulation in *Y. lipolytica* by manipulating fatty acid, lipid, or central carbon metabolism, but no attempt has been made to simultaneously alter these metabolic functionalities (Beopoulos et al. 2008; Dulermo and Nicaud 2011; Tai and Stephanopoulos 2013). We sought to concurrently control these aspects of lipid synthesis by overexpressing three enzymes that control metabolic flux from central carbon metabolism into fatty acid synthesis (AMPDp, ACLp, and MEA1p) or two isozymes that control lipid synthesis (DGA1p and DGA2p) in four genomic backgrounds with 25 altered fatty acid catabolic ability. These four genomic backgrounds included the PO1f (WT) strain, a PO1f MFE1 deletion strain (AMFE1), a PO1f PEX10 deletion strain (APEX10), and a MFE1 PEX10 double knockout strain (APEX10ΔMFE1). The majority of enzymatic overexpressions were driven by the high strength UAS1B₁₆-TEF constitutive promoter (Blazeck et al. 2011), were integrated into *Y. lipolytica*'s genomic rDNA repeats (Blazeck et al. 2013a; Ledall et al. 1994), and alleviated either PO1f's uracil or leucine auxotrophy. In our previous work, we noticed that 30 alleviation of the leucine auxotrophy tended to increase lipid (e.g. triacylglyceride) accumulation far more than alleviation of the uracil auxotrophy. Therefore, nearly identical

strains were routinely created differing only in the marker utilized to integrate an enzymatic overexpression cassette, enabling either uracil synthesis (S1) or leucine synthesis (S2). Initial overexpressions of the DGA1p and DGA2p enzymes occurred episomally with an identical UAS1B₁₆-TEF promoter on a leucine-marker containing plasmid, though final strain construction entailed integrating these cassettes. Strain names included background (WT, ΔMFE1, ΔPEX10, or ΔPEX10ΔMFE1), markers used (S1, S2, S1-S2, or pMCS), and enzymes overexpressed (AMPD, MEA, ACL1, ACL2, DGA1, DGA2) so a strain overexpressing the AMPDp enzyme with a leucine marker in the ΔPEX10ΔMFE1 background is called ΔPEX10ΔMFE1 S2-AMPD. S1-Ø, S2-Ø, and S1, 2-Ø refer to strains without protein overexpressions but with uracil, leucine, or uracil+leucine auxotrophies alleviated. ACL1p and ACL2p form a heterodimer in vivo so were tested as concurrent overexpressions.

Our combinatorial approach generated over 46 distinct genotypes that were analyzed for lipid (e.g. triacylglyceride) accumulation with nile red fluorescence flow cytometry after four days growth in C₈₀N₅ media and produced a large range in lipid (e.g. triacylglyceride) accumulation ability, culminating in a 60-fold improvement over PO1f WT control (FIG. 3). We saw that the deletion of the pex10 peroxisomal biogenesis transcription factor combined with overexpression of an acyl-CoA:diacylglycerol acyltransferase (DGA1 or DGA2) are essential for the highest lipid (e.g. triacylglyceride) production (FIG. 3). When comparing ammonia depletion in PO1f WT and our highest lipid producer, ΔPEX10ΔMFE1 pMCSDGA1, we observed a pronounced reduction in steady state nitrogen concentration in the ΔPEX10ΔMFE1 pMCSDGA1 strain. We saw a very noticeable correlation between the ability to synthesize leucine and lipid (e.g. triacylglyceride) accumulation ability, with an average increase of five fold in lipid content between comparable strains with and without a leucine marker present (FIG. 4). Deletion of mfe1 drastically reduced this increase in lipid (e.g. triacylglyceride) content. ΔMFE1 and ΔPEX10ΔMFE1 saw only a 1.42 fold and 2.58 fold increases in lipid (e.g. triacylglyceride) content granted from the capacity to synthesize leucine compared to 8.16 and 7.45 fold increases in WT and ΔPEX10 backgrounds (FIG. 4). In three of our four backgrounds, DGA1p outperformed DGA2p (FIG. 3); WT pMCSDGA2 was not included, but subsequent testing showed WT pMCSDGA1 to give higher lipid (e.g. triacylglyceride) levels than WT pMCSDGA2. Overall, fluorescence levels were highest in the ΔPEX10 and ΔPEX10ΔMFE1 backgrounds (~3-fold WT), and lowest in the ΔMFE1 background (~65% of WT), although mfe1 deletion has been shown to increase lipid (e.g. triacylglyceride) accumulation in media containing higher C:N ratio in eight day cultivation periods (Blazeck et al. 2013a). Because mfe1 deletion should further inhibit fatty acid degradation in the ΔPEX10ΔMFE1 background in long-scale fermentations, the DGA1p was integrated into the ΔPEX10ΔMFE1 background with S2 cassette and a S1-Ø to form our final fully heterotrophic rationally engineered strain. This ΔPEX10ΔMFE1 S1-S2-DGA1 strain displayed similar lipid (e.g. triacylglyceride) content to strains containing episomally expressed DGA1p and could accumulate lipids (e.g. triacylglyceride) effectively without any amino acid supplementation (Table 4) and yielded a highest % lipid (e.g. triacylglyceride) content of 32% dry cell weight for a total of 1.32 g/L. Furthermore, we saw no significant difference in LEU3 or DGA1 mRNA levels between these two strains.

During bioreactor runs, these strains are able to produce significant amounts of lipids and cells exhibit 88% by dry cell weight lipids. Improved lipid production with one of the highest producing strains, ΔPEX10ΔMFE1-S1-S2-DGA1 in a bioreactor. Lipid levels have reached 22 g/L in media containing only 80 g/L glucose, 5 g/L ammonium sulfate, and 1.7 g/L Yeast Nitrogen Base (without amino acids or ammonium sulfate). Increasing dissolved oxygen content and maintaining pH at or above 5.0 enabled this yield. This represents ~86% of the theoretical yield. Furthermore, in these strains, we identify the presence of unique C17 fatty acids (FIG. 15).

Complex control of cellular processes, like lipid accumulation, is coordinated by transcription factors that regulate gene networks. In particular, the TupI general transcriptional repressor and the Had leucine zipper transcription factor involved in unfolded protein response have been shown to be upregulated in lipid (e.g. triacylglyceride) accumulation cell states (Morin et al. 2011). However, overexpression of these two proteins decreased lipid (e.g. triacylglyceride) accumulation in the PO1f WT background.

Dissection of Genotype-Dependence Towards Media Induction.

We more fully examined how C:N ratio and genotype interacted towards enabling lipid (e.g. triacylglyceride) accumulate on a larger scale by examining the response of twelve strains grown in thirteen different C:N ratios (Table 5). We were pleased to observe a strong tendency towards high lipid (e.g. triacylglyceride) levels in most high producers at a single media formulation—C₈₀N₅ (FIG. 5-8), allowing us to pinpoint a formulation for later use. Two trends stand out—(1) The 0.2 g/L ammonium sulfate formulations rarely enable lipid (e.g. triacylglyceride) accumulation, so that (2) the difference in induction from media containing 1 g/L and 5 g/L is slight, making glucose concentration seem more important towards increasing content than nitrogen content (after a certain threshold is reached).

Lipid Accumulation on Multiple Carbon Sources.

Viability of lipid (e.g. triacylglyceride) production depends on the capacity to fully convert all sugars from lignocellulosic biomass to lipids or to use carbon from industrial waste streams for lipid production. We analyzed the ability PO1f WT, ΔPEX10 S1-MEA, ΔPEX10 S2-AMPD, ΔPEX10ΔMFE1 S2-DGA1, and ΔPEX10ΔMFE1 pMCSDGA1 to generate lipids (e.g. triacylglyceride) when utilizing glucose, glycerol, xylose, fructose, mannose, ribose, sucrose, or a lignocellulosic sugar blend as their carbon source (FIG. 9). ΔPEX10ΔMFE1 S2-DGA1 and ΔPEX10ΔMFE1 pMCSDGA1 generated the highest lipid (e.g. triacylglyceride) content across the board under conditions tested, and all engineered strains demonstrated the capacity to utilize each carbon source for lipid (e.g. triacylglyceride) production. Glucose, mannose, and the lignocellulosic saccharide blend were utilized easiest while ribose utilizations generated the least lipid (e.g. triacylglyceride) content of the conditions tested. The PO1f WT and ΔPEX10ΔMFE1 S2-DGA1 strain were tested to determine if decreasing carbon content or increasing initial inoculum amount could increase xylose-generate lipid (e.g. triacylglyceride) accumulation. Increasing xylose concentration and decreasing inoculum amount increased lipid (e.g. triacylglyceride) content in the ΔPEX10ΔMFE1 S2-DGA1 strain, while little difference was noticeable in the PO1f WT strain. However, PO1f WT demonstrated a surprising capacity to utilize pure glycerol for lipid (e.g. triacylglyceride) generation.

Isolation of a Novel MGA2 Mutation with Whole Genome Sequencing.

During the screening of a gDNA overexpression library intended to increase *Y. lipolytica*'s lipid (e.g. triacylglyceride) production, we isolated a strain, dubbed L36, with incredible lipid (e.g. triacylglyceride) accumulation ability (FIG. 10). L36's lipid (e.g. triacylglyceride) production could be enhanced with micronutrient supplementation (FIG. 10). Complete sequencing of the L36 genome revealed a missense mutation in the MGA2 lipid synthesis regulator (MGA2G643R) as the most likely potential cause for L36's lipid (e.g. triacylglyceride) production capacity. Overexpression of a truncated MGA2p in a PO1f WT background reconstituted 58% of the observed L36 phenotype.

Directed Evolution with EMS Mutagenesis to Increase Lipid Accumulation

Direct evolution is commonly utilized to increase growth rate or to decrease sensitivity to a toxic metabolite. However, directed evolution has never been evaluated as a tool to increase lipid (e.g. triacylglyceride) production in oleaginous organisms. As evidenced by the isolation of strain L36, it is likely that *Y. lipolytica* is amenable to this approach. We subjected both L36 (FIG. 11) and ΔPEX10ΔMFE1 S2-DGA1 to EMS mutagenesis followed by serial selection via subculturing and then nile red staining. Both backgrounds proved highly responsive towards the directed evolution approach, and an increase in fluorescence with a large increase in final cell concentration (Table 6).

Besides the minerals, during the experiments, we also observed a critical phenotype for lipid (e.g. triacylglyceride) production in *Yarrowia lipolytica*: the lipid (e.g. triacylglyceride) de novo lipid (e.g. triacylglyceride) accumulation is close related to leucine biosynthesis pathway. A 5 fold lipid (e.g. triacylglyceride) level increase was achieved with strain harboring complete LEU biosynthesis pathway comparing to the one without complete pathway. Although this phenotype has been reported with engineered *Saccharomyces cerevisiae* (Kamisaka et al. 2007), this is the first observation in oleaginous yeast to our best knowledge. Understanding of this phenotype could be essential to understand the basic differences between oleaginous microbes and normal ones. However, to the date, the fundamental reason is still missing. Two possible routes may contribute to this, one is through TOR pathway (Kim and Guan 2011; Laplante and Sabatini 2009) and the other one is through leucine degradation and ketone body generation (Endemann et al. 1982). Either pathway heavily interacts with the whole cell metabolism which requires deep analysis to reveal the true mechanism behind.

Engineering with Known: Biosynthesis pathways and basic regulations. Rational systematic engineering *Yarrowia lipolytica* for high lipid production. Engineering with Unknown: Pathway interactions and complex regulation networks. Engineering lipid production in *Yarrowia lipolytica* through Inverse combinatorial metabolic Engineering. Confirmed lipid enhancers include DGA1 (Diacylglycerol acyltransferase) 300% improvement, MRM2 (Mitochondrial 2' O-ribose methyltransferase) 25% improvement, MGMT (O-6-methylguanine-DNA methyltransferase) 15% improvement.

C. FATTY ACID CHARACTERIZATION BY NILE RED STAINING COUPLE WITH FLOW CYTOMETRY OR FLUORESCENCE MICROSCOPY

Nile Red is commonly utilized to stain oleaginous cellular material, and can be coupled with fluorescence flow cytometry to gauge relative lipid content (Greenspan et al. 1985).

Y. lipolytica strains were routinely inoculated from glycerol stock in biological triplicate in appropriate media for 72 hours at 30° C. with shaking. Cell concentrations were normalized to a specific OD₆₀₀ for reinoculation in fresh media and further incubation. In general, 2 mL cultures were inoculated to an OD₆₀₀=2.5, and larger volume cultures were inoculated to an OD₆₀₀=0.1. Cultures were incubated for two to eight days at 30° C. with constant agitation. 2 mL cultures were incubated in a rotary drum (CT-7, New Brunswick Scientific) at speed seven and flasks were shaken at 225 rpm in a standing incubator. To harvest, one OD₆₀₀ unit of each culture was spun down at 1000 g for three minutes and resuspended in 500 μL Phosphate Buffered Saline solution (PBS) (Sigma Aldrich). 6 μL of 1 mM Nile Red (dissolved in DMSO) was added, and then cells were incubated in the dark at room temperature for 15 minutes. Cells were spun down at 1000 g for three minutes, resuspended in 800 μL ice cold water, spun down again, and resuspended again in 800 μL ice cold water. 300 μL of stained cells were added to 1 ml ice cold water and tested with a FACS Fortessa (BD Biosciences), a voltage of 350, a 10,000 cell count, a forward scatter of 125, a side scatter of 125, and the 535LP and 585/42BP filters for fluorescence detection using the GFP fluorochrome. Samples were kept on ice and in the dark during the test and the data was analyzed using FlowJo software (Tree Star Inc., Ashland, Oreg.) to compute mean fluorescence values. Day-to-day variability was mitigated by analyzing all comparable strains on the same day. An average fluorescence and standard deviation were calculated from the mean values of biological replicates. Stained cells were routinely examined with fluorescence microscopy under a 100× oil immersion objective using the FITC channel on an Axiovert 200M microscope (Zeiss).

D. LIPID QUANTIFICATION AND FATTY ACID PROFILE ANALYSIS

Lipids from ~20-30 OD₆₀₀ equivalents were extracted following the procedure described by (Folch et al. 1957) and modified for yeast (Schneiter and Daum 2006). Dried lipids were transesterified with N-tert-Butyldimethylsilyl-N-methyltrifluoroacetamide (Sigma-Aldrich) following the procedure of (Paik et al., 2009), and 2 μL samples were injected into a GC-FID (Agilent Technologies 6890 Network GC System) equipped with an Agilent HP-5 column (5% phenyl-95% methylsiloxane—product number 19091J-413) to analyze fatty acid fractions. Briefly, the following settings were used: Detector Temp=300° C., He Flow=1.0 mL/min, Oven Temp=80° C. for 2 min, increased at 30° C./min to 200° C., increased at 2° C./min to 229° C., increased at 1° C./min to 232° C., increased at 50° C./min to 325° C. Fatty acid standards for C16:0 palmitic acid, C16:1(n-7) palmitoleic acid, C18:0 stearic acid, C18:1 (n-9) oleic acid, and C18:2 (n-6) linoleic acid were purchased from Sigma-Aldrich, transesterified, and analyzed by GC to identify fatty acid peaks.

E. CITRIC ACID QUANTIFICATION

A 2 mL culture sample was pelleted down for 5 minutes at 3000×g, and the supernatant was filtered using a 0.2 mm syringe filter (Corning Incorporated). Filtered supernatant was analyzed with a HPLC Ultimate 3000 (Dionex) and a Zorbax SB-Aq column (Agilent Technologies). A 2.0 μL injection volume was used in a mobile phase composed of a 99.5:0.5 ratio of 25 mM potassium phosphate buffer (pH=2.0) to acetonitrile with a flow rate of 1.25 mL/min.

The column temperature was maintained at 30° C. and UV-Vis absorption was measured at 210 nm. A citric acid standard (Sigma-Aldrich) was used to detect and quantify citric acid production.

F. EMS MUTAGENESIS AND ISOLATION OF HIGH LIPID PRODUCING STRAINS

10 OD units from cultures grown overnight were spun down in sterile microcentrifuge tubes at 5000 g for 10 seconds. Cell pellets were resuspended in 1 mL H₂O, repelleted, and resuspended in 1 mL PBS. Two samples were spun down from each culture, one for EMS mutagenesis (30 µl of EMS added) and one as a control to determine the prevalence of spontaneous beneficial mutation (no EMS added). Cells were incubated for 1 hr at 30° C., with agitation, pelleted and resuspended in 200 µl of 5% sodium thiosulfate, transferred to fresh microcentrifuge tubes, washed twice in 200 µl of 5% sodium thiosulfate, and resuspended in 1 mL H₂O. Cells were then grown to stationary phase in YSC media, and then reinoculated at an OD₆₀₀=2.5 in 1 mL C₈₀N₅ media and grown for four days. Three to six serial transfers of the cell cultures followed in which the 1 mL cultures were spun down at 1000 g for two minutes, and the top 200 µL of the supernatant was transferred to 1 mL of fresh YSC media and allowed to grow to stationary phase before again spinning down and transferring. Final cultures (top 200 µL after spin down) were plated on YSC plates containing 0.01 mM Nile Red. After four days, high lipid producers were selected by viewing plates under a blue fluorescent light and picking colonies with brighter pink fluorescent color. Lipid amount was determined by coupling Nile Red staining with flow cytometry as described above.

The EMS mutagenesis procedures were performed following the protocol described by Winston (Winston 2001). Briefly, an overnight culture was cultivated to OD about 10. Cells were then harvested, washed and resuspended with 0.1 M sodium phosphate buffer (pH 7). 30 µl of EMS were added and incubated with unmutagenized control for 1 hr at 30° C., with agitation. The cells were then washed with 5% sodium thiosulfate and ready for serial transfer experiments to enrich the high lipid population. The EMS treated cells and unmutagenized cells were first cultured YSC media for 72 hours and then cultured in high glucose media with starting OD at 2.5 for 96 hours. The cells were centrifuged down with 100 g, the unclear supernatant, which contains high lipid accumulation strains, was used as seed for another round of cultivation. After five rounds of transfer, the cells were plated on Nile Red YSC plate to facilitate the isolation of high lipid production strains. Individual colonies were picked from the EMS treated cells as well as unmutagenized cells for characterization.

Characterization of EMS mutagenesis and floating cell transfer selection procedure selected strain E13 and E26. Second generation sequencing platform illumina paired ended sequencing PE 2×100 were performed with genomic DNA extracted from strain E26, E13 as well as PO1f by Genomic Sequencing and Analysis Facility in The University of Texas at Austin. 6424381 reads for strain E26 and 6565093 reads for strain E13 were collected from illumina HiSeq, which lead to a coverage approximately 65x. The Illumina reads were mapped to the CLIB122 genome using BWA (Li and Durbin 2009) and analyzed with Samtools (Beopoulos, Cescut et al. 2009) and BEDTools (Quinlan and Hall 2010). The SNPs identified were then filtered with

SnpSift with QUAL>=30 (Pablo, Viral et al. 2012). The SNPs identified from PO1f, EMS26 and EMS13 were compared to extract the authentic SNPs in EMS26 and EMS13. The identified SNPs were then visualized in the IGV genome visualization software to validate as well as study the location of the SNPs in the genome due to the high false error rate in SNP calling process (Liu, Guo et al. 2012).

Information on identified targets in E26 and E13 strains following mutagenesis. Succinate semialdehyde dehydrogenase (SSADH), which converts succinate semialdehyde to succinate after UGA1,4-aminobutyrate aminotransferase, deaminates GABA to succinate (Ramos, El Guezzar et al. 1985). Higher levels of accumulation of α-ketoglutarate were found in uga2 mutants in *Saccharomyces cerevisiae* (Cao, Barbosa et al. 2013) (3VZ1; 3VZ3). In the same time, lower levels of succinic acid (more than 5 fold decrease) were also identified in the yeast (Kamei, Tamura et al. 2011). The identified mutation in UGA2 in sequenced strains of Proline 209 is a highly conserved residual and close to a hydrogen bond forming Serine (Yuan, Yin et al. 2013). 15 GABA metabolism is closely related to nitrogen assimilation in yeast and nitrogen limitation has been studied as a key function for triggering lipogenesis in *Yarrowia lipolytica* (Beopoulos, Cescut et al. 2009). Nitrogen sources have also been proven as an important factor for lipid accumulation 20 inside cells (Evans and Ratledge 1984). A relationship between GABA metabolism and the TOR pathway, an important signaling pathway for lipid accumulation (Blazeck, Hill et al. 2014), has also been suggested (Cardenas, Cutler et al. 1999; Staschke, Dey et al. 2010). 25 YALI0E17215 g codes for a protein with similarity to *Saccharomyces cerevisiae* RME1, which is a zinc finger protein involved in the control of meiosis (Covitz, Herskowitz et al. 1991). A similar protein has shown significant levels of increase in mRNA levels in a lipid accumulation-improved snf1 mutant in *Yarrowia lipolytica* (Xue, Sharpe et al. 30 35 40 45 50 55 60 65 70 75 80 85 90 95 100 105 110 115 120 125 130 135 140 145 150 155 160 165 170 175 180 185 190 195 200 205 210 215 220 225 230 235 240 245 250 255 260 265 270 275 280 285 290 295 300 305 310 315 320 325 330 335 340 345 350 355 360 365 370 375 380 385 390 395 400 405 410 415 420 425 430 435 440 445 450 455 460 465 470 475 480 485 490 495 500 505 510 515 520 525 530 535 540 545 550 555 560 565 570 575 580 585 590 595 600 605 610 615 620 625 630 635 640 645 650 655 660 665 670 675 680 685 690 695 700 705 710 715 720 725 730 735 740 745 750 755 760

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TABLE 1

| List of select strains used in this study | | |
|---|--|---------------------|
| Host Strain Name | Genotype | Reference or Source |
| <i>Escherichia coli</i> strains | | |
| DH10B | F ⁻ mcrA Δ(mrr-hsdRMS-mcrBC) φ80d ^r /acZΔM15 ^r ΔlacX74 endA1 recA1 deoR Δ(ara, leu)7697 araD139 galU galK nupG rpsL λ | Open Biosystems |

TABLE 1-continued

| List of select strains used in this study | | |
|--|--|---------------------|
| Host Strain Name | Genotype | Reference or Source |
| <i>Yarrowia lipolytica</i> base strains | | |
| WT (PO1f) | MatA, leu2-270, ura3-302, xpr2-322, axp1-2 | Madzak et al. 2000 |
| AMFE1 (PO1f-Amfe1) | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1 | Blazeck et al. 2013 |
| APEX10 (PO1f-Apex10) | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δapex10 | Blazeck et al. 2013 |
| APEX10ΔMFE1 (PO1f-Δapex10-Δmfe1) | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δapex10, Δmfe1 | This work |
| ΔACO1 (PO1f-Δaco1) | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δaco1 | This work |
| Selected <i>Yarrowia lipolytica</i> overexpression strains | | |
| WT-S1-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, URA3 (S1) | This work |
| WT-S2-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (S2) | This work |
| WT-S1-S2-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, URA3, LEU2 (S1, S2) | This work |
| WT-pMCS | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (pMCS) | This work |
| WT-pMCS-TUP1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (pMCS), UAS1B16-TEF-TUP1 | This work |
| WT-pMCS-HAC1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (pMCS), UAS1B16-TEF-HAC1 | This work |
| WT-S1-AMPD | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, URA3 (S1), UAS1B ₁₆ -TEF-AMPD | This work |
| WT-S2-AMPD | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (S2), UAS1B ₁₆ -TEF-AMPD | This work |
| WT-S1-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, URA3 (S1), UAS1B ₁₆ -TEF-MEA1 | This work |
| WT-S2-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (S2), UAS1B ₁₆ -TEF-MEA1 | This work |
| WT-S1-S2-AMPD-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, URA3, LEU2 (S1, S2), UAS1B16-TEF-AMPD, UAS1B16-TEF-MEA1 | This work |
| WT-pMCS-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (pMCS), UAS1B ₁₆ -TEF-DGA1 | This work |
| AMFE1-S1-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, URA3 (S1) | This work |
| AMFE1-S2-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, LEU2 (S2) | This work |
| AMFE1-S1-S2-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, URA3, LEU2 (S1, S2) | This work |
| AMFE1-S1-AMPD | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, URA3 (S1), UAS1B16-TEF-AMPD | This work |
| AMFE1-S2-AMPD | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, LEU2 (S2), UAS1B16-TEF-AMPD | This work |
| AMFE1-S1-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, URA3 (S1), UAS1B16-TEFMEA1 | This work |
| AMFE1-S2-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, LEU2 (S2), UAS1B16-TEF-MEA1 | This work |
| AMFE1-S1-S2-AMPD-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, URA3, LEU2 (S1, S2), UAS1B16-TEF-AMPD, UAS1B16-TEF-MEA1 | This work |
| AMFE1-S1-S2-ACL1-ACL2 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, URA3, LEU2 (S1, S2), UAS1B16-TEF-ACL1, UAS1B16-TEF-ACL2 | This work |
| AMFE1-pMCS-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, LEU2 (pMCS), UAS1B ₁₆ -TEF-DGA1 | This work |
| AMFE1-pMCS-DGA2 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δmfe1, LEU2 (pMCS), UAS1B ₁₆ -TEF-DGA2 | This work |
| APEX10-S1-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δapex10, URA3 (S1) | This work |
| APEX10-S2-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δapex10, LEU2 (S2) | This work |
| APEX10-S1-S2-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δapex10, URA3, LEU2 (S1, S2) | This work |
| APEX10-S1-AMPD | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δapex10, URA3 (S1), UAS1B16-TEF-AMPD | This work |
| APEX10-S2-AMPD | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δapex10, LEU2 (S2), UAS1B16-TEF-AMPD | This work |
| APEX10-S1-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δapex10, URA3 (S1), UAS1B16-TEFMEA1 | This work |

TABLE 1-continued

| List of select strains used in this study | | |
|---|---|---------------------|
| Host Strain Name | Genotype | Reference or Source |
| ΔPEX10-S2-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, LEU2 (S2), UAS1B ₁₆ -TEF-MEA1 | This work |
| ΔPEX10-S1-S2-AMPD-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, URA3, LEU2 (S1, S2), UAS1B ₁₆ -TEF-AMPD, UAS1B ₁₆ -TEF-MEA1 | This work |
| ΔPEX10-pMCS-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, LEU2 (pMCS), UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔPEX10-pMCS-DGA2 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, LEU2 (pMCS), UAS1B ₁₆ -TEF-DGA2 | This work |
| ΔPEX10ΔMFE1-S1-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3 (S1) | This work |
| ΔPEX10ΔMFE1-S2-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, LEU2 (S2) | This work |
| ΔPEX10ΔMFE1-S1-S2-Ø | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3, LEU2 (S1, S2) | This work |
| ΔPEX10ΔMFE1-S1-AMPD | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3 (S1), UAS1B ₁₆ -TEF-AMPD | This work |
| ΔPEX10ΔMFE1-S2-AMPD | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, LEU2 (S2), UAS1B ₁₆ -TEF-AMPD | This work |
| ΔPEX10ΔMFE1-S1-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3 (S1), UAS1B ₁₆ -TEF-MEA1 | This work |
| ΔPEX10ΔMFE1-S2-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, LEU2 (S2), UAS1B ₁₆ -TEF-MEA1 | This work |
| ΔPEX10ΔMFE1-S1-S2-AMPD-MEA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3, LEU2 (S1, S2), UAS1B ₁₆ -TEF-AMPD, UAS1B ₁₆ -TEF-MEA1 | This work |
| ΔPEX10ΔMFE1-S1-S2-ACL1-ACL2 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3, LEU2 (S1, S2), UAS1B ₁₆ -TEF-ACL1, UAS1B ₁₆ -TEF-ACL2 | This work |
| ΔPEX10ΔMFE1-pMCS-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, LEU2 (pMCS), UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔPEX10ΔMFE1-pMCS-DGA2 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, LEU2 (pMCS), UAS1B ₁₆ -TEF-DGA2 | This work |
| ΔPEX10ΔMFE1-S2-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, LEU2 (S2), UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔPEX10ΔMFE1-S1-Ø-S2-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3 (S1), LEU2 (S2), UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔPEX10ΔMFE1-S1-AMPD-S2-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3 (S1), LEU2 (S2), UAS1B ₁₆ -TEF-AMPD, UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔPEX10ΔMFE1-S1-MEA1-S2-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3 (S1), LEU2 (S2), UAS1B ₁₆ -TEF-MEA1, UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔPEX10ΔMFE1-S1-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3 (S1), UAS1B ₁₆ -TEF-DGA1 | This work |
| WT-S2-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Leu2 (S2), UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔPEX10ΔMFE1-S1-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3 (S1), UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔPEX10-S2-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, LEU2 (S2), UAS1B ₁₆ -TEF-DGA1 | This work |
| WT-pMCS-DGA2 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (pMCS), UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔPEX10ΔMFE1-S1-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3, UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔMFE1-S2-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Amfe1, LEU2(S2), UAS1B ₁₆ -TEF-DGA1 | This work |
| ΔMFE1-S2-DGA2 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Amfe1, LEU2 (S2), UAS1B ₁₆ -TEF-DGA2 | This work |
| ΔPEX10ΔMFE1-S1-Ø-pMCS-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δpex10, Amfe1, URA3 (S1), LEU2 (pMCS), UAS1B ₁₆ -TEF-DGA1 | This work |
| Po1f pMCSMga2 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2 LEU2 (pMCS), UAS1B ₁₆ -TEF-Mga2 | This work |
| Po1f pMCSMga2dTm | MatA, leu2-270, ura3-302, xpr2-322, axp1-2 LEU2 (pMCS), UAS1B ₁₆ -TEF-Mga2dTm (truncated of transmembrane span) | This work |

TABLE 1-continued

| List of select strains used in this study | | |
|---|---|---------------------|
| Host Strain Name | Genotype | Reference or Source |
| Po1f pMCSMga2L36 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2 LEU2 (pMCS), UAS1B ₁₆ -TEF-Mga2L36 (has SNP found in L36 strain) | This work |
| Po1f pMCSMRM2 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (pMCS), UAS1B ₁₆ -TEF-MRM2 | This work |
| Po1f pMCSO6M | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (pMCS), UAS1B ₁₆ -TEF-O6M | This work |
| ΔACO1-DGA1 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, Δaco1, URA3 (S1), LEU2 (S2), UAS1B ₁₆ -TEF-DGA1 | This work |
| L36 and EMS derived strains | | |
| L36 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, LEU2 (pMCS) - isolated and fully sequenced to determine source of high lipid accumulation - most likely from mutation in MGA2 ORF. | This work |
| L36 E1S6-4 | L36 strain mutagenized further with EMS | This work |
| L36 E1S6-5 | L36 strain mutagenized further with EMS | This work |
| L36 E1S6-6 | L36 strain mutagenized further with EMS | This work |
| APEX10ΔMFE1-S2-DGA1 E1 | APEX10ΔMFE1-S2-DGA1 strain mutagenized with EMS | This work |
| APEX10ΔMFE1-S2-DGA1 E6 | APEX10ΔMFE1-S2-DGA1 strain mutagenized with EMS | This work |
| APEX10ΔMFE1-S2-DGA1 E12 | APEX10ΔMFE1-S2-DGA1 strain mutagenized with EMS | This work |
| E13 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, ΔapeX10, Δmfe1, URA3 (S1), LEU2 (S2), UAS1B ₁₆ -TEF-DGA1 strain mutagenized with EMS and selected | This work |
| E26 | MatA, leu2-270, ura3-302, xpr2-322, axp1-2, ΔapeX10, Δmfe1, URA3 (S1), LEU2 (S2), UAS1B ₁₆ -TEF-DGA1 strain mutagenized with EMS and selected | This work |

TABLE 2

| List of primers used in this study | |
|------------------------------------|--|
| JB387 YL AMPD 5' | TTGGCGCGCCatggcccgaccaaggaaatgg (SEQ ID NO.: 1) |
| AscI | |
| JB388 YL AMPD 3' | CCTTAATTAAattaacccatgcagccgctcaaac (SEQ ID NO.: 2) |
| PacI | |
| JB402 YL ACL1 5' | TTGGCGCGCCatgtctgccaacgagaacat (SEQ ID NO.: 3) |
| AscI | |
| JB403 YL ACL1 + 4 5' AscI | TTGGCGCGCtctgccaacgagaacatctc (SEQ ID NO.: 4) |
| PacI | |
| JB404 YL ACL1 3' | CCTTAATTAAactatgatcgagtcttgcccttg (SEQ ID NO.: 5) |
| PacI | |
| JB405 YL ACL2 5' | TTGGCGCGCCATGTCAGCGAAATCCATTCAAG (SEQ ID NO.: 6) |
| AscI | |
| JB406 YL ACL2 + 4 5' AscI | TTGGCGCGCCTCAGCGAAATCCATTCAAG (SEQ ID NO.: 7) |
| PacI | |
| JB407 YL ACL2 3' | CCTTAATTAAATTAAACTCCGAGAGGGAGTGGAA (SEQ ID NO.: 8) |
| PacI | |
| JB862 Loxleu 5' | CCAccgcggataacttcgtataatgtatgtatcgaagtttatggatggaaaga (SEQ ID NO.: 9) |
| SacII | |
| JB863 Loxleu 3' | CGGTTCGAAataacttcgtatacatacattatacgaagttatcagtcggccagcttaagatata (SEQ ID NO.: 10) |
| BstBI | |
| JB865 hygR 3' | GgaacggTAGATCtCGAGCGTCCAAAACCTTCTC (SEQ ID NO.: 11) |
| bgIII | |

TABLE 2-continued

List of primers used in this study

| | |
|-----------------|--|
| JB883 hygR 5' | GtggacGGccggcggtttggcgccgttttcg |
| Nae | (SEQ ID NO.: 12) |
| JB911 DGA1 5' | CattcaaaGGCGCGCCatgactatcgactcacaatactaca |
| AscI | (SEQ ID NO.: 13) |
| JB912 DGA1 3' | GcGGATCCTTAATTAAattactcaatcattcggaactctgg |
| PacI | (SEQ ID NO.: 14) |
| JB913 DGA2 5' | CattcaaaGGCGGCCATGGAAGTCCGACGACGAAA |
| AscI | (SEQ ID NO.: 15) |
| JB914 DGA2 3' | GcGGATCCTTAATTAACTACTGGTTCTGCTTGAGTTGT |
| PacI | (SEQ ID NO.: 16) |
| AH011 Tup1 5' | GACTGGCGCGCATGAGCTTCCCCAACAAAGTA |
| Asc | (SEQ ID NO.: 17) |
| AH012 Tup1 3' | GTCCTTAATTAAATTATCTGTTGACAGGAAAGTATCGC |
| PacI | (SEQ ID NO.: 18) |
| AH007 HacI 5' | GACTGGCGGCATGTCTATCAAGCGAGAAGAGT |
| AscI | (SEQ ID NO.: 19) |
| AH008 HacI 3' | GTCCTTAATTAACTAGATCAGCAATAAGTCGTGCT |
| PacI | (SEQ ID NO.: 20) |
| AH020 MAE 5' | GAETGGCGGCCATGTTACGACTACGAACCATGC |
| AscI | (SEQ ID NO.: 21) |
| AH021 MAE 3' | GTCCTTAATTAACTAGTCGTAATCCGCACATG |
| PacI | (SEQ ID NO.: 22) |
| LQ310 Mga2 5' | ACTGGGCGGCC atggctaaagacaaggaaatcgactttgac |
| AscI | (SEQ ID NO.: 23) |
| LQ303 Mga2TM 3' | ACTGTTAATTAA tcagtaaatgtaaagccagaacatcg |
| PacI | (SEQ ID NO.: 24) |
| LQ309 Mga2 3' | ACTGTTAATTAA tcatgcagccgtggcctgg |
| PacI | (SEQ ID NO.: 25) |
| LQ294 O6M 5' | ACTGGGCGGCC atgtttcacccaagccccgaccgg |
| AscI | (SEQ ID NO.: 26) |
| LQ295 O6M 3' | ACTGTTAATTAA ttagagagtccccacatgtcaccc |
| PacI | (SEQ ID NO.: 27) |
| LQ259 MRM2 5' | ACTGGGCGGCC Atgcgc当地atgtccgttcaac |
| AscI | (SEQ ID NO.: 28) |
| LQ260 MRM2 3' | ACTGTTAATTAA ttatggctcccttctgccacatc |
| PacI | (SEQ ID NO.: 29) |
| LQ261 DGA1 5' | ACTGGGCGGCC Atgactatcgactcacaatactac |
| AscI | (SEQ ID NO.: 30) |
| LQ262 DGA1 3' | ACTGTTAATTAA ttactcaatcattcggaactctgg |
| PacI | (SEQ ID NO.: 31) |

TABLE 3

TABLE 3-continued

| Media formulations used for two strain testing | | | Media formulations used for two strain testing | | |
|--|-----------------------------------|--|--|-----------------------------------|--|
| Media Name | Carbon Source Glucose (g/L) | Nitrogen Source Ammonium Sulfate (g/L) | Media Name | Carbon Source Glucose (g/L) | Nitrogen Source Ammonium Sulfate (g/L) |
| C ₁₀ N ₅ | 10 | 5 | C ₄₀ N _{0.2} | 40 | 0.2 |
| C ₂₀ N _{0.04} | 20 | 0.04 | C ₄₀ N ₁ | 40 | 1 |
| C ₂₀ N _{0.2} | 20 | 0.2 | C ₄₀ N ₅ | 40 | 5 |
| C ₂₀ N ₁ | 20 | 1 | C ₈₀ N _{0.04} | 80 | 0.04 |
| C ₂₀ N ₅ (YSC) | 20 | 5 | C ₈₀ N _{0.2} | 80 | 0.2 |
| C ₂₀ N ₁₀ | 20 | 10 | C ₈₀ N ₁ | 80 | 1 |
| | | | 60 | | |
| | | | 65 | | |

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TABLE 3-continued

| Media formulations used for two strain testing | | |
|--|-----------------------------------|--|
| Media Name | Carbon Source Glucose (g/L) | Nitrogen Source Ammonium Sulfate (g/L) |
| C ₈₀ N ₅ | 80 | 5 |
| C ₈₀ N ₁₀ | 80 | 10 |
| C ₁₆₀ N _{0.2} | 160 | 0.2 |
| C ₁₆₀ N ₁ | 160 | 1 |
| C ₁₆₀ N ₅ | 160 | 5 |
| C ₃₂₀ N _{0.2} | 320 | 0.2 |
| C ₃₂₀ N ₁ | 320 | 1 |
| C ₃₂₀ N ₅ | 320 | 5 |

TABLE 4

| Media | Sample | Day 4 OD | Day 4 GFP Fluorescence |
|-----------------------|--------|----------|------------------------|
| CSM - C80N5 | A | 16.83 | 36696 |
| CSM - C80N5 | B | 16.76 | 34397 |
| CSM - C80N5 | C | 16.31 | 39166 |
| Minimal Media - C80N5 | A | 11.7 | 29365 |
| Minimal Media - C80N5 | B | 11.46 | 52520 |
| Minimal Media - C80N5 | C | 11.87 | 32427 |

TABLE 5

| Media Formulations used for 12 strain testing | | |
|---|-----------------------------------|--|
| Media Name | Carbon Source Glucose (g/L) | Nitrogen Source Ammonium Sulfate (g/L) |
| C ₂₀ N _{0.2} | 20 | 0.2 |
| C ₂₀ N ₁ | 20 | 1 |
| C ₂₀ N ₅ (YSC) | 20 | 5 |
| C ₄₀ N _{0.2} | 40 | 0.2 |
| C ₄₀ N ₁ | 40 | 1 |
| C ₄₀ N ₅ | 40 | 5 |
| C ₈₀ N _{0.2} | 80 | 0.2 |
| C ₈₀ N ₁ | 80 | 1 |
| C ₈₀ N ₅ | 80 | 5 |
| C ₈₀ N ₁₀ | 80 | 10 |
| C ₁₆₀ N _{0.2} | 160 | 0.2 |

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TABLE 5-continued

| Media Formulations used for 12 strain testing | | |
|---|-----------------------------------|--|
| Media Name | Carbon Source Glucose (g/L) | Nitrogen Source Ammonium Sulfate (g/L) |
| C ₁₆₀ N ₁ | 160 | 1 |
| C ₁₆₀ N ₅ | 160 | 5 |
| TABLE 6 | | |
| RFU and OD for EMS data | | |
| | RFU | OD |

| | | |
|-----------------------------|----------|-------|
| APEX10ΔMFE1 S2-DGA1 Control | 23750 | 8.81 |
| E1 | 31800 | 21.91 |
| E6 | 35400 | 18.86 |
| E12 | 37100 | 22.5 |
| L36 Control | 23133.33 | 11.83 |
| E1S6 4 | 34350 | 20.61 |
| E1S6 6 | 34250 | 20.58 |
| E1S6 8 | 28750 | 18.31 |

| Media | Sample | Day 4 OD | Day 4 GFP Fluorescence |
|-----------------------|--------|----------|------------------------|
| CSM - C80N5 | A | 16.83 | 36696 |
| CSM - C80N5 | B | 16.76 | 34397 |
| CSM - C80N5 | C | 16.31 | 39166 |
| Minimal Media - C80N5 | A | 11.7 | 29365 |
| Minimal Media - C80N5 | B | 11.46 | 52520 |
| Minimal Media - C80N5 | C | 11.87 | 32427 |

TABLE 7

| List of genes and genetic changes | |
|---|----------------------|
| Gene | Type of Modification |
| Leucine Biosynthesis Gene (LEU2) - Note may also be able to include rest of genes of leucine biosynthetic pathway, have yet to test these additional ones | Over-expression |
| Uracil Biosynthesis gene (URA3) multifunctional enzyme (MFE1) in b-oxidation | Over-expression |
| Deletion pathway | |
| Transcription Factor (PEX10) | Deletion |
| AMP Deaminase (AMPD) | Over-expression |
| ATP-Citrate Lyase (ACL1 and/or ACL2) | Over-expression |
| Malic Enzyme (MAE/MEA) | Over-expression |
| Acetyl-CoA Carboxylase (ACC) | Over-expression |
| acetyl-CoA:diacylglycerol acyltransferases (DGA1 and/or DGA2) | Over-expression |
| Mitochondrial 2' O-ribose methyltransferase(MRM2) | Over-expression |
| O-6-methylguanine-DNA methyltransferase (MGMT) | Over-expression |
| Aconitase (ACO1) | Deletion |
| Citrate Synthase (CIT1) | Over-expression |

TABLE 8

| Strain L36 important SNP list | | | | | |
|-------------------------------|----------|-----------|---|---|-------------------|
| | Mutation | | | | Accession numbers |
| Chromosome | Position | type | sequence | Gene | |
| B | 1644655 | SNP | C > T | mga2 | 12342g |
| D | 2401168 | Insertion | A > AG | sorbitol utilization protein SOU2 | 18964g |
| E | 1837892 | SNP | C > A | CENOE | 15444s |
| | 1837894 | SNP | T > A | CENOE | 15444s |
| | 4025540 | SNP | C > A | DEHA0A1298g IPF 95.1 | 33891g |
| | 4025542 | SNP | G > C | DEHA0A1298g IPF 95.1 | 33891g |
| F | 2861334 | Insertion | A > AGAGGG CTAGAGAG AGGGAGA A (SEQ ID NO.: 32) | RLF2 chromatin assembly complex subunit p90 | 21637g |

Gene Targets: The reference number given for each name corresponds to the Genolevures database: <http://www.genolevures.org/>. YALI0 stands for *Yarrowia lipolytica*. A.B.

C,D,E,F specifies chromosome, and the following number specifies location. Note: Leu2 and Ura3 given as GenBank Accession numbers

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aagttcaacctcaagtacaaccctgtcggtgagtcgactgcgagaaatctccataagacccgacaact
 acatccagggtcgatacctagctgagatcacaaaggagggtgtccaggatctcgagaactcgaaacttaca
 gatggcggagtgaccgtatccatcacggtcggccaaggacgcgactggacaaactggctggctgg
 ctggacaacaaactgtttcgccaaatgtcggtgttgcattcgactgtacgacattaca
 agaaggctggctggtaaacacccatgtcgacattgtcgagaacgtcttgagccctttcgaggtcac
 caaggatcccagtagccatccaaactgcacgtgtccctgcagcgagttgtggcttgactctgtcgat
 gacgagtcgaagctggaccgcacgtttccaccgaaagttccaaactgcagcatactggacagcgcacaga
 accctccctactcgtaactggcactatctatacgccaaatcgccatcaacacccatggagacagcg
 ttggctataatactttgagttcgacccatgtggagaggctggacccagagcatctctgtgc
 acttatctggcttcagggtatcaaccacccatgttgcgaaagggtgccttcattcagtagcc
 atacactggaccagatccccatgcgttgcctgttgcgaaatgcgttgccttcacgttgcacaa
 gaacccttctactcataacttcaagcgggtcaacgtgtccatgttgcgatcgttgcagtt
 gcttacactaaggaggctgttgcgatcgtggctgcgttgcgatcgttacaagcttccaaacgtgg
 atatgttgagcttgcgaaactcggtactgtggctgcgttgcgatcgttacaaggagcatggat
 cggcggaaaactacgagatccatggcccgaggcaacaccatccagaagacaaacgtgcccattgtgcgt
 ctggccttccgagacgagacttgcgatcgttgcgatcgttgcgatcgttacaaggagcatggat
 ttgagcggctgcgttgcgatggta

(SEQ ID NO.: 34)

Amino Acid =
 MPQQAMDIKGKAKSVPMPPEEDDDSHFVGPISPRPHGADEIAGYVGCEDEDDELEELGMLGRSASTHFSY
 AEERHLIEVDAKYRALHGHLPHQHSQSPVSRSSSFVRAEMNHPPPPSSHTHQQPEDDDASSTRSRRSSR
 ASGRKFNRNRTKSGSSLKGQLQQLNMTGSLEEPEYESDDDARLSAEDDIVYDATQKDTCKPISPTLKRT
 TKDDMKNMSINDVKITTTEDPLVAQELSMMFEKVQYCRDLRDKYQTDSLQDGDNPKDDKTHWKIYPEP
 PPPSWHETEKFRGSSKKEHQKDPMDEFKFEDCEIPGPNDMVFKRDPCTVYQVYEDESSLNENKPFVA
 IPSIRDYYMDLEDLIVASSDGPFAFRRLQYLEAKWNLYYLLNEYETETTESKTNPHRDFYNVRKVDT
 VHHSCAMNQKHLRLFIKYKMKNCDEVVIHRDGRELTLSQVFESNLNTAYDLSIDTLDMAHKDSFHRFD
 KFNLKYNPVGESRLREIFLKDNYIQGRYLAETKEVFQDLENSKYQMAEYRISIYGRSKDEWDKLA
 LDNKLFSPNVRWLQIVPRLYDIYKKAGLVNTFADIVQNVFEPFLFVTKDPSTHPKLHVFLQRVVGFDSVD
 DESKLDRRFHRKPTAAWDSAQNPPYSWQYYLYANMASINTWRQRLGYNTFELRPHAGEAGDPEHLLC
 TYLVAQGINHGILLRKVPFIQYLYLDQIPIAMSPVSNNALFTFDKNPFYSYFKRGLNVSLSSDPLQF
 AYTKEALIEEYSAALIYKLSNVDMCELARNSQLQSGFERIIKEHWIGENYEIHPEGNTIQKTNVPNVR
 LAFRDETLTHELALVDKYTNLEEFERLHG*

Leu2 - AF260230

(SEQ ID NO.: 35)

Nucleotide =
 atggaacctcgaaactaagaagaccaagactgactccaagaagattgttctcgccggcacttctgt
 gccccgaggttgcgaggccgtcaagggtgctcaagtcgttgtcgaggcctccggcaccgagttgt
 gtttggaggaccgactcatggaggagctgcattgagaaggaggcgagccatcaccgcacgtactctc
 gacatctgccgaaaggctgactctattatgctcggtgtcgaggcgctgccaacaccgtatggacca
 ctccccgacggaccaaccgacgtgcgaccggcagggttcctcaagtcgtcgaaaggacctgaaacctgt
 cgccaaacctcgacccctgcccagctgtcgccaaagctcgccatctcccccattcgaaacgttgc
 ggccaccgacttcatatgtccgagagctgtcgaggatctactttggagagcgaaaggaggatgacg
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cttcctggcccttcagcacaaccccccattcccgatgtggcttgcacaaggccaacgtgctggccctcc
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agctgatcgactccgcgcatgtatctcatcaaggcagccctcaagatgaatggtatcatcataccac
caacatgtttggcgatcatctccgacgaggcctccgtatcccggtctctgggtctgctgcctcc
gcctcttggttctgtccgacaccaacgaggcgttcggtctgtacgagccgtcacggatctgccc
ccgatctcgcaaggcagaaggtaacccattgccaccattctgtctgcccgtatgtcaagttctc
tcttaacatgaagccgcggtgacgctgttgaggctgcccgtcaaggagtccgtcaggctgtatca
accggcatacgaggcttccacccatccgaggctggagacttgttgcacaaggtaaggagct
gtcaagaaggagaagtcgtttctacgacgcattgtatggaaaggagcaaactgacgcgcctgcgggttgg
tctaccggcagggtccgctagtgtataa

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(SEQ ID NO.: 36)

Amino Acid =
MEPETKKTDTSKVILVLLGGDFCGPEVIAEAVKVLKSVAEASGTEFVFEDRLIGGAAIEKEGEPIIDATL
DICRKADSIMLGAVGGAANTVWTTPDGRDVRPEQGLLKLRIKDLNLYANLRPCQLLSPKLADLSPIRNVE
GTDFIIVRELVGGIYFGERKEDDGSVASDTETYSVPEVERIARMAAFLALQHNPLPVWSLDKANVLAS
SRLWRKTVTRVLKDEPPQLELNHQLIDSAMILIKQPSKMNGIIITTNMFEDIISDEASVIPGSLGLPS
ASLASLPDTNEAFGLYEPCHGSAFDLGKQKVNPATLSAAMMLKFSLNMKPAGDAVEAAVKESVEAGIT
TADIGGSSSTSEVGDLLPTRSRSKSFLRRIDGRSKLTRLRVGLPAGSASV*

Ura3 - YLU40564

(SEQ ID NO.: 37)

Nucleotide =
atgcctccatcagaagctcgagctaactccacaaggcccttgcgtcgactgtcaagctcggt
cagccaagaaaaccacccatgtgttaccaccaccaaggagcttgcgttgc
taagggtcgacccatgtgtcatcaagacccatatcgacatcattgcgttgc
actgtgtcccccataaggaaacttgcattaaacgcgttccctgtcgaggac
atattggcaacactgtcaagcaccagtacaagaacgggttgcgttgc
caacgcacccacgggttgcgttgcgttgcgttgcgttgcgttgcgttgc
gaacagaagaaggaggacgtctgtactacgagaactccatgttgcgttgc
acgagaagctggccagaggctgtcatgttgcgttgcgttgcgttgcgttgc
gtactccaaggcaggatgttgcgttgcgttgcgttgcgttgcgttgcgttgc
cgacctaaggccactgttgcgttgcgttgcgttgcgttgcgttgcgttgc
ctctcgacaggactgttgcgttgcgttgcgttgcgttgcgttgcgttgc
aggctgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
taccagaagattaactgttag

(SEQ ID NO.: 38)

Amino Acid =
MPSYEARANVHKSAFAARVLKLVAAKKTNLCASLDVTTKELIELADKVGPYVCMIKTHIDIDDFTYAG
TVLPLKELALKHGFLLFEDRKFADIGNTVKHQYKNGVYRIAESDITNAHGVPGTGIIAGLRAAGAEETVS
EQKKEDVSDYENSQYKEFLVPSPNEKLARGLLMLAELSCKGSLATGEYSKQTIELARSDPFVVGFIAQN
RPKGDSEDWLILTPGVGLDDKGDALGQQYRTVEDVMSTGTDIIIVGRGLYQONRDPIEEAKRYQKAGWEA
YQKINC*

ACLsubunit1 - YALI0E34793

(SEQ ID NO.: 39)

Nucleotide =
atgtctgccaacgagaacatctccgattcgacgcccgtgtggcaaggagcacccgcatacgactct
tccataaccacacacatcttcgtctatggtctccagcctcgagccaggatgtggacttcga

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ttcatactgtaaagcgagagaacccctccgtggccgggtgtatctatcccttcggccggccagtttgtacc
aagatgtactgggcaccaaggagactcttccctgttaccagcaggtcgagaaggcccgtgccaagc
accccgaggctcgatgtcggtcaactttgcctctcgatccgttactcttaccatggagtgct
cgagtaccccaagttccgaaccatgccattattgocgagggtgtccccgagcgcacgagcccgagatc
ctccacaaggcccagaagaagggtgtgaccatcttggccgttaccgtcgaggtatcaagcccggtt
gttcaagggtggaaacccggaggatgtatggacaacattgtcgccctcaagctctaccgaccgggtc
cggtgcctacgtctccaagtcggaggaatgttcaacgcgtgaacaacattatcttcacaccaccgac
ggtgtctacgagggtattgttatggtggtgaccgataccctgttactacatttcatgaccatattctc
gatacgaggcccaccccaagtgtaagatcatcgctcccttggtgagggtgggtgttgaggagtacc
agtcatcgaggctgttaagaacccgcacatcaagaagccatcgctcgctggccattggtaacttgtcc
tccatgttcaagactgaggttcaagtccggcacgcggccatggccaaactccgacctggagactgcca
aggctaagaacccgcacatgaagtcgtgttccatgtccccgataccctcgaggacatggccagggt
ccctggccagcttacgagaagatggtcgccaagggcgagctgtctcgatctcgaccctgagggtcccc
aagatcccttactgactacttggcccaaggagcttggttatccgaaagcccgctgtttcatctcca
ctatttccatgtgaccgaggccaggagcttctgtacgctggcatgcccattccgagggtttcaaggagg
catggtatcgccgtgtcatgtctgtgttcccgacgacgactcccgactacgccttcaaggat
cttgagatggttctcatgttactgtgaccacggtcccgctatccggccatgaacaccattatcca
ccaccccgagctggtaaggatcttccctgggtgtggtotctcgaccatggtaaccggatccg
agggtgtctgtacgggtgtccacccggagtttaccactgcctacgacaagggtctgtccccccgacagtt
gttgatcatgtcgaaaagcagaacaagctgttccctgggtatggcategagtcagttctcgaa
cccgatcccgactcgagctgtcaaggactttgttaagaagaacttccctccacccagctgtcgact
cgcccttgcgtcgaggagggtcaccaccccaagaaggacaacctgatttgcacccgttgcatt
gtgtttctttgtcgatctcatgegatcttgcgggtgccttactgtggaggagactgaggactacctca
agaacgggtttcaacggctgttgcgttccgtcgatccatggtctattggccaccatcgatca
gaagcgactcaagaccggctgttaccgacatccctgggacgatcacctacccgttggccaggaggct
atccagaqaqaadccqatcqagatcacqccqccqccqacqttccaaqgccaqactcqatcatq

(SEQ. ID. NO.: 40)

Amino Acid =

MSANENISRFDAPVGKEHPAYELFHNHTRSFVYGLQPRACQGMLDFDFICKRENPSVAGVIYPFGQFVT

KMYWGTKEPLLKVYQQVEKAACKHPEVDVVNFASSRSVSYSTMELLEYPQFRTRIAIIAEGVPERRAREI
LHKAKQKGVTIIGPATVGGIKPGCFKVGNTGGMMDNIVASKLYRPGSVAYVSKGGMSENLLNIISHTTD
GVYEGIAIGGDYRPGTTFIDHILRYEADPKCKIIVLLGEVGGVEEYRVIEAVKNGQIKKPIVAWAIGTC
SMFKTEVQFGHAGSMANSDETAKNAAMKSAGFYVPDTFEDMPPEVLAELYEKMVAKGELSRISEPEVP
KIPIDYSWAQELGLIRKPAAFIISTISDDRGQELLYAGMPISEVFKEDIGIGGVMSLLWPRRLPDYASKF
LEMVLMLTADHGPAVSGAMNTIITTRAGKDLISSLVAGLLTIGTRFGGALDGAATEFTTAYDKGLSPRF
VDTMRKQNKLIPGIGHRVKSRRNPDRVELVKDFVKKNFPSTQLDYALAVEEVTSKKDNLILNVDGAI
AVSFVDSLRSGAGTVEETEDYLKNGVLNLGFVLGRSIGLIAHHLDQKRLKTGLYRHPWDITTYLVQGEA

ACI-unknown-2 UNITS OF

(SEQ ID NO.: 41)

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ggacggcgtggcccccgagcagatttcgccgcgctgaaaagacctaaccctggctgctggagtccggc
gccaagggttggccaagcccgaccagctcatcaagcgcacgaggcaaggccggctgctggtaactcaaca
agtctgggaggagtgcagaagccctggatcgccgacggggcccaagccatcaacgtggagggttgcattga
cgaggatgcgacgttcctggcggcccttgcggccacgaccagaacgacgactatcaac
atccactccgtgcgagagggcactggatecttttacccacgaggaggatgcacgtggcgacgtgg
acgccaaggccccaagatcctcatccccgttgcattgagaacgacttacccctcaacgcacgctcac
caaggagctgtggcacacgtgcccggaggaccagcaccagaccctgtcgacttcatcaacggcttac
gcgtctacgtogatctgcagttacgtatctggatcaacccctgtgtatccccacccggcagg
ggcgcgaggteactacctggatcttgcggcaagctcgaccagaccgcagatgggtgatgcggcccaa
gtgggctgtgcgggtccccccggctctggccaggtegtcaettgcacgcggctccaccaagggt
tcacategacgcggccccccatggcttcccgctcttcggatgcagatgttcaatgcaaggaggcgt
acattgcggagctcgattcaagacggagctctgtgaagctgacttcaatgcaaggaggcgaat
ctggacccttggctggatggaggacccgtcgatctacgcgcacgcattgcgtctgcggcttgc
gacgagctgcacaactacggcggactctggcgctccaaacgagaccgcacgtacgcgggg
ccgtactggatctcatgacccgggggacgcctcaccggcaggatgcggacttgcgggg
cgccaaacttcacccagggttatccacccatggatccacccatccggggctccggactaccagg
ctgcacaaccacaagggttatcgtgcacgaggcggtccaaactggcaggagggttgcgggt
tcaagtgccgtgcacgagctgaatctggccatggatattacggccggacatgcacgtgtcggtat
tgttcccttggctctgtggaaagcgcccaagaatgtcaaggctttggcacggacccatctactgag
gttccactcttcggagttaa

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(SEQ ID NO.: 42)

Amino Acid =
MSAKSIHEADGKALLAHFLSKAPVVAEQQPINTFEMGTPKLASLTfedGVapeqifAAAEKTypwllesg
AKFVAKPDQLIKRKGAGLLVLINKSWECKPWIAERAAPINVEGIDGVLRFLVEPFVPHDQKHEYyin
IHSVREGDWILFYHEGGVDVGDVDAAKAIIIPVDIENEYPSNATLTKELLAHVPEQHQTLDFINRLy
AVYVDLQFTYLEINPLVVIPTAQGVHDLAGKLDQTAEEFECGPWAAARSPAALGQVVTIDAGSTKV
SIDAGPAMVPAPFGRELSKEEAYIAELDSKTGASLKLTVLNAGRIWTLVAGGGASVYADAIASAGFA
DELANYGEYSAPNETQTYEAKTVLDLMTRGDAHPEGKVLFIGGGIANFTQVGSTFKGIIRAFRDYQSS
LHNHKVKIYVRGGPNWEGGLRLIKSAGDELNLPMEIYGPDMHVSGIVPLALLGKRPKNVKFGTGPSTE

ASTPLGV*

MEA1 - YALI0E18634

(note: 4 nucleotide difference compared to the reference sequence.
In embodiments, MEA1 is the reference sequence associated with
YALI0E18634. In embodiments, MEA1 is the reference sequence with
the four nucleotide differences from the reference sequence
shown below.)

(SEQ ID NO.: 43)

Nucleotide =
atgttacgactacgaaaccatgcgacccacacagaccaggcgtcaggccggcgcttggccacgcgtcgccg
cccgaaacatgtccctccagccctccagcttcgaatactcgtccatcgtaaggccacgcggggaaat
cgccaccgaaaggcgcccacaacccgtctgtcggttggggcccatctacgtggcttcgacggcatt
cgcttcataacctgcgcatactcaacaagggtctggattcccccataacgagcgcacgggattcggac
tcagtggcttcgtccctgtccgaaggccaccctggaggaacaggctcgaccgagcatccaattcaa
aaagtgtggactcccttagccaaaacgggttctgcacccgtcaagttccaaacgaggtgtctac
tacgcctgtgtcaagcacttgcacccatcatatacaccgactcaggagaagcca

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ttgaacagtactcgccgttccggcgccgaggcttcgtcgacatcaccagtcacgcgt
ggaggagcgtctggagcgttggagaccatgacgacattgactacattgtcgactgactccgagggt
attctcgaaattggagccaaggagtggcggtattgttccatcgcaagctggctctcatgactc
tatgtcggtcgactcaaccctcacgacttccgtgttctggatacggaaaccaacaaccaggagct
gtgcacgaccccctgtatctggccgacgaatgccccgagtgcgaggaaagcagtacgacgacttcac
gacaactttgtcgacttgcggcgaggctgtatccaaaggcggtatccattcgaggacttggctcg
ctaacgcacacaagatcctcgacaagttacgacccggagatccctgcttcaacgacgacatccaggcac
tggagccgtactctggccatcacggcgctcaagggtctggcaaaaatatacagatactcg
attctcggtacggagctggcgatggatttgcgaaacaggctatgataacctgggttgc
agggtctcgacgacaagactgcgcacaaaacatcttcatggaccgaccggctactgaccaccgc
acttaccgacgacatgacgtgcagaagccgtttgcacaggcaattacgaggagtg
gacccaagactctggagcacgtgggtctgcgtcaagcccatattctattggatgttccactcagc
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ttccaacccacacgtttcatgaggctgtccctgcagatctgtacaagtggaccgacggcaaggctctg
gttgcacccggctcgcccttgaccctgtctcgatcaaagctcatcacaacaccatgattgtctgc
tccccggatcggctggagccattctgtctcgatcaaagctcatcacaacaccatgattgtctgc
catcgagtgcctcgccgaacaggccccattctcaagaaccacgacgaggagacttccgacgtagct
ctcatccagatcattcggccgggtggccactgcccgttctcaggccaaaggctgagggctagcca
ctgtcgaggaagagactcaagccggaccaagaaacatgtgcagattccgacaaactttgacgagtg
cgcttgggtcgagactcagatgtggccggccgttacccgcctctcatccatgtgcggattacgactag

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(SEQ ID NO.: 44)

Amino Acid =
MLRLRTMRPTQTSVRAALGPTAAARNMSSSPSSFEYSSYVKGTREIGHRKAPTRLSVEGPIYVGF
RLLNLPHLNKGSGFPLNERREFGLSGLPSAETLEEQVDRAYQQFKKCGTPLAKNGFCTSLKFQNEVLY
YALLLKHVKEVFPPIIYTPTQGEAIEQYSRLFRRPECCFLDITSPYDVEERLGAFGDHDDIDYIVVT
ILGIGDQGVGGIGISIAKLALMTLCAGVNPSRVIPVVLDGTNNQELLHDPLYLGRMPVRGKQYDDFI
DNFVQSARRLYPKAVIHFDGLANAHKILDKYRPEIPCFNDDIQGTGAVTLASITAALKVLGKNITDTR
ILVYGAGSAGMGIAEQVYDNLVAQGLDDKTARQNIFLMDRPGLLTALTDEQMSDVQKPF
DKTLEHVVAAVKPHILIGCSTQPGAFNEKVKEMLKHTPRPIILPLSNPTRLHEAVPADLYKWT
VATGSPFDPVNGKETSENNNCFVFPGIGLGAIRSKLITNTMIAAAIECLAEQAPI
LQIIISARVATAVVLQAKAEGLATVEEELKPGTKEHVQIPDNFDECLAWETQMWRP
DVA LIQIIISARVATAVVLQAKAEGLATVEEELKPGTKEHVQIPDNFDECLAWETQMWRP
VYRPLIHVRYD*

DGA1 - YALIOE32769

(SEQ ID NO.: 45)

Nucleotide =
atgactatcgactcacaatactacaaggcgagacaaaaacgcacccgcacccaaatcgccggaaatcc
gatatggcccgctatcgacaccattactcaaccgtatgtcgacccctctctggcttgcacatttcag
cattccactttccatcaaaatttcatgtatgtcgcaattccactgtctggccatttgtgattgc
tatgtatgtacgtgttacggactcccgatccacggaggatgttgcgatactcgccatatt
caagaaaccttcatgttgcgatcttgcgacttccgcataactctgcacaagacggatct
ggagccacgcacacatactaccctctggacgtccaggatgtatccatgttgcgatactggcc
cagaacaaggatccgcgacatcatccacccatcgatcttgcgccttcatgaaacggatct
tttctatcaacgagcaggaggcgcgtccgcgacatctctctgtctccgttctccagctctcc
gggttctcaacctgacaaggatggattaaccacgcacgcacatagccgtggagaatcatctgg
gtccac

116

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ggccacgcctcggtccggaaacttaacggcaacggcaacaatggcaccactaaccgacgacccatgt
ccgcctctgctggctccactgcacatctgattccacgcttcttaacgggtccctcaactctacgccaacca
gtcattggcggaaaacgaccacacagctgtgcggcccaaaaaactcaagggccacttggcagaaaatacatctt
ggctaccacccccacggcattatcgcatgggagcccttgggtggaaattggccaccggaggagctggatggt
ccaagctttccggcatccctgtttcttatgacttcaccaacaacttccgagtgcctctcacag
agagtacctcatgagttctggagtcgttctgtctccaagaagtcctgcacggcccttcataagcggaaac
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tgctactcaagcggaaagggttggacttgcacttggatggaggtcgaaatgtcgccctgttcccacat
ggccttggtgagaacgaccctatgaccaggtagcaacgacaagtcgtccaaagctgtaccgattccg
cagtttgtcaagaacttcctggattcacccctttgatgcacatgcccggaggcgtttcaactacgatg
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acacccacccacgcgaagaagtgtccgaataccacgaccgatacatcgccgagtcgcagcgaatctacaac
gagcacaaggatgaatattcatcgatggaccgaggaggcaaggagcccccaggttcggatgatt
agtaa

(SEQ. TD. NO.: 46)

SEQ ID NO.:
Amino Acid = MTIDSQYYKSRDKNDTAPKIAIGIRYAPLSTPLLNRCETFSLVWHIFSIPFLTIFMLCCAPIPLLWPFVIA
YVVYAVKDDSPSNGGVVKRYSPISRNFIFIWKLFGRYFPITLHKTVDLEPTHTYYPLDVQEYHLIAERYWP
QNKYLRAlISTIEYFLPAFMKRSLSINEQECPAERDPLLSPVSPPSGSPQDKWINHDSRYSRGESSGSN
GHASGSELNGNGNNNTNRRPLSSASAGSTASDSTLLNGSLNSYANQIIGENDPQLSPTKLKPTGRKYIF
GYPHGIIIGMGAFFGIATEGAGWSKLFPGIPVSLMTLTNNPRVPLYREYLMMSGVGAVSKKSCKALLKRN
QSICIVVGGAQESLLARPGVMDLVLLRKKGFGVRLGMEVGNVALVPIMAPGENDLYDQVSNDKSSKLYRFQ
QFVKNFLGFTLPLMHARGFVNVDGLVPYRRPVNIVVGSPIDLPYLPHTDEEVSEYHDRYIAELQRINY
EHKDEYFIDWTEEGKGAAPEFRIE*

DGA2 - YALI0D07986

(SEQ ID NO.: 47)

Nucleotide =
atggaaagtccgcacgacaaaaatcgacgtgtcaaggcccagaaaaacggctacgaatcgggcccacat
ctcgacaatcgtcgcagccctcctcaagagcatcgccagaaccggcaacaaacactcctcgccaccct
gtcgctcagcgactgaccatgaaagtccagaagaacacctcgccggaccggcggactccaaacgcga
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accccgactcaagggttcaaaaacatccggatgtatccatgttgcggaaatctacggctcgatt
cgaaaactacctaatacggcattccaaccggtttgcacccaaaattactcctccgagtggcag
ctctcaggcttgctcatagtcgtggcctacgcacatatcctcatggcctacgctattgagagcgtgcca
agctgctgttctctctagcaaacaccactacatggccgtggggcttctgcataccatgacacttggc
gtccatctcggtgtgtccatcgctgtctactactacacctgcccacccggcaggcacaatagtcgag
tttgtggccgttattctgttctctcaaaactcgccctacgcacccactaactcgatctccgaaaaggcc
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tttcagatgacgcagagacttggcagacatttgacgtcattctgcataactacgcacagctggccctaccc
cagaatgtgacgctgtgaacctgtgtacttctggttgtccccactggctaccaggcccgtgtacc
ccaaagacggagcgtattcgacccaaggcacgtgatccgaaacctgtttagctgctctctgtgcgtact
tattcagtttctcatttccagtgacgcctacccatcatgcagtgctggctgttctccagccc
aaqctcgattatqccaaacatctccqaqcqccatqaaqtqqccctcqqtctatqatqqtctqqqctca

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ttggattctacgcttccagaacggctcaatcttattgccagactcacctgtttggaaacagaac
cttctaccaggcagtggaaattcccgctccatggccagactggactctatgaaacaagcagtcac
cagttacttttagacaccacgttacgtgcctctctcgctcgcccgtatgcgtcggtgg
tggttttttttcgcgtatccatgaactgttgcgtccatccccactcacaacatcatcgagc
cgcccttctcgcatgtatgtcgaggtgcctctgtatggctactgagaacccatcgatattaactcc
tctctggggcccttcgttgcactgtgcattctggttcaccttttctggacaacccacttgtcat
tccttattatctggcttacaactacaacgcagaaccaggtag

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(SEQ ID NO.: 48)

Amino Acid =
MEVRRRKIDVLKAQKNGYESGPPSRQSSQPSSRASSRTRNKHSSSTLSGLTMKVQKKPAGPPANSKTP
FLHIKPVHTCCSTSMLSDYDGSNPSPKGFKNIGMIILIVGNLRLAFENYLKYGISNPFPDPKITPSEWQ
LSGLLIVVAYAHILMAYAIESAKLLFLSSKHHYMAVGLLHMTLSSISLLSYVYYYYLPNPVAGTIVE
FVAVILSLKLASYALTNSDLRAAIHAQKLDKTQDDNEKESTSSSSSSDAETLADIDVIPAYYAQLPYP
QNVTLSNLLYFWFAPTLVYQPVYPKTERIRPKHVIRNLFELVSLCMLIQFLIFQYAYPIMQSCLALFFQP
KLDYANISERLMKLASVSMMVWLGFYAFFQNGLNLIAEELTCFGNRTFYQQWNNSRSIGQYWTLNKPVN
QYFRHHVYVPLLARGMSRFNASVVVFFSAVIHELLVGIPTHNIIGAAFFGMMSQVPLIMATENLQHINS
SLGPFLGNCAFWFTFFLGQPTCAFLYYLAYNYKQNQ*

MGA2- YALIOB12342

(SEQ ID NO.: 49)

Nucleotide =
atggctaaagacaaggaaatcgactttgactacacgggagaactggatggacgattcgagttccca
tcgacgacatgtccacaacgcacggagatgactttgtcaagaaggaaacgtggacgagggtttggtt
cggaacaaatggcgccgtgggtgcgcagatggacgtccagaccagccattagegaccctgttttgc
ggcgtggagcaggccctgacatgtgggtctcatggatacaaacatgaaccacatcaacggtagtcaca
acatgaacagegtcgtcaagcaggagactactacacccgtccatggcactccatgaaccccaaca
gcaacagtccatgaccctcaacagcagcatcacatgaaccacaaccgcctctcagtcacatcttg
catcaacagtcacaggctcaaccacacgcaccaacaacagccacatcagtcacaggagtcata
gcataatcacaaaggcatacaccaggcagcaggagacctaccgtacggacgaaagtactcacgacaact
caacaagtaccccgaggacgtggagattcatcttcgaccatcgctatggacatgtgtgaccaac
tcggaaactccgtaccaataccagatacatgtccattccatgcccggaaatcacgtgtggagacccaaa
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ggcgtccaaatcccgactcaagcaggccacatcccgactcgtgtctccatggaaatcacgtgtggagacccaaa
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gtcgaaagaaactcttgcgagtcggaggagctgtcggtcgagactcgtcaacgcacgtctggctgt
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acagttaccgcacaggcgtggctctggccctgcgtctggctactgttagacaccacccggggaaaa
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cctgccaccgcgtggcttccatgcacccccccacccagggttcctaccccgctgcatttcgtcgacgagct
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ccatttcattactccaactatggttctaaacgacgacgacggcttccatcagcgttggagcggc
atgatgaacgtgcgaggcatggatagacaggcttccattaccagcattccggaaatgggtgtggcatgt
cgaacatgactgtggccagtgttccggtagcgcactaatctggctgcataacaacatgaacaacccgc

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agacgaaaacctgccgtcatcaagcgaatcatccctcgagggttccattcgaggcggcattgaagta
 accctgttgatctggttcaagtccaatctggggctgtttcggtgacaacaaggccgtggcaccc
 actgtggtctgattcgaccatcgatcgaccatctgcgcctcgaccatcggttccgtggtgc
 ttcgaagggtttgtgtcgacaagcctcagatttacatatttgcacacagacagccagttgatt
 gagttggcgctccagggttgtgggtctcaagatgaacggacggctgaaagacgcccacatgc
 gaatcggtggcaacaatggaggcggtgcgggcacaaggccatggcaggcggaaacatgtcta
 agacgttggaaagtgtgtgcagacagttcggttcaaccgtatgcctccacagaccacgaa
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 ggctcgaggagtcgtgtggatgttccgacaatggtggattcactccttcatttcgtctcttgc
 ggccgtcgaaaagattgccaagaactacttcgtgtcaacgctgacccttacaaacgtaacc
 gaaattggcg
 aaaccgtttgtgttgcgttgcctcacattctcgatcttgcgttgcgttgcctcaggcatgc
 ctgcgttgcgtataactccgattaccatcgtcagcgtcgatcttcatttcactctggcttgc
 attgcattccatccaggattcgtgttagtacggtttatgaccatggaaatgattccaacct
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 cggtaatttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 gagctactgtgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 ggaccatccccatcacttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 gccggcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 agggccatccccatcacttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 tggccatgggggtgtccacgcataagggtgttgcgttgcgttgcgttgcgttgc
 gggcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 tagccccatcacttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 cgatcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 ttgggtgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 gagcagttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 atttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 attcatcaagccccaggccccaggctgc

(SEQ ID NO.: 50)

Amino Acid =
 MAKDKEIDFDYTGELVMDDFEFPIDDMHLHNDGDDFVKKETWDEGFVGTNGAVGAQMDVQTSPFSDPVFG
 GVAGAGPDMGLMDTNMNHINGSHNMNSVVVKQEDYYTPTSMGTPMNPQQQQSMTPQQQHHMNHNQPSQLQSL
 HQQSQKAQPQQQQQPHQSTGVDSIITKAYTRAAGDLPYGRKYSRQLNKYPEDVEYSSPDPSLWSNLLTN
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 GEQNPSKPVNLCSRCIKREQKRACRKLFDESEELSWVETQRRLAVFNCEVLEFKDVERRVYIPESGT
 TVTAQQLVLPLRLACYCRHHGEKKGFRILFCLRDEGGQIVVGVGQSGTTVMITDDHKVVGDAVAMPTTATA
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TLLGSGFKSNLVAVFGDNKAVGTHCWSDSTIVTHLPPSTIVGPVVVFEGFVLDKPQIFTYFDDTDGQLI
 ELALQVVGKMNMRLEDARNIAMRIVGNNGGVAGAQGAMAGGNMSNGDVGMESAAADSSVQPVSPTDHE
 DVVLRCALTDIPGGRIANWQLTNAEGQTMVHLASILGYSRVLVALVARGARVDVSDNGFTPPLHFAALF
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 SVRKHAKAKSVESPLSEEEERLVRHIEAEDQAVERVAAGIVSSNVPDVVSSNDSDHVRSDTSTENKSFS
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 AGRGRSHSSISRMRWRYLKNSSADEATRSRSRDANGAGAPPAYEEIFPGHGVVHDKKVQMAAASAENSS
 GPVGASSSAVASTSAAAAPPSPPLAPIVEDEEOLVEAWRRQRSMANDRMLFAFWLPVLLMAIGYMIKA
 FGLFPDQVSAVESVAETVGVCRGAVAKLWFQOYPVHRGQPLKDTCSFEPNSLVESALRQMNGWDREV
 IHQAQAA*

Mga2-L36-mutant version

(SEQ ID NO.: 51)

Nucleotide =
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(SEQ ID NO.: 52)

Amino Acid =
MAKDKEIDFYTGELVMDDFEFPIDDMHLHDGDDFVKKETWDEGFVGTNGAVGAQMDVQTSPFDPVFG
GVGAGPDMGLMDTNMNHINGSHNMSVVVKQEDYYTPSMGTPMNPQQQSMTPQQQHHMNHNQPSQLQSL
HQQSQKAQPQQQQQPHQSTGVDSIITKAYTRAAGDLPYGRKYSRQLNKYPEDVEYSSFDPSLWSNLLTN
SETPYQYQIHVHSMPGKSRVETQIKCALSIYPPPPQQSVRLPTDTISRPKFQLQGHIPDSCLSLEVYIV
GEQNPSKPVNLCSRCIKREQKRAKRKKLFDSEEEELSWVETQRRLAVFCSEVLEFKDVERRYIPESGT
TVTAKQLVLPLRLACYCRHHGEKGFRILFCLRDDEGGQIVVGQSGTTVMITDDHKVVGDAVAMPTTATA
PATAGSSQPPTQVPTPAASSSTSYPNRNSLPLSPTSMEDSSSEFTSDHSHYSNYGSKRRDGSSISDWG
MMNVRGMDRQASITSIPEMVGGMSNMTVASASGSATNLAAHMNNPADENLPVIKRIIPSQGSIRGGIEV
TLLGSGFKSNLVAVFGDNKAVGTHCWSDESTIVTHLPPSTIVGPVVVSFEGFVLDKPQIFTYFDDTDGQLI
ELALQVVGKMNRRLEDARNIAMRIVGNNGGVAGAQGAMAGGNMSNDVGMESAADSSVQPSPPTDHE
DVVLRCALTDIPGGRIANWQLTNAEGQTMVHLASILGYSRVVALVARGARVDVSDNGGFTPLHFAALF
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IASIQDSREYGFYDHGMISNLSHIPSTCSIRSSTSQFDAEDEWDERDEEDGDFDDSDEDSDDDSDALFM
SVRKHAKAKSVEPLSEEEERLVRHIEAEDQAVEARVAAGIVSSNVPDVSSNDHVRSDTSTENKSFS
RYFDRTLSMASWDDVLAYIYRPKRATVPNKRSGAPPSPRSTRSPSLDHPITSSGDESRTISAHAPSGG
AGRGRSHSSISRWMWRYLKNSSADEATRSRSRDANGAGAPPAYEEIFPGHGVVHDKKVVQMAASAAENS
GPVGASSSAVASTSAAAAPPSPPLAPIVEDEEQLVEAWRRQRMSANDRMLFAFWLPVLLMAIGYMVIKA
PGLFPDQVSAVESVAETVGVHCRGAVAKLWPKQYPVHRGQPLKDTCSFEPNSLVESALRQMNGWSREV
IHQAQQAA*
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Mga2-truncated version removing of transmembrane span.

(SEQ ID NO.: 53)

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Nucleotide =
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(SEQ ID NO.: 54)

Amino Acid =
MAKDKEIDFDYTGEVLMDDFEFPIDDMLHNDGDDPVKETWDEGFVFGTNGAVGAQMDVQTSPFSDPVG

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HQSQSKAQPOQQQQQPHQSTGVDSIITKAYTRAAGDLPYGRKYSRQLNKYPEDVEYSSFPDSLWSNLLTN

SETPYQYQIHVSMPGKSRVETQIKCALSIYPPPPQSVRLPTDTISRPKFQLKQGHIPDSCSLSLEVYIV

GEQNPSKPVNLCSRCIKREQKRACRKLFDESEELSWVETQRRLAVFNCEVULFKDVERRYVIPESGT

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ELALQVVGKLMNMRLEDARNIAMRIVGNNGVAGAQGAMAGGNMSNDVGMESAADSSVQPSPPTDHE

DVVLRLCLALTDIPGGRIANWQLTNAEGQTMVHLASILGYSRVLVALVARGARVDVSDNGGFTPLHFAALF

GRRKIAKKLLRCNADPYKRNRIGETVFDVACPHILDLLVGPGQGMPMAVQTSYTPDYHRQRSSSSSTLAS

IASIQDSDREYGFYDHGMSINLSHIPSTCSIRSSTSQFDAEDEWDERDEEDGFD-DDSDEDSDDDSDALFM

SVRKHAKAKSVESPLSEEEERLVRHIEAEDQAVEARVAAGIVSSNVPDVSSNDSDHVRSDTSTENKSFS

RYFDRTLSMASWDDVLAYI*

Sou2L36 YALI0D18964g

(SEQ ID NO.: 55)

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Nucleotide =
Atgtctggacacctccaccctcgccacggactgcacccttccccacagagacccaaagtccccacca
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129

130

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CEN0EL36 YALI0D15444s

(SEQ ID NO.: 56)

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Nucleotide =
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DEHA0A1298q IPF 95.1 YAL10E33891q

(SEQ ID NO.: 57)

RLF2 chromatin assembly complex subunit p90 YALI0F21637g

(SEQ ID NO.: 58)

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gccaccattcagcactatgtgagcgacagggtcctaagagcgacaagcggtgggtctgaaggatatct
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ag

TUP1 - YALI0A14542

(SEQ ID NO.: 59)

Nucleotide

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(SEO ID NO. : 60)

Amino Acid

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GVCVKTLGKDFVLSVASTLDGQWILSGSKDRGVQFWDPRTGQVQLMLQGHRNSVISVAPSPMGLFAT

GSGDCKARIWRYFPVNR*

HAC1 - YALI0B12716

(SEQ ID NO.: 61)

Nucleotide =

atgtcttatcaagcgagaagagtcccttactcccaccccccggaggacctggatctccccctgacagctgatt
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(SEQ ID NO.: 62)

Amino Acid =

MSIKREESFTPTPEDLGSPLTADSPGPESGDKRKKDLTLPLPAGALPPRKRAKTENEKEQRRIERIMRN
RQAAHASREKKRRHLEDLEKKCSELSENNDLHHQVTESKTNMHLMEQHYSVLAKLQQLSLVNMAKSS
GALAGVDVPDMSDVSMAPKLEMPTAAPSQPMGLASAPTLFNHDNETVVPDSPIVKTEEVNSTNLLHTES
SSPPPELAESTGSGSPSSTLSCDETDLVDRARHPAVMTVATTDQQRRHKISFSSRTSPLTSLDCMDCRM
TSPCLKTTSLLPSTTLLI*

MRM2 - YALI0E31933

(SEQ ID NO.: 63)

Nucleotide =

Atgcgcacaaagctgcgttcaacccgcctccagtcgttctcccgcaatctttgtgcggggaaaaaac
acgatgcgcgcagccgcgtggaaatgcgcagatgaaagacaagcatgtggccatggccaaaggctgcgg
atcccggtctcgagccgcgtacaagactacaggaaactcgactccatgttccggctgttcaagccggcatg
acgggtggatgtggatcttccttcatttcctccaggatgtccagcatccaggaaattt
cctgtccaaagaaacacaaaacgactcaaacgtgtgcggccgttcggcatggagttcccaaggac
aaggactctggatggccataggcactgcctccgtttatctggacactgaacgcgagctggcagta
ttAACAGCAACAGCAACGAACCCCAATTGGCGACGACTACCCGGTAGATAAGTGCTTAGTGACATGT
cgaaacgttaccccgaggaaacacggattttcaaagaactattaatgaccatactataggatggcaat
gttccggcatagctgtgagggaccatgcgcagtattgtgagtgaaaggaaaggcgttggatgt
gtgcagccagctcgatgtggcagaaggcataa

(SEQ ID NO.: 64)

Amino Acid =

MRQKLPLFNPLQSLLPRIFVRGKHKDARSWRWEMRQMVKDKHVAMAKADGFRSRAAYKLQELDSMFRLFPGM
TVVDSLGFAPGAWSQVAQRVRPGGRVIGVDILPCIPPPGVSSIQGNFLSKETQNELKRVLAWSAMGVPKD
KDSGGAIGTAPPSSYLDTERELGSINSNSNEPQFGDDYPVDIVLSDMCETLPQEHGFFQRTINDPYYRMAN
VSGIAVRDHAAStVSEGRKRIGCGAASFVVAEGKP*

06M- YALI0C10010p

135

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136

(SEQ ID NO.: 65)

(SEQ ID NO.: 66)

(SEQ ID NO.:
Amino Acid =
MFYTKPDPVVDYSRLKDMMDMPEYDNGQNMGFSNMNMTDLYDGGLNMSSMAQPVALNQMGSMGPMGSLSN
MPMGFVSONQPQTQAAQQAQSQNQNQNQNQNQNQPQNHNTVMSDNHNHTHNNTHNTNVTHNTPSMGGH
TTSVGGHDINDSAHVGGAHSANVTSPTPATPASTSSVPATSPQIPTFTVAPPAPSGKYVTDDERWQALVDRD
PEADGAFIYCVTSTKVCRTPTCSARLALRSNIVYFDTMKEAVAAGYRPCRRCNPDVSEMNSQRRAVGSC
NLIHSLEPDVKPRVKLAEVGLTLWHFHRLFKRYTGLTPRQYITEFHKRKLRLGPLQLQSKVVTKKSYE
RQQRRQGSNGSTPQQSPQVGASSPAGEVEAIKLETPTVETVQPLYYDSNGVTHNAANVGAHSSNVTHNTSH
VGSNATSATSSIATPLSNTTSPDTSPAQDSAYIIAHGSNASNAAPVVAPGPATGSGDNWIKTEPSMDFM
PRYEPRYDOSISIDAPMFIPDGNEYHHNGEMLGDMWGLT*

CIT1 - YALI0E02684

(SEQ. ID. NO.: 67)

Nucleotide =
atgatttctgttatccgtcccgccgttcgatcttccgttgcgtttggccatggccaacaccgccttc
gggcctacttacccaggatgtgagtatTTTtttcatcaattggttgtgtgcacggattcgt
tgtcgatcgccgttgcacacgccttaggccccatTTTcgacctgttcttgccctggaaagttttcc
gaatgcatgtgacacgcgtcgaaatgtgggtgttcaagcagcagcagcagcataaaaatatggaatgtgttgc
gtgcagaagtgcacattacataaccccgccgcaaccatacgagatggcagtataacaatgtcaatttag
caatacaaaaccacactqcaacccactaaaaaaaqaaaacacqactaacaataqqqtcttaaqqqaqcqattcq

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cgtcgaggtgeccggaggaaccaacaacaactacgccaacgtcgagctgattgtcgacgtggctgag
 cgattcggcgtcgatgcgcgtgtggccggatggggccatgccagtaaaatccccgtcccccgagtcgc
 tagccggcctcccccgaagatgttcatccgcctccggagctgccatgagatctctggagacaa
 aatttcttctaccattgtggccagcacgcaaaggcccgatgtatccctggcttggaaaccggagtgac
 gaggttgggttggacaagagcaccaacctcgatccgtgtccggagggatgtacaccaaggctgcacca
 ccggcccaaggcagggtctggagaaggctaaggatggatcccgatgtccggatgtcaaggctccgagg
 aggaggaggaaagggtatcgaaagggttgcgcagaggaggactcgaggctgttaccaccaggctcgag
 ggagagatccccggctcgccatcttattatgcgcgttgcaggcaatccccggatttggaggtgcagc
 ttctggctgatcagtacggcaacaatatttactgtttggctgagatgttccggatcgcacggcatca
 aaagattattgaggaggctctgtgactgtggctggccagcagacccactgcattatggaaaggctgca
 gtgcactcggttaagttgtcgatgtctgcaggatccgttgcaggatggatctgttattccatgaggac
 acaagttctacttcttggagctgaatccctcgatcttgcaggatggatccatccaccaccatggatcc
 tgcacccctggctggccatggatccatggatccatccatggatccatggatccatggatccatgg
 ctctttacgggttaaccctcacaccaccactccaaattgttgcacttctccggcgaggatgtgata
 agacacagcgacgtccgtcccgaggttgcacaccactgttgcgaatcacatcccgaggaccctggaga
 gggttcaagccctccggaggtactatgcacgagctcaacttccgatccgtccaaacgtgtgggttac
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 gtgagaaccgaagtgcgtctcgaaaggcatgggttgcaggactatcttgcaggatccatccatgg
 cccaaccaccgtcgagtatccatcaagctgttgcaggacaccggacttcgaggacaacaccatcacc
 ggctggctggatgagttatccatcaagctgtactgtccgcggacccactgttccctcgatgttgc
 gtgggtgtgtccatccaaggccatcgagttccgaggacttgcacccatggatccatggatccatgg
 gggccaggtccctgtcgagacattctcaagaccctttccctgttgcattcatctacgaggccagcg
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 ttggagtttagacttgcgttgcagggttgcgttgcaggacttgcgttgcacccatcgatgttact
 gaaggaggagggtggagccacgcgttgcgttgcaggacttgcgttgcacccatcgatgttgc
 cccactcagttcgatccatccatccatccatccatccatccatccatccatccatccatccatcc
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 gatgtatccatccatccatccatccatccatccatccatccatccatccatccatccatccatcc
 tcagcgtaacaaggctcatcagcgatacgagactgcgcagaacgtgtccatccatccatccatcc
 cgataaccagggtgtatgcaggacttccacttgcaggatgttgcgttgcacccatccatccatcc
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 ctggatccatccatccatccatccatccatccatccatccatccatccatccatccatccatccatcc
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 ccgagattcttcgaaagggtgtatggccagccatccatccatccatccatccatccatccatccatcc
 gtgtggcccttcgtatgcataaggtggccgaccaggctggcaccgcacttgcgttgcgttgc
 acgttgtcaaaagtacttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgcgttgc
 atctctgaaagcccgagagatttcatccatccatccatccatccatccatccatccatccatccatcc
 gagcacatttcgtcgatccatccatccatccatccatccatccatccatccatccatccatccatcc

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tgctctggttgactacaaggcagccatcatggtgtacatcccccacccggtagctgcgagggtggttct
tgggttggttgacccaccatcaactcgacatgatggagatgtacgctgacgtcgagtctcgagggtg
gtgtgtggccgaggaaatggcggtcgatcaagttaccgacgacaaagactggacaccatggctcg
tctggatcccagactctctctcaagaaggcagcttggaggatctccgatctgaggagtcaggtc
aagctcagcgtgcgagagaagtctctcatgccatctaccagcagatctccgtcagttgcgacttgc
atgaccgagctggccgaatggaggccaagggtgtcattcgtgaggcttggtaaggatgtcgte
attcttcttctggcgaatccgacgacgattagtcgaggagttacctattaccaagatcaatagcattctg
ccctcttgcactcggcttgagttcggctcgaatcaagtcgtggaaaggcttgcacttgc
ctgaccgggggtgttgcgagtggttgacgagaactctgtatgcgtctctgcactcagcagctcaa
gaaggacgcttgcggccagtcgtttgttcaactgagaaaggacccacagggtacttccaggcatg
aaggcaggctctcgcttctcttctgaggctgagcggctgagctgctcaagggttgc

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(SEQ ID NO.: 70)

Amino Acid =
MRLQLRTLTRRFMSMASGSSTPDVAPLVDPIHKGLASHFFGLNSVHTAKPSKVKEFVASHGGHTVINKV
LIANGNIAAVKEIRSVRKWAYETFGDERAISFTVMATPEDLAANADYIRMADQYVEVPGGTNNNNYANVE
LIVDVVAERFGVDAWAGWGHASENPPLPESLAASPRKIVFIGPPGAAMRSLGDKISSTIVAQHAKVPCIP
WSGTGVDEVVVDKSTNLVSVSEEVYTKGCTTGPQGLEKAQIGFPVMIKASEGGGGKGIRKVEREEDFE
AAHYQVEGEIPGSPIFIMQLAGNARHLEVQLLADQYGNNSLFGRDCSVQRHQKIEEAPVTAGQQTF
TAMEKAAVRLGKLGVYVSAGTVEYLISHEDDKFYFLELNPRLQVEHPTTEMVTGVNLPAAQLQIAMGIPL
DIRKDIRLFYGVNPHTTPIDPDFSGEDADKTQRRPVPRGHTTACRITSEDPGEGFKPSGGTMHELNFRS
SSNVWGYFSVGNQGGIHSFSDSQFGHI FAFGENRSASRKHMVALKELSIRGDFRVVYLIKLLTPDF
EDNTITTGWLDLISNKLTAEPRDSFLAVVCGAATKAHRASEDSIATYMASLEKGQVPARDILKTLFPVD
FIYEGQRYKFTATRSSEDSYTLFINGSRCIDIGVRPLSDGGILCLVGGRSHNVYKVEVGATRLSVDSKTC
LELEVENDPTQLRSPSPGKLVFKLVEENGDHVRANQPYAEIEVMKMYMLTAQEDGIVQLMKQPGSTIEAGD
ILGILALDDPSVKHAKPFECQLPELGPTLSCGNKPHQRYEHQNVLHNILLGFDNQVVMKSTLQEMVGL
LRNPELPYQWAHQVSSLHTRMSAKLDATLAGLIDKAKQRGGEFPAKQLLRALEKEASSGEVDALFOQTL
APLFDLAREYQDGLAIHELQVAAGLLQAYYDSEARFCGPVRDEDVILKLREENRDSLKVVMQLSHSR
VGAKNNLVALLDEYKVADQAGTDPASNVHVAKYLRPVLRKIVELESRASAKVSLKAREILIQCALPSL
KERTDQLEHILRSSVVESRYGEVGLEHRTPRADILKEVVDSKYIVFDVLAQFFAHDDPWIVLAALELYIR
RACKAYSILDINYHQSDLPPVISWRFRRLPTMSSALYNNSVSSGSKTPSPSVRADSVDFTSYTVERDS
APARTGAIVAVPHLDDLEDALTRVLENLPKRGAGLAISVGASNKSAAAASARDAAAAAASSVDTGLSNICN
VMIGRVDESDDDDTLIARIQSQIVEDFKDEFEAWSLRRITFSFGNSRGTYPKYFTFRGPAYEEDPTIRHIE
PALAFQLELARLSNFDIKPVHTDRNIHVYEATGKNAASDKRFFTRGIVRPGRLRENIPTSEYLISEADR
LMSDILDALVEIGTTNSDNLNHFINFSAVFALKPEEVEAAGFFGLERFGRRWLRLVTGAEIRMMVSDPE
TGSAFPLRAMINNVSGYVVQSELVAEAKNDKGQWIFKSLGKPGSMHRSINTPYPTKEWLQPKRYKAHLM
GTTCYDFPELFRQSIEDWKKYDGKAPDMLTCNELILDEDSGELQEVNREPGANNVGMVAWKPEAKTP
EYPRGRSFIVVANDITFQIGSFGPAEDQFFFVTELARKLGIPRIYLSANS GARIGIADELVGKYKAWN
DETDPSKGFKLYFTPESLATLKPDVTVTTEIEEEGPNGVEKRVIDYIVGEKDGLGVECLRGSGLIAGA
TSRAYKDIFTLTVTCRSVGIGAYLVRLGQRAIQIEGQPIILTGA PAINKLLGREVYSSNLQGGTQIMY
NNGVSHLTARDDLNGVHKIMQWLSYIPASRGLPVPVLPHKTDVWDRDVTQPVVRGEQYDVRWLISGRITLE
DGAFESGLFDKDSFQETLSGWAKGVVVGRARLGGIPFGVIGVETATVDNTTPADPANPDSIEMSTSEAGQ
VWYPNSAFKTSQAINDFNHGGEALPLMILANWRGFSGGQRDMDYNEVLKYGSFIVDALVDYKQPIMVYIPPT

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GELRGGSWVVVDPTINSMDMEMYADVESRGGVLEPEGMVGKIKYRRDKLLDTMARLDPEYSSLKKQLEESP
DSEELKVKLSVREKSLMPIYQQISVQFADLHDHAGRMEAKGVIREALVWKDARRFFFWRIRRRLVEEYLI
TKINSILPSCTRLECLARIKSWKPATLDDQGSDRGVAEWFDENSDAVSARLSELKKDASAQSFSARLKRDR
QGTLOGMKOALASLSEAERAELLKGL*

Knockouts:
PEX10 - YALI0C01023g

(SEQ ID NO.: 71)

Nucleotide =
atgtggggaaagtccacatgcattcgctggtaatctgatctgacactacaactacacaccagggtccaaca
tgagcgacaatacgcacaatcaaaaagccgatccgacccaaaccgatccggacggAACGCGCTGCGTTACGC
tggggccgcagaatcatccggccaaaccgaaagaccactactttgagtcgtgtggacacgcatactc
gtcacgtttctgcagaaatggaaggggagtagcattatccaccagtacaaggaggagctggagacggcgt
ccaagttgcataatctcggttgtgtacgttgcggctcaagactctcgagaaagtagtacaccaatct
catgtacactatcagagaccgaacagcttacccgggggtggtagacggttggctacgtgtttccaaat
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gttttgtgaacaagttgacaagttcacggcgtggagggttaccgcgtccacttggcgtttctac
gtctacggctgtactaccagctcagtaagggatctggggcatgcgttatgtatggacacggactgg
acaagaatgagocctcgaatcggttacgagatgctcggtctgtgtatttgcgggttggccacgttatt
tgtgcagacggaaagagagtagctcgagcgctgtggaaaagagcgtggagaaagaggcaggggagaag
gaagatggaaaggaaagcgggtgtgcggaaaaaaagactgtcaattcggttattgaggatacagaagggg
agacggaaagacaagatcgatctggaggaccctcgacagctcaagttcattcctgaggcgccagacgctg
cactctgtgtctgtcatacattgtgcgcggcatgtacggcatgtggacactttctgtggactgt
atttccgaatgggtgagagagaagggccgagttgtcccttgcggcagggtgtgagagagcagaacttgt
tgcctatcaqataa

(SEQ ID NO.: 72)

Amino acid =
MWGSSHAFAGESDLTLQLHTRSNMSDNTTIKKPIRKPPIRTERLPYAGAAEIIIRANQKDHYFESVLEQH
VTFLQWKWGVRFIHQYKEELETASKFAYLGLCTLVGSKTLGEEYTNLMTIRDRTALPGVVRFGYVLSN
TLPYPLFVRYMGKLRAKLMREYPHLVEYDEDEPVPSPETWKERVIKTFVNKFDFKFTALEGFTAIHLAIFY
VYGSYQQLSKRIWGMRVFGRHLDKNEPRIGYEMLGLLIFARFATSFVQTGREYLGALLEKSVEKEAGEK
EDEKEAVVPKKSSIPFIEDTEGETEDKIDLEDPRQLKFIPEASRACTLCLSYISAPACTPCGHFFCWDC
ISEWVREKPECPCLRQGVREQNLLPIR*

MFE1 - YALI0E15378

(SEQ ID NO.: 73)

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Nucleotide =
atgaccgcacaaggactggatcttgtctacaaggccacgtttcggtgcataaggttaccggactg
cctggccttacttccgaaagcagaagtacggtcgagttatcttaccttccgctgtggtttaegg
aaacctcgcccgacccaactactccgctgccaagctcgccctgggttcggtgagactctcgccaag
gagggtgccaagttacaacattacttccaaacgtcatcgctcttgcgtgttcccaatgaccgagacag
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cgactctgtcaccgagtcttatggtattacgaggctgggtgcgttacatggctaaaatccgatgggg
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tgttaagcgaccccgagaaccccccaaggacccaccgtctccctcaaggaccagggtgtcatgtcaactgg
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gccgggtctggcattggccgagttactctcaccccttgcataagttggtgccaaggcgatgtttaacg
atttcggtaaccctcagaagggtgtcgatcaaattaaggccctcggtgtatcgccgtcgatgacaagaa
caacgtcatccacggtgagaagggtgttcagaccgtatcgacgccttcggtgctgtccacggcggtgc
aacaacgctggatttcccgagacaagtcttcgccaacatggatgatgagatgtggcagctgatcttg
atgtccacctaaccggtaacttactccgttaccaaggccgtggccccacttcctaagcagaagttacgg
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aaggctggtatctcggttctcccgagctttgtcgagagggtgagaagttacaacattctgtcaaca
ccattggccctaaccgtgtactgcatgactgtttctgttcaactggatcaggctccggtaaccgggtatcttcaa
gcccgatttcatcgacccatcacggctctgcttgcgttcccgatcaggctccggtaaccgggtatctgtt
gagactgggtctgtggatcgagactcgatggcagcggatctgggttaaggccctcaacaccaaga
agggtgtcaceccccgaaatgggtcgagacagctggctaaagatcgacttgcattcgatgttactccac
ccatcccaccactccctccgagttactacttcagattttcaacacgtgcctgtatggag
gtttagggagactgtctcggtgtggtcccggtatctcaacaaggagggcgaaccttgc
actacacttacacttaccggagacttacttcagattttcaacacgttgcgttgcaggctatggatggc
gtatgttccgagggtgtatggatgtacttccagaccgtgtccacttccgtgttacatgggtggc
ctcatcaacttacactatgggacttcgttccatcttcaacacgttgcgttgcaggctatggatgt
accttggaaatccgacagtggcatttccatccaatgttacatggagaacaaggctaaaggctatcgatgt
cggtgacaaggccgaaaggctgccttgcgttactgttaccaccaccacgaaacaaggagactgggtggag
gttttctacaacgagttctcttcttcatccggatctgggtttcggtgttacatgggttacatgggtggc
accgtggcgctgcaacttgcgttccatcttccatccggatctgggtttcggtgttacatgggttacatgggtggc
gtgtgttccgagggtgtatggatgtacttccagaccgtgtccacttccgtgttacatgggtggc
tttacgttacatgggttccatccaagaacgttacatgggttacatgggttacatgggtggc
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accggccatcagoaatggccatttggatcgatgttccatccaaggatgttacatgggttacatgggtggc
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(SEQ ID NO.: 74)

Amino Acid =
MTDKDWDLVYKVHVFGAYKVTRAAPYFRKQKYGRVISTSSAAGLYGNFGQTNYSAAKLALVGFGETLAK
EGAKYNITSNVIAPLAASRMTEVMPEDILKLLKPEYVVPLVGYLTHDSVTESYGIYEVGAGYMAKIRWE
RNGAVFKGDDFTPSAILKRWDEVTSFESPTYPNGPADFFKYAEESVKR PENPQGPTVSFKDQVVIVTG
AGAGIGRAYSHLLAKLGAKVVNDFGNPQKVVDIEKALGGIAVADKNNVIHGEKVVQTAIDAFGAVHAVV
NNAGILRDKSFANMDEMWQLIFDVHLNGTYSVTKAAPHPLKQKYGRVINTTSTSGIYGNFGQANYSA
KAGILGFSRALAREGEKYNILVNTIAPNAGTAMTASVFTEEMLELFPDFIAPITVLLASDQAPVTGDLF
ETGSAWIGQTRWQRAGGKAFNTKKGVTPEMVRDSWAKIVDPDDGNSTHPPTPSESTTQILENIFNVPDEE
VEETALVAGPGPGIILNKEGEFDYTYTDLILYLNGLGAKANELKYVFEGLDDDFQTVPTFGVIPYMGG
LITTNYGFVNPFPNPMMLHGEQYLEIRQWPIPTNATLENKAKVIDVVDKGKAALLVTATTTNKETGEE
VFYNESSLFIRGSGGFGGKSTGDRGAATAANKPPARAPDFVKEIKIQEDQAAIYRLSGDYNPLHIDPAF
AAVGNFDRPILHGLCSFGVSGKALYDQFGPKNAKVRFAGHVFPGETLKVEGWKEGNKVIQOTKVVERGT
TAISNAAIELFFPKDAKL*

AC01- YALI0D09361

(SEQ ID NO.: 75)

Nucleotide =
atgctggcttcgagttccatcaaggctgtgagttatcgatggtgaagaaagacaccgacaatcgccac

149

150

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(SEQ ID NO.: 76)

Amino Acid =

MLASRVSIAKAPRLARSLATTTNASLNLDSDKVRMNNWEANNFLNFKKHTENVQIVKERLNRPLTYAEKILY

151

152

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GHLDKPKHEQEIVRGQSYLKLRLPDRACQDATAQMAILQFMSAGIPTVQTPTTVHCDHLIQAQVGGEQDLA
RAIDINKEVYNFLGTASAKYDIGFWKAGSGIIHQIILENYAFCGALLIGSDSHTPNAGGLGMLAIGVGGA
DvvvDMAGLPWELKAPKIIGVLTGKLSGWTSPKDIIKVAGILTVKGGTGAIIVEYFGDGVDNLSTCGMG
TICNMGAEIGATTSTPPFNERMADYLNATGRKEIADFARLYNHFLSADEGCEYDQLIEIDLNTLEPYVNG
PFTPDLATPISKLKDVAVENGWPLEVKVGLIGSCTNSSYEDMERSASIKAKDAMAHGLKSXIYTVPGSE
QIRATIERDGQLQTFLDFGGIVLANACGPCIGQWDRRDIKKGKEKNTIVSSYNRFTGRNDNSNPATHAFVT
SPDLVTAFIAIGDLRFNPLTSDLKDSEGKEFKLKEPTGKGLPDRGYDPGMDTYQAPPADRSAVEVDVSPT
SDRLQILKPFKPWDGKDGIAMDPILIKSLGKTTDHISQAGPWLKYRGHLQNISNNYMIGAINAENEENN
VRNQITGEWGGVPETAIAYRDNGIRVVVVGDNFGEGRSSREHAALEPRFLGGFAITKSFARIHETNLKK
QGLLPLNFVNAGDYDKIQPSDKISILGLKDLAPGKNTIEVTPKDGAKWTTEVSHTYNSEQLEWFKYGSA
LNKMAASKK*

YLYOX1 YALI0E20449g

(SEQ ID NO.: 77)

Nucleotide =
atggatctggcgaaaatcaccgacggcttcgtcaagcacgagacacctcgctgcgtccctcttgcgtccaa
ccaccaaacacaggcccaccccagacttgttccagtgcgcctccaaggaaatgtgagaagcgccacg
agaggacgacctgaaagagtgcgcacacgagcgcggccacaagcaacaacacgcgtacgcgtct
ctcatgtccaccccagagccaaagtgcgtctccccccgactgtgcatttcgcacacctgtatgcaaa
agtccggacaccatgttaccgacagaacctcaactcgaccagtatctactcgacgaggagaaggagaa
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ccccaatatgcattttatcacatccatcacctgcgtaccctgtgaacgcgcctcagattgacaacgcac
ggctggcgccgaaaacgacgcccgaacgttccacggaaactcgcgctgtggagcaggagtttgcgg
caaccagaagcctccaagcacattcgctcgacattgcggcagactcgacatgactgaaaaggctgtg
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ttccaaacgtcagcaccagcgttaagcaacaagtcaatcaactttccatcacagacaacaagtgcgcctg
geacagtcaaccaccggcactctggtgcacacgcacgcacgcacgcacaca
ccgcattccacttccacaaacgactccgaaattgcattccgtcgccccaaaaacaaacggcagctatt
ctctgtttcgaaagatacccccgagactcccgaaaaagaaaccaggactgtctccgcactgtccatgcgt
ggtgggaaggetacttttatctacggcggcaagccaaagggtgtacgtgttccctggaaagacgttgg
gggtccctggccacaccctctcccgccaaacaacaatctggctggggctgcgtccatggccacatc
gtctccatgaccagggaccgcgtcgcaactgaaccaggcatotgcattttccatctggctgtt
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(SEQ ID NO.: 78)

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153

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(SEQ ID NO.: 79)

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155**156**

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It is understood that the examples and embodiments described herein are for illustrative purposes only and that various modifications or changes in light thereof will be suggested to persons skilled in the art and are to be included

within the spirit and purview of this application and scope of the appended claims. All publications, patents, and patent applications cited herein are hereby incorporated by reference in their entirety for all purposes.

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ggccgatctg cgtccaccca cttctttaac gccaagaac gccacccatcg cgagggtat 240

gccaagtaca gagctttca tggccatctg cctcatcagc actctcagag tccctgtcc 300

agatcttcgtt cattttgtcgcc ggccgaaatg aaccacccccc ctccccccacc ctccagccac 360

acccaccaac agccagagga cgtatgcgcata tttccactc gatctcgatc gtcgtctcg 420

gtttctggac gcaaggttcaa cagaaacaga accaagtctg gatcttcgtt gagcaagggt 480

ctccagcagc tcaacatgac cggatcgctc gaagaagagc cctacgagag cgtatgcac 540

gccccactat ctgcggaaaga cgcacattgtc tatgtatgtt cccagaaaaa caccgtcaag 600

ccatatctc ctactctcaa acgcacccgc accaaggacg acatgaagaa catgtccatc 660

aacgacgtca aaatcaccac caccacagaa gatccttttg tggcccgagga gctgtccatg 720

atgttcgaaa aggtgcagta ctgcggagac ctccggagaca agtaccaaac cgtgtcgcta 780

cagaaggacg gagacaaccc caaggatgc aagacacact ggaaaattta ccccgagcct 840

ccaccacccct cttggcacgaa gaccggaaag cgattccgag gctcgccaa aaaggagcac 900

caaaagaaaag acccgacaat ggtatggattc aaattcgagg actgcgaaat ccccgagccc 960

aacgacatgg tcttcaagcg agatcctacc tttgtctatc aggtctatga ggtgaaagc 1020

tctctcaacg aaaataagcc gtttggccat cttccctcaa tccgagatata ctacatggat 1080

ctggaggatc tcattgtggc ttcgtctgac ggacccgtccaa agtctttgc tttccgacgca 1140

ctgcaatatac tagaagccaa gtggAACCTC tactacctgc tcaacgagta cacggagaca 1200

accgagtcacca agaccaaccc ccatcgagac ttttacaacg tacgaaaggt cgacacccac 1260

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| | |
|--|------|
| gttcaccact ctgcctcgat gaaccagaag catctgctgc gattcatcaa atacaagatg | 1320 |
| aagaactgcc ctgatgaagt tgtcatccac cgagacggtc gggagctgac actctccca | 1380 |
| gtgtttgagt cacttaacct gactgcctac gacctgtcta tgcataccct tgatatgcat | 1440 |
| gctcacaagg actcggttcca tcgatttgac aagttcaacc tcaagtacaa ccctgtcggt | 1500 |
| gagtcctcgac tgcgagaaaat cttccctaaag accgacaact acatccaggg tcgataccta | 1560 |
| gctgagatca caaaggagggt gttccaggat ctgcagaact cgaagtacca gatggcggag | 1620 |
| taccgttattt ccatctacgg tcggtccaaag gacgagttggg acaagctggc tgccctgggt | 1680 |
| ctggacaaca aactgttttc gcccaatgtt cggtggttga tccaggtgcc tcgactgtac | 1740 |
| gacatttaca agaaggctgg tctggtaaac accttgcgg acatttgca gaacgtcttt | 1800 |
| gagcctttt tcgaggtcac caaggatccc agtaccatcc ccaagctgca cgtttccctg | 1860 |
| cagcgagttt tgggctttga ctctgtcgat gacgagtcga agctggaccg acgttccac | 1920 |
| cgaaagttcc caactgcagc atactggac agegcacaga accctcccta ctgcgtactgg | 1980 |
| cagtaactatc tatacgccaa catggcctcc atcaacaccc ttggagacagcg tttggcttat | 2040 |
| aatactttt agttgcgacc ccatgcttga gaggctggg acccagagca tcttctgtgc | 2100 |
| acttatctgg ttgctcaggg tatcaaccac ggtattctgt tgcgaaaggt gcccatttcatt | 2160 |
| cagtacctt actacacttggc ccagatcccc attgccatgt ctccctgtgc caacaatgcg | 2220 |
| ctgttccctca cgttcgacaa gaacccttc tactcatact tcaaggggg tctcaacgtg | 2280 |
| tccttgtcat cggatgatcc tctgcagtt gcttacacta aggaggctct gattgaggag | 2340 |
| tactctgtgg ctgcgtcat ttacaagctt tccaaacgtgg atatgtgtga gcttgcgtca | 2400 |
| aactcggtac tgcaatctgg ctttgagcga atcatcaagg agcattggat cggcgaaaaac | 2460 |
| tacgagatcc atggccccga gggcaacaccc atccagaaga caaacgtgcc caatgtgcgt | 2520 |
| ctggccttcc gagacgagac tttgacccac gagcttgctc tgggtggacaa gtacaccaat | 2580 |
| cttgaggagt ttgagcggtt qcatggta | 2609 |

<210> SEQ ID NO 34
<211> LENGTH: 869
<212> TYPE: PRT
<213> ORGANISM: *Yarrowia lipolytica*

<400> SEQUENCE: 34

Met Pro Glu Glu Asp Asp Leu Asp Ser His Phe Val Gly Pro Ile Ser
20 25 30

Pro Arg Pro His Gly Ala Asp Glu Ile Ala Gly Tyr Val Gly Cys Glu
35 40 45

Asp Asp Glu Asp Glu Leu Glu Glu Leu Gly Met Leu Gly Arg Ser Ala
50 55 60

Ser Thr His Phe Ser Tyr Ala Glu Glu Arg His Leu Ile Glu Val Asp
65 70 75 80

Ala Lys Tyr Arg Ala Leu His Gly His Leu Pro His Gln His Ser Gln
25 30 35

Ser Pro Val Ser Arg Ser Ser Ser Phe Val Arg Ala Glu Met Asn His

Pro Pro Pro Pro Pro Ser Ser His Thr His Gln Gln Pro Glu Asp Asp

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Asp Ala Ser Ser Thr Arg Ser Arg Ser Ser Arg Ala Ser Gly Arg
 130 135 140
 Lys Phe Asn Arg Asn Arg Thr Lys Ser Gly Ser Ser Leu Ser Lys Gly
 145 150 155 160
 Leu Gln Gln Leu Asn Met Thr Gly Ser Leu Glu Glu Glu Pro Tyr Glu
 165 170 175
 Ser Asp Asp Asp Ala Arg Leu Ser Ala Glu Asp Asp Ile Val Tyr Asp
 180 185 190
 Ala Thr Gln Lys Asp Thr Cys Lys Pro Ile Ser Pro Thr Leu Lys Arg
 195 200 205
 Thr Arg Thr Lys Asp Asp Met Lys Asn Met Ser Ile Asn Asp Val Lys
 210 215 220
 Ile Thr Thr Thr Glu Asp Pro Leu Val Ala Gln Glu Leu Ser Met
 225 230 235 240
 Met Phe Glu Lys Val Gln Tyr Cys Arg Asp Leu Arg Asp Lys Tyr Gln
 245 250 255
 Thr Val Ser Leu Gln Lys Asp Gly Asp Asn Pro Lys Asp Asp Lys Thr
 260 265 270
 His Trp Lys Ile Tyr Pro Glu Pro Pro Pro Ser Trp His Glu Thr
 275 280 285
 Glu Lys Arg Phe Arg Gly Ser Ser Lys Lys Glu His Gln Lys Lys Asp
 290 295 300
 Pro Thr Met Asp Glu Phe Lys Phe Glu Asp Cys Glu Ile Pro Gly Pro
 305 310 315 320
 Asn Asp Met Val Phe Lys Arg Asp Pro Thr Cys Val Tyr Gln Val Tyr
 325 330 335
 Glu Asp Glu Ser Ser Leu Asn Glu Asn Lys Pro Phe Val Ala Ile Pro
 340 345 350
 Ser Ile Arg Asp Tyr Tyr Met Asp Leu Glu Asp Leu Ile Val Ala Ser
 355 360 365
 Ser Asp Gly Pro Ala Lys Ser Phe Ala Phe Arg Arg Leu Gln Tyr Leu
 370 375 380
 Glu Ala Lys Trp Asn Leu Tyr Tyr Leu Leu Asn Glu Tyr Thr Glu Thr
 385 390 395 400
 Thr Glu Ser Lys Thr Asn Pro His Arg Asp Phe Tyr Asn Val Arg Lys
 405 410 415
 Val Asp Thr His Val His His Ser Ala Cys Met Asn Gln Lys His Leu
 420 425 430
 Leu Arg Phe Ile Lys Tyr Lys Met Lys Asn Cys Pro Asp Glu Val Val
 435 440 445
 Ile His Arg Asp Gly Arg Glu Leu Thr Leu Ser Gln Val Phe Glu Ser
 450 455 460
 Leu Asn Leu Thr Ala Tyr Asp Leu Ser Ile Asp Thr Leu Asp Met His
 465 470 475 480
 Ala His Lys Asp Ser Phe His Arg Phe Asp Lys Phe Asn Leu Lys Tyr
 485 490 495
 Asn Pro Val Gly Glu Ser Arg Leu Arg Glu Ile Phe Leu Lys Thr Asp
 500 505 510
 Asn Tyr Ile Gln Gly Arg Tyr Leu Ala Glu Ile Thr Lys Glu Val Phe
 515 520 525
 Gln Asp Leu Glu Asn Ser Lys Tyr Gln Met Ala Glu Tyr Arg Ile Ser
 530 535 540
 Ile Tyr Gly Arg Ser Lys Asp Glu Trp Asp Lys Leu Ala Ala Trp Val

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| | | | |
|---|-----|-----|-----|
| 545 | 550 | 555 | 560 |
| Leu Asp Asn Lys Leu Phe Ser Pro Asn Val Arg Trp Leu Ile Gln Val | | | |
| 565 | 570 | 575 | |
| Pro Arg Leu Tyr Asp Ile Tyr Lys Ala Gly Leu Val Asn Thr Phe | | | |
| 580 | 585 | 590 | |
| Ala Asp Ile Val Gln Asn Val Phe Glu Pro Leu Phe Glu Val Thr Lys | | | |
| 595 | 600 | 605 | |
| Asp Pro Ser Thr His Pro Lys Leu His Val Phe Leu Gln Arg Val Val | | | |
| 610 | 615 | 620 | |
| Gly Phe Asp Ser Val Asp Asp Glu Ser Lys Leu Asp Arg Arg Phe His | | | |
| 625 | 630 | 635 | 640 |
| Arg Lys Phe Pro Thr Ala Ala Tyr Trp Asp Ser Ala Gln Asn Pro Pro | | | |
| 645 | 650 | 655 | |
| Tyr Ser Tyr Trp Gln Tyr Tyr Leu Tyr Ala Asn Met Ala Ser Ile Asn | | | |
| 660 | 665 | 670 | |
| Thr Trp Arg Gln Arg Leu Gly Tyr Asn Thr Phe Glu Leu Arg Pro His | | | |
| 675 | 680 | 685 | |
| Ala Gly Glu Ala Gly Asp Pro Glu His Leu Leu Cys Thr Tyr Leu Val | | | |
| 690 | 695 | 700 | |
| Ala Gln Gly Ile Asn His Gly Ile Leu Leu Arg Lys Val Pro Phe Ile | | | |
| 705 | 710 | 715 | 720 |
| Gln Tyr Leu Tyr Tyr Leu Asp Gln Ile Pro Ile Ala Met Ser Pro Val | | | |
| 725 | 730 | 735 | |
| Ser Asn Asn Ala Leu Phe Leu Thr Phe Asp Lys Asn Pro Phe Tyr Ser | | | |
| 740 | 745 | 750 | |
| Tyr Phe Lys Arg Gly Leu Asn Val Ser Leu Ser Ser Asp Asp Pro Leu | | | |
| 755 | 760 | 765 | |
| Gln Phe Ala Tyr Thr Lys Glu Ala Leu Ile Glu Glu Tyr Ser Val Ala | | | |
| 770 | 775 | 780 | |
| Ala Leu Ile Tyr Lys Leu Ser Asn Val Asp Met Cys Glu Leu Ala Arg | | | |
| 785 | 790 | 795 | 800 |
| Asn Ser Val Leu Gln Ser Gly Phe Glu Arg Ile Ile Lys Glu His Trp | | | |
| 805 | 810 | 815 | |
| Ile Gly Glu Asn Tyr Glu Ile His Gly Pro Glu Gly Asn Thr Ile Gln | | | |
| 820 | 825 | 830 | |
| Lys Thr Asn Val Pro Asn Val Arg Leu Ala Phe Arg Asp Glu Thr Leu | | | |
| 835 | 840 | 845 | |
| Thr His Glu Leu Ala Leu Val Asp Lys Tyr Thr Asn Leu Glu Glu Phe | | | |
| 850 | 855 | 860 | |
| Glu Arg Leu His Gly | | | |
| 865 | | | |

<210> SEQ ID NO 35

<211> LENGTH: 1218

<212> TYPE: DNA

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 35

| | | | | | | |
|-------------|------------|------------|------------|------------|-------------|-----|
| atggaacctcg | aaactaagaa | gaccaagact | gactccaaga | agattgtct | tctccggccgc | 60 |
| gacttctgtg | gccccgaggt | gattgccag | gccgtcaagg | tgctcaagtc | tgttgcttag | 120 |
| gcctccggca | ccgagttgt | gtttgaggac | cgactcattg | gaggagctgc | cattgagaag | 180 |
| gagggcgagc | ccatcacgca | cgctactctc | gacatctgcc | gaaaggctga | ctcttattatg | 240 |
| ctcgggtctg | tcggaggcgc | tgccaacacc | gtatggacca | ctcccgacgg | acgaaccgac | 300 |

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| | | | | | | |
|------------|-------------|-------------|------------|-------------|------------|------|
| gtgcgacccg | agcagggtct | cctcaagctg | cgaaggacc | tgaacctgta | cgcacactg | 360 |
| cgaccctgcc | agctgtgtc | gccaagctc | ggcgatctct | ccccatccg | aaacgtttag | 420 |
| ggcacccact | tcatcattgt | ccgagagctc | gtcgaggta | tctactttgg | agagcgaag | 480 |
| gaggatgacg | gatctggcgt | cgcttccgac | accgagacct | actccgttcc | tgagggttag | 540 |
| cgaattgccc | aatggccgc | cttctggcc | cttcagcaca | accccccctct | tcccgtgtgg | 600 |
| tctcttaca | aggccaacgt | gtcgccctcc | tctcgacttt | ggcgaaagac | tgtcactcga | 660 |
| gtcctcaagg | acgaattccc | ccagctcgag | ctcaaccacc | agctgatcga | ctcgccgc | 720 |
| atgatcctca | tcaaggcagcc | ctccaagatg | aatggtatca | tcatcaccac | caacatgttt | 780 |
| ggcgatatac | tctccgacga | ggcctccgctc | atccccggtt | ctctgggtct | gctgcctcc | 840 |
| gctctctgg | tttctctgcc | cgacaccaac | gaggcggttc | gtctgtacga | gcccgtcac | 900 |
| ggatctgccc | ccgatctcg | caagcagaag | gtcaacccca | ttgccaccat | tctgtctgcc | 960 |
| gcacatgtgc | tcaagttctc | tcttaacatg | aagcccgccc | gtgacgctgt | tgaggctgcc | 1020 |
| gtcaaggagt | ccgtcgaggc | ttgttatca | accgcccata | tggaggctc | ttcctccacc | 1080 |
| tcggaggctc | gagacttgtt | gccaacaagg | tcaaggagct | gtcaagaag | gagtaagtcg | 1140 |
| tttctacgac | gcattgtatgg | aaggagcaaa | ctgacgcgc | tgcggttgg | tctaccggca | 1200 |
| gggtccgcta | gtgtataa | | | | | 1218 |

<210> SEQ ID NO 36

<211> LENGTH: 400

<212> TYPE: PRT

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 36

| | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Glu | Pro | Glu | Thr | Lys | Lys | Thr | Lys | Thr | Asp | Ser | Lys | Ile | Val |
| 1 | | | | | 5 | | | 10 | | | | 15 | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Leu | Gly | Gly | Asp | Phe | Cys | Gly | Pro | Glu | Val | Ile | Ala | Glu | Ala | Val |
| | | | | | 20 | | | | 25 | | | 30 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Lys | Val | Leu | Lys | Ser | Val | Ala | Glu | Ala | Ser | Gly | Thr | Glu | Phe | Val | Phe |
| | | | | | 35 | | | 40 | | | 45 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|--|
| Glu | Asp | Arg | Leu | Ile | Gly | Gly | Ala | Ala | Ile | Glu | Lys | Glu | Glu | Pro | |
| | | | | | 50 | | | 55 | | | 60 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ile | Thr | Asp | Ala | Thr | Leu | Asp | Ile | Cys | Arg | Lys | Ala | Asp | Ser | Ile | Met |
| 65 | | | | | 70 | | | 75 | | | 80 | | | | |

| | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Gly | Ala | Val | Gly | Gly | Ala | Ala | Asn | Thr | Val | Trp | Thr | Pro | Asp |
| | | | | | 85 | | | 90 | | | 95 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gly | Arg | Thr | Asp | Val | Arg | Pro | Glu | Gln | Gly | Leu | Leu | Lys | Leu | Arg | Lys |
| | | | | | 100 | | | 105 | | | 110 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Asp | Leu | Asn | Leu | Tyr | Ala | Asn | Leu | Arg | Pro | Cys | Gln | Leu | Leu | Ser | Pro |
| | | | | | 115 | | | 120 | | | 125 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Lys | Leu | Ala | Asp | Leu | Ser | Pro | Ile | Arg | Asn | Val | Glu | Gly | Thr | Asp | Phe |
| | | | | | 130 | | | 135 | | | 140 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ile | Ile | Val | Arg | Glu | Leu | Val | Gly | Gly | Ile | Tyr | Phe | Gly | Glu | Arg | Lys |
| 145 | | | | | 150 | | | 155 | | | 160 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Glu | Asp | Asp | Gly | Ser | Gly | Val | Ala | Ser | Asp | Thr | Glu | Thr | Tyr | Ser | Val |
| | | | | | 165 | | | 170 | | | 175 | | | | |

| | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Pro | Glu | Val | Glu | Arg | Ile | Ala | Arg | Met | Ala | Ala | Phe | Leu | Ala | Gln |
| | | | | | 180 | | | 185 | | | 190 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| His | Asn | Pro | Pro | Leu | Pro | Val | Trp | Ser | Leu | Asp | Lys | Ala | Asn | Val | Leu |
| | | | | | 195 | | | 200 | | | 205 | | | | |

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179**180**

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Ala Ser Ser Arg Leu Trp Arg Lys Thr Val Thr Arg Val Leu Lys Asp
 210 215 220

Glu Phe Pro Gln Leu Glu Leu Asn His Gln Leu Ile Asp Ser Ala Ala
 225 230 235 240

Met Ile Leu Ile Lys Gln Pro Ser Lys Met Asn Gly Ile Ile Ile Thr
 245 250 255

Thr Asn Met Phe Gly Asp Ile Ile Ser Asp Glu Ala Ser Val Ile Pro
 260 265 270

Gly Ser Leu Gly Leu Leu Pro Ser Ala Ser Leu Ala Ser Leu Pro Asp
 275 280 285

Thr Asn Glu Ala Phe Gly Leu Tyr Glu Pro Cys His Gly Ser Ala Pro
 290 295 300

Asp Leu Gly Lys Gln Lys Val Asn Pro Ile Ala Thr Ile Leu Ser Ala
 305 310 315 320

Ala Met Met Leu Lys Phe Ser Leu Asn Met Lys Pro Ala Gly Asp Ala
 325 330 335

Val Glu Ala Ala Val Lys Glu Ser Val Glu Ala Gly Ile Thr Thr Ala
 340 345 350

Asp Ile Gly Gly Ser Ser Ser Thr Ser Glu Val Gly Asp Leu Leu Pro
 355 360 365

Thr Arg Ser Arg Ser Cys Ser Arg Arg Ser Lys Ser Phe Leu Arg Arg
 370 375 380

Ile Asp Gly Arg Ser Lys Leu Thr Arg Leu Arg Val Gly Leu Pro Ala
 385 390 395 400

<210> SEQ ID NO 37
 <211> LENGTH: 861
 <212> TYPE: DNA
 <213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 37

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atgcccctc acgaaagctcg agctaacctc cacaagtccg cctttgcgc tcgagtgtc 60
aagctcgatgg cagccaagaa aaccaacctg tggatgttac caccaccaag 120
gagctcattt gatgtccggta taaggatcgatgttgc ctttatgtgttgc 180
gacatcattt acgacttcac ctacggccgc actgtgtcc ccctcaagga acttgcttt 240
aagcacggtt tcttcctgtt cgaggacaga aagttcgacatgttgc 300
caccatgttata agaacgggtt ctaccgttgc gccgatgttgc 360
ggtgttacccg gaaccggaat cattgtggc ctgcgatgttgc gtggcgagga aactgtctt 420
gaacagaaga aggaggacgt ctctgactac gagaactccc agtacaagga gttcctggc 480
ccctctccca acgagaagct ggccagaggt ctgctcatgc tggccgagct gtcttgcaag 540
ggctctgttgc ccactggcgatgttgc gatgttgc 600
gatgttgc 660
attctgaccc cccgggtggg tcttgacgttgc aaggagacgt ctctggaca gcaatgttgc 720
actgttgc 780
ggccagaacc gagatcctat tgaggaggcc aagcgatacc agaaggctgg ctgggaggct 840
taccagaaga ttaactgttgc 861

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<210> SEQ ID NO 38
 <211> LENGTH: 286
 <212> TYPE: PRT

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<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 38

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Pro | Ser | Tyr | Glu | Ala | Arg | Ala | Asn | Val | His | Lys | Ser | Ala | Phe | Ala |
| 1 | | | | 5 | | | | 10 | | | | | | 15 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ala | Arg | Val | Leu | Lys | Leu | Val | Ala | Ala | Lys | Lys | Thr | Asn | Leu | Cys | Ala |
| | | 20 | | | | 25 | | | | | 30 | | | | |

| | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ser | Leu | Asp | Val | Thr | Thr | Lys | Glu | Leu | Ile | Glu | Leu | Ala | Asp | Lys |
| | | 35 | | | 40 | | | | 45 | | | | | |

| | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Val | Gly | Pro | Tyr | Val | Cys | Met | Ile | Lys | Thr | His | Ile | Asp | Ile | Asp |
| | | | | 50 | | 55 | | 60 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Asp | Phe | Thr | Tyr | Ala | Gly | Thr | Val | Leu | Pro | Leu | Lys | Glu | Leu | Ala | Leu |
| 65 | | | | | 70 | | | 75 | | | | 80 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Lys | His | Gly | Phe | Phe | Leu | Phe | Glu | Asp | Arg | Lys | Phe | Ala | Asp | Ile | Gly |
| | | | | 85 | | | 90 | | 95 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Asn | Thr | Val | Lys | His | Gln | Tyr | Lys | Asn | Gly | Val | Tyr | Arg | Ile | Ala | Glu |
| | | 100 | | | | | 105 | | | 110 | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Trp | Ser | Asp | Ile | Thr | Asn | Ala | His | Gly | Val | Pro | Gly | Thr | Gly | Ile | Ile |
| | | 115 | | | | 120 | | | 125 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ala | Gly | Leu | Arg | Ala | Gly | Ala | Glu | Glu | Thr | Val | Ser | Glu | Gln | Lys | Lys |
| | | 130 | | | 135 | | | 140 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Glu | Asp | Val | Ser | Asp | Tyr | Glu | Asn | Ser | Gln | Tyr | Lys | Glu | Phe | Leu | Val |
| 145 | | | | 150 | | | 155 | | | 160 | | | | | |

| | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Pro | Ser | Pro | Glu | Lys | Leu | Ala | Arg | Gly | Leu | Leu | Met | Leu | Ala | Glu |
| | | 165 | | | 170 | | 175 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Ser | Cys | Lys | Gly | Ser | Leu | Ala | Thr | Gly | Glu | Tyr | Ser | Lys | Gln | Thr |
| | | 180 | | | | 185 | | | 190 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ile | Glu | Leu | Ala | Arg | Ser | Asp | Pro | Glu | Phe | Val | Val | Gly | Phe | Ile | Ala |
| | | 195 | | | | 200 | | | 205 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gln | Asn | Arg | Pro | Lys | Gly | Asp | Ser | Glu | Asp | Trp | Leu | Ile | Leu | Thr | Pro |
| | | 210 | | | 215 | | | 220 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gly | Val | Gly | Leu | Asp | Asp | Lys | Gly | Asp | Ala | Leu | Gly | Gln | Gln | Tyr | Arg |
| 225 | | | 230 | | | 235 | | | 240 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Thr | Val | Glu | Asp | Val | Met | Ser | Thr | Gly | Thr | Asp | Ile | Ile | Ile | Val | Gly |
| | | 245 | | | 250 | | | 255 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Arg | Gly | Leu | Tyr | Gly | Gln | Asn | Arg | Asp | Pro | Ile | Glu | Glu | Ala | Lys | Arg |
| | | 260 | | | | 265 | | | 270 | | | | | | |

| | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Tyr | Gln | Lys | Ala | Gly | Trp | Glu | Ala | Tyr | Gln | Lys | Ile | Asn | Cys |
| | | 275 | | | 280 | | | 285 | | | | | |

<210> SEQ ID NO 39

<211> LENGTH: 1953

<212> TYPE: DNA

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 39

atgtctgcca acgagaacat ctcccgattc gacgcccctg tgggcaagga gcaccccgcc 60

tacgagctct tccataacca cacacgatct ttctgtatg gtctccagcc tcgagcctgc 120

cagggttatgc tggacttcga cttcatctgt aagcgagaga accccctccgt ggccggtg 180

atctatccct tcggggcca gttcgtcacc aagatgtact ggggcaccaa ggagactctt 240

ctccctgtct accagcaggt cgagaaggcc gctgccaagg acccccgaggt cgatgtcg 300

gtcaactttg cttccctctcg atccgtctac tcctctacca tggagctgtc cgagtaccc 360

cagttccgaa ccatacgccat tattgccgag ggtgtccccg agcgacgagc ccgagagatc 420

-continued

| | | | | | | |
|------------|-------------|--------------|-------------|-------------|-------------|------|
| ctccacaagg | cccagaagaa | gggtgtgacc | atcattggtc | ccgctaccgt | cgaggtatc | 480 |
| aagcccggtt | gcttcaaggt | tggaaacacc | ggaggtatga | tggacaacat | tgtcgctcc | 540 |
| aagctctacc | gaccggctc | cgttgctac | gtctccaagt | ccggaggaat | gtccaacgag | 600 |
| ctgaacaaca | ttatctctca | caccaccgac | ggtgtctacg | agggttattgc | tattgggtgt | 660 |
| gaccgatacc | ctggtaactac | cttcattgac | catatcctgc | gatacggaggc | cgacccaaag | 720 |
| tgtaagatca | tcgtctctct | tggtgagggt | ggtggtgttg | aggagtacccg | agtcatcgag | 780 |
| gtgttaaga | acggccagat | caagaagccc | atcgtcgctt | gggccattgg | tacttgcgc | 840 |
| tccatgttca | agactgaggt | tcagttcggc | cacggccggc | ccatggccaa | ctccgacactg | 900 |
| gagactgcca | aggctaagaa | cgccgcccattg | aagtctgtcg | gcttctacgt | ccccgatacc | 960 |
| ttcgaggaca | tgcccgaggt | ccttgcgcag | ctctacgaga | agatggtcgc | caagggcgag | 1020 |
| ctgtctcgaa | tctctgagcc | tgaggtcccc | aagatcccc | ttgactactc | ttgggcccag | 1080 |
| gagcttggtc | ttatccgaaa | gccccgtgt | ttcatctccca | ctatccgaa | tgaccgaggc | 1140 |
| caggagttc | tgtacgctgg | catgcccatt | tccgagggttt | tcaaggagga | cattggatc | 1200 |
| ggcggtgtca | tgtctctgt | gtgggtccga | cgacgactcc | ccgactacgc | ctccaagttt | 1260 |
| tttgagatgg | ttctcatgt | tactgctgac | cacggtcccg | ccgtatccgg | tgccatgaac | 1320 |
| accattatca | ccacccgagc | tggtaaggat | ctcatttctt | ccctggttgc | ttggctctcg | 1380 |
| accattggta | cccgattcgg | agggtctt | gacggtgctg | ccaccgagtt | caccactgcc | 1440 |
| taclacaagg | gtctgtcccc | ccgacagtcc | gttgatacc | tgcgaaagca | gaacaagctg | 1500 |
| attcctggta | ttggccatcg | agtcaagtct | cgaaacaacc | ccgatttccg | agtcgagctt | 1560 |
| gtcaaggact | ttgttaagaa | gaacttcccc | tccacccagc | tgctcgacta | cgcccttgct | 1620 |
| gtcgaggagg | tcaccaccc | caagaaggac | aacctgattc | tgaacgttga | cggtgctatt | 1680 |
| gtctgttctt | ttgtcgatct | catcgatct | tgcgggtcct | ttactgtgga | ggagactgag | 1740 |
| gactaccta | agaacgggt | tctcaacgg | ctgttcgttc | tccggcgtatc | cattggtctc | 1800 |
| attggccacc | atctcgatca | gaagcgactc | aagaccgggt | tgtacccgaca | tccttggac | 1860 |
| gatatcacct | acctgggtgg | ccaggaggct | atccagaaga | agcgagtcga | gatcagcgcc | 1920 |
| ggcgacgttt | ccaaggccaa | gactcgatca | tag | | | 1953 |

<210> SEQ ID NO 40

<211> LENGTH: 650

<212> TYPE: PRT

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 40

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Ser | Ala | Asn | Glu | Asn | Ile | Ser | Arg | Phe | Asp | Ala | Pro | Val | Gly | Lys |
| 1 | | | | 5 | | | | 10 | | | | | | 15 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Glu | His | Pro | Ala | Tyr | Glu | Leu | Phe | His | Asn | His | Thr | Arg | Ser | Phe | Val |
| | | | | 20 | | | | 25 | | | | | | 30 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Tyr | Gly | Leu | Gln | Pro | Arg | Ala | Cys | Gln | Gly | Met | Leu | Asp | Phe | Asp | Phe |
| | | | | 35 | | | | 40 | | | | | | 45 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ile | Cys | Lys | Arg | Glu | Asn | Pro | Ser | Val | Ala | Gly | Val | Ile | Tyr | Pro | Phe |
| | | | | 50 | | | | 55 | | | | | | 60 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gly | Gly | Gln | Phe | Val | Thr | Lys | Met | Tyr | Trp | Gly | Thr | Lys | Glu | Thr | Leu |
| | | | | 65 | | | | 70 | | | | | | 75 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Pro | Val | Tyr | Gln | Gln | Val | Glu | Lys | Ala | Ala | Ala | Lys | His | Pro | Glu |
| | | | | | | | | 85 | | | | | | 90 | |

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Val Asp Val Val Asn Phe Ala Ser Ser Arg Ser Val Tyr Ser Ser
100 105 110

Thr Met Glu Leu Leu Glu Tyr Pro Gln Phe Arg Thr Ile Ala Ile Ile
115 120 125

Ala Glu Gly Val Pro Glu Arg Arg Ala Arg Glu Ile Leu His Lys Ala
130 135 140

Gln Lys Lys Gly Val Thr Ile Ile Gly Pro Ala Thr Val Gly Gly Ile
145 150 155 160

Lys Pro Gly Cys Phe Lys Val Gly Asn Thr Gly Gly Met Met Asp Asn
165 170 175

Ile Val Ala Ser Lys Leu Tyr Arg Pro Gly Ser Val Ala Tyr Val Ser
180 185 190

Lys Ser Gly Gly Met Ser Asn Glu Leu Asn Asn Ile Ile Ser His Thr
195 200 205

Thr Asp Gly Val Tyr Glu Gly Ile Ala Ile Gly Gly Asp Arg Tyr Pro
210 215 220

Gly Thr Thr Phe Ile Asp His Ile Leu Arg Tyr Glu Ala Asp Pro Lys
225 230 235 240

Cys Lys Ile Ile Val Leu Leu Gly Glu Val Gly Gly Val Glu Glu Tyr
245 250 255

Arg Val Ile Glu Ala Val Lys Asn Gly Gln Ile Lys Lys Pro Ile Val
260 265 270

Ala Trp Ala Ile Gly Thr Cys Ala Ser Met Phe Lys Thr Glu Val Gln
275 280 285

Phe Gly His Ala Gly Ser Met Ala Asn Ser Asp Leu Glu Thr Ala Lys
290 295 300

Ala Lys Asn Ala Ala Met Lys Ser Ala Gly Phe Tyr Val Pro Asp Thr
305 310 315 320

Phe Glu Asp Met Pro Glu Val Leu Ala Glu Leu Tyr Glu Lys Met Val
325 330 335

Ala Lys Gly Glu Leu Ser Arg Ile Ser Glu Pro Glu Val Pro Lys Ile
340 345 350

Pro Ile Asp Tyr Ser Trp Ala Gln Glu Leu Gly Leu Ile Arg Lys Pro
355 360 365

Ala Ala Phe Ile Ser Thr Ile Ser Asp Asp Arg Gly Gln Glu Leu Leu
370 375 380

Tyr Ala Gly Met Pro Ile Ser Glu Val Phe Lys Glu Asp Ile Gly Ile
385 390 395 400

Gly Gly Val Met Ser Leu Leu Trp Phe Arg Arg Arg Leu Pro Asp Tyr
405 410 415

Ala Ser Lys Phe Leu Glu Met Val Leu Met Leu Thr Ala Asp His Gly
420 425 430

Pro Ala Val Ser Gly Ala Met Asn Thr Ile Ile Thr Thr Arg Ala Gly
435 440 445

Lys Asp Leu Ile Ser Ser Leu Val Ala Gly Leu Leu Thr Ile Gly Thr
450 455 460

Arg Phe Gly Gly Ala Leu Asp Gly Ala Ala Thr Glu Phe Thr Thr Ala
465 470 475 480

Tyr Asp Lys Gly Leu Ser Pro Arg Gln Phe Val Asp Thr Met Arg Lys
485 490 495

Gln Asn Lys Leu Ile Pro Gly Ile Gly His Arg Val Lys Ser Arg Asn
500 505 510

Asn Pro Asp Phe Arg Val Glu Leu Val Lys Asp Phe Val Lys Lys Asn

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| | | |
|---|-----|-----|
| 515 | 520 | 525 |
| Phe Pro Ser Thr Gln Leu Leu Asp Tyr Ala Leu Ala Val Glu Glu Val | | |
| 530 | 535 | 540 |
| Thr Thr Ser Lys Lys Asp Asn Leu Ile Leu Asn Val Asp Gly Ala Ile | | |
| 545 | 550 | 555 |
| Ala Val Ser Phe Val Asp Leu Met Arg Ser Cys Gly Ala Phe Thr Val | | |
| 565 | 570 | 575 |
| Glu Glu Thr Glu Asp Tyr Leu Lys Asn Gly Val Leu Asn Gly Leu Phe | | |
| 580 | 585 | 590 |
| Val Leu Gly Arg Ser Ile Gly Leu Ile Ala His His Leu Asp Gln Lys | | |
| 595 | 600 | 605 |
| Arg Leu Lys Thr Gly Leu Tyr Arg His Pro Trp Asp Asp Ile Thr Tyr | | |
| 610 | 615 | 620 |
| Leu Val Gly Gln Glu Ala Ile Gln Lys Lys Arg Val Glu Ile Ser Ala | | |
| 625 | 630 | 635 |
| Gly Asp Val Ser Lys Ala Lys Thr Arg Ser | | |
| 645 | 650 | |

<210> SEQ_ID NO 41
<211> LENGTH: 1494
<212> TYPE: DNA
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 41

| | |
|---|------|
| atgtcagcga aatccattca cgaggccgac ggcaaggccc tgctcgacata ctttctgtcc | 60 |
| aaggcgcccg tggggccgaa gcagcagccc atcaacacgt ttgaaatggg cacacccaag | 120 |
| ctggcgcttc tgacgttcga ggacggcggt gcccccgagc agatcttcgc cgccgctgaa | 180 |
| aaagacctacc cctgggtgct ggagtccggc gccaagttt tggccaagcc cgaccagctc | 240 |
| atcaagcgac gaggcaaggc cggcctgttg gtactcaaca agtcgtgggaa ggagtgcag | 300 |
| ccctggatcg ccgagcgggc cgccaagccc atcaacgtgg agggcattga cggagtgttg | 360 |
| cgaacgttcc tggtcgagcc ctgttgccca caccgaccaga agcacgagta ctacatcaac | 420 |
| atccactccg tgcgagaggg cgactggatc ctcttctacc acgaggaggagg agtcgacgatc | 480 |
| ggcgacgtgg acgccaaggc cgccaagatc ctcattcccc ttgacattga gaacgagttac | 540 |
| ccctccaacg ccacgctcac caaggagctg ctggcacacg tgcccggagga ccagcaccag | 600 |
| accctgtctcg acttcatcaa ccggctctac gccgtctacg tcgatctgca gtttacgtat | 660 |
| ctggagatca acccccttgtt cgtatcccc accggccagg gctcgaggt ccactacctg | 720 |
| gatcttgccg gcaagctcgaa ccagaccgca gagtttgagttt gggccccaa gtgggctgct | 780 |
| ggcgccgtccc ccggcgctct gggccaggtc gtcaccatttg acgcccggctc caccaagggt | 840 |
| tccatcgacg ccggccccgc catggcttcc cccgctcattt tcggtcgaga gctgtccaa | 900 |
| gaggaggcgatc acattgcggaa gctcgattcc aagaccggag cttctctgaa gctgactgtt | 960 |
| ctcaatgcca agggccgaat ctggaccctt gtggctggtg gaggagccctc cgtcgtctac | 1020 |
| ccgcacgcca ttgcgtctgc cggcttgcgt gacgagctcg ccaactacgg cgagtactct | 1080 |
| ggcgctccca acgagaccca gacctacgag tacgccaaaa ccgtactgga tctcatgacc | 1140 |
| cgggggcagc ctcaccccgaa gggcaaggta ctgttcatttg gggggagaaat cgccaaacttc | 1200 |
| accaggatgg gatccacccctt caaggccatc atccggccctt tcggggacta ccagtcttct | 1260 |
| ctgcacaacc acaagggtgaa gatttacgtg cgacgaggcg gtcccaactg gcaggagggt | 1320 |
| ctgcgggttga tcaagtcggc tggcgacgag ctgaatctgc ccatggagat ttacggcccc | 1380 |

US 9,896,691 B2

189**190**

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gacatgcacg tgcgggtat tgcccttg gctctgcttg gaaagcggcc caagaatgtc 1440
 aaggcttttg gcacccggacc ttctactgag gcttccactc ctctcgaggat ttaa 1494

 <210> SEQ_ID NO 42
 <211> LENGTH: 497
 <212> TYPE: PRT
 <213> ORGANISM: Yarrowia lipolytica

 <400> SEQUENCE: 42

 Met Ser Ala Lys Ser Ile His Glu Ala Asp Gly Lys Ala Leu Leu Ala
 1 5 10 15

 His Phe Leu Ser Lys Ala Pro Val Trp Ala Glu Gln Gln Pro Ile Asn
 20 25 30

 Thr Phe Glu Met Gly Thr Pro Lys Leu Ala Ser Leu Thr Phe Glu Asp
 35 40 45

 Gly Val Ala Pro Glu Gln Ile Phe Ala Ala Ala Glu Lys Thr Tyr Pro
 50 55 60

 Trp Leu Leu Glu Ser Gly Ala Lys Phe Val Ala Lys Pro Asp Gln Leu
 65 70 75 80

 Ile Lys Arg Arg Gly Lys Ala Gly Leu Leu Val Leu Asn Lys Ser Trp
 85 90 95

 Glu Glu Cys Lys Pro Trp Ile Ala Glu Arg Ala Ala Lys Pro Ile Asn
 100 105 110

 Val Glu Gly Ile Asp Gly Val Leu Arg Thr Phe Leu Val Glu Pro Phe
 115 120 125

 Val Pro His Asp Gln Lys His Glu Tyr Tyr Ile Asn Ile His Ser Val
 130 135 140

 Arg Glu Gly Asp Trp Ile Leu Phe Tyr His Glu Gly Val Asp Val
 145 150 155 160

 Gly Asp Val Asp Ala Lys Ala Ala Lys Ile Leu Ile Pro Val Asp Ile
 165 170 175

 Glu Asn Glu Tyr Pro Ser Asn Ala Thr Leu Thr Lys Glu Leu Leu Ala
 180 185 190

 His Val Pro Glu Asp Gln His Gln Thr Leu Leu Asp Phe Ile Asn Arg
 195 200 205

 Leu Tyr Ala Val Tyr Val Asp Leu Gln Phe Thr Tyr Leu Glu Ile Asn
 210 215 220

 Pro Leu Val Val Ile Pro Thr Ala Gln Gly Val Glu Val His Tyr Leu
 225 230 235 240

 Asp Leu Ala Gly Lys Leu Asp Gln Thr Ala Glu Phe Glu Cys Gly Pro
 245 250 255

 Lys Trp Ala Ala Ala Arg Ser Pro Ala Ala Leu Gly Gln Val Val Thr
 260 265 270

 Ile Asp Ala Gly Ser Thr Lys Val Ser Ile Asp Ala Gly Pro Ala Met
 275 280 285

 Val Phe Pro Ala Pro Phe Gly Arg Glu Leu Ser Lys Glu Glu Ala Tyr
 290 295 300

 Ile Ala Glu Leu Asp Ser Lys Thr Gly Ala Ser Leu Lys Leu Thr Val
 305 310 315 320

 Leu Asn Ala Lys Gly Arg Ile Trp Thr Leu Val Ala Gly Gly Ala
 325 330 335

 Ser Val Val Tyr Ala Asp Ala Ile Ala Ser Ala Gly Phe Ala Asp Glu
 340 345 350

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Leu Ala Asn Tyr Gly Glu Tyr Ser Gly Ala Pro Asn Glu Thr Gln Thr
 355 360 365

Tyr Glu Tyr Ala Lys Thr Val Leu Asp Leu Met Thr Arg Gly Asp Ala
 370 375 380

His Pro Glu Gly Lys Val Leu Phe Ile Gly Gly Ile Ala Asn Phe
 385 390 395 400

Thr Gln Val Gly Ser Thr Phe Lys Gly Ile Ile Arg Ala Phe Arg Asp
 405 410 415

Tyr Gln Ser Ser Leu His Asn His Lys Val Lys Ile Tyr Val Arg Arg
 420 425 430

Gly Gly Pro Asn Trp Gln Glu Gly Leu Arg Leu Ile Lys Ser Ala Gly
 435 440 445

Asp Glu Leu Asn Leu Pro Met Glu Ile Tyr Gly Pro Asp Met His Val
 450 455 460

Ser Gly Ile Val Pro Leu Ala Leu Leu Gly Lys Arg Pro Lys Asn Val
 465 470 475 480

Lys Pro Phe Gly Thr Gly Pro Ser Thr Glu Ala Ser Thr Pro Leu Gly
 485 490 495

Val

<210> SEQ ID NO 43

<211> LENGTH: 1890

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 43

| | | | | | |
|------------------------|-------------|------------|-------------|-------------|------|
| atgttacgac tacgaaccat | gcgaccacaca | cagaccagcg | tcagggcggc | gcttggggcc | 60 |
| accgctgcgg cccgaaacat | gtcctcctcc | agcccctcca | gcttcgata | ctcgcttac | 120 |
| gtcaaggcga cgccggaaat | cggecacgga | aaggcgccca | caacccgtct | gtcggttag | 180 |
| ggccccatct acgtgggctt | cgaaggcatt | cgtcttctca | acctgcccga | tctcaacaag | 240 |
| ggctcgggat tccccctcaa | cgagcgacgg | gaattcggac | tcaagtggct | tctgcctct | 300 |
| gccgaagcca ccctggagga | acaggtcgac | cgagcatacc | aacaattcaa | aaagtgtggc | 360 |
| actcccttag ccaaaaacgg | gttctgcacc | tcgctcaagt | tccaaaacga | ggtgtctac | 420 |
| tacgcccgtc tgctcaagca | cgttaaggag | gtcttcccc | tcatctatac | accgactcag | 480 |
| ggagaagcca ttgaaacagta | ctcgccgctg | ttccggccgg | ccgaaggctg | cttccctcgac | 540 |
| atcaccagtc cctacgacgt | ggaggagcgt | ctgggagcgt | ttggagacca | tgacgacatt | 600 |
| gactacattg tcgtgactga | ctccgagggt | attctcgaa | ttggagacca | aggagtggc | 660 |
| ggtattggta ttccatcgc | caagctggct | ctcatgactc | tatgtgctgg | agtcaacccc | 720 |
| tcacgagtca ttccctgtgg | tctggatacg | ggaaccaaca | accaggagct | gctgcacgac | 780 |
| ccccctgtatc tcggccgacg | aatgccccga | gtgcgaggaa | agcagtagcga | cgacttcatc | 840 |
| gacaactttg tgcagtctgc | ccgaaggctg | tatcccaagg | cggtgatcca | tttcgaggac | 900 |
| tttgggtctcg otaacgcaca | caagatcctc | gacaagtatc | gaccggagat | ccccctgcttc | 960 |
| aacgacgaca tccagggcac | tggagccgtc | actctggcct | ccatcacggc | cgctctcaag | 1020 |
| gtgctggcga aaaatatcac | agataactcga | attctcggt | acggagctgg | ttcggccggc | 1080 |
| atgggtattg ctgaacaggt | ctatgataac | ctgggtgccc | agggtctcga | cgacaagact | 1140 |
| gccccgacaaa acatcttct | catggaccga | ccgggtctac | tgaccaccgc | acttaccgac | 1200 |

-continued

gagcagatga ggcgacgtgca gaagccgttt gccaaggaca aggccaaatta cgagggagtg 1260
 gagaccaaga ctctggagca cgtgggttgc gccgtcaagc cccatattct cattggatgt 1320
 tccactcagc ccggcgccctt taacgagaag gttgtcaagg agatgttaa acacaccct 1380
 cgaccatca ttctccctctt tcacaacccc acacgtttc atgaggctgt ccctgcagat 1440
 ctgtacaagt ggaccgacgg caaggctcg gttgccaccg gtcgcgcctt tgaccaggc 1500
 aacggcaagg agacgtctga gaacaataac tgctttgtt tccccggaat cgggctggga 1560
 gccattctgt ctgcgtcaaa gctcatcacc aacaccatga ttgctgctgc catcgagtgc 1620
 ctgcggacac aggccccat tctcaagaac cacgacgagg gагtacttcc cgacgttagct 1680
 ctcatccaga tcatttcggc ccgggtggcc actgcccgtgg ttcttcaggc caaggctgag 1740
 ggcctagcca ctgtcgagga agagctcaag cccggcacca aggaacatgt gcagattccc 1800
 gacaactttg acgagtgtct cgcctgggtc gagactcaga tgtggcggcc cgtctaccgg 1860
 cctctcatcc atgtgcggga ttacgactag 1890

<210> SEQ ID NO 44

<211> LENGTH: 629

<212> TYPE: PRT

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Synthetic polypeptide

<400> SEQUENCE: 44

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Leu | Arg | Leu | Arg | Thr | Met | Arg | Pro | Thr | Gln | Thr | Ser | Val | Arg | Ala |
| 1 | | | | | | 5 | | | 10 | | | | 15 | | |

| | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ala | Leu | Gly | Pro | Thr | Ala | Ala | Ala | Arg | Asn | Met | Ser | Ser | Ser | Pro |
| | | | | | 20 | | | 25 | | | 30 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ser | Ser | Phe | Glu | Tyr | Ser | Ser | Tyr | Val | Lys | Gly | Thr | Arg | Glu | Ile | Gly |
| | | | | | 35 | | | 40 | | | 45 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| His | Arg | Lys | Ala | Pro | Thr | Thr | Arg | Leu | Ser | Val | Glu | Gly | Pro | Ile | Tyr |
| | | | | | 50 | | | 55 | | | 60 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Val | Gly | Phe | Asp | Gly | Ile | Arg | Leu | Leu | Asn | Leu | Pro | His | Leu | Asn | Lys |
| | | | | | 65 | | | 70 | | 75 | | 80 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gly | Ser | Gly | Phe | Pro | Leu | Asn | Glu | Arg | Arg | Glu | Phe | Gly | Ile | Ser | Gly |
| | | | | | 85 | | | 90 | | | 95 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Leu | Pro | Ser | Ala | Glu | Ala | Thr | Leu | Glu | Glu | Gln | Val | Asp | Arg | Ala |
| | | | | | 100 | | | 105 | | | 110 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Tyr | Gln | Gln | Phe | Lys | Lys | Cys | Gly | Thr | Pro | Leu | Ala | Lys | Asn | Gly | Phe |
| | | | | | 115 | | | 120 | | | 125 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Cys | Thr | Ser | Leu | Lys | Phe | Gln | Asn | Glu | Val | Leu | Tyr | Tyr | Ala | Leu | Leu |
| | | | | | 130 | | | 135 | | | 140 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Lys | His | Val | Lys | Glu | Val | Phe | Pro | Ile | Ile | Tyr | Thr | Pro | Thr | Gln |
| | | | | | 145 | | | 150 | | 155 | | 160 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gly | Glu | Ala | Ile | Glu | Gln | Tyr | Ser | Arg | Leu | Phe | Arg | Arg | Pro | Glu | Gly |
| | | | | | 165 | | | 170 | | | 175 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Cys | Phe | Leu | Asp | Ile | Thr | Ser | Pro | Tyr | Asp | Val | Glu | Glu | Arg | Leu | Gly |
| | | | | | 180 | | | 185 | | | 190 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ala | Phe | Gly | Asp | His | Asp | Asp | Ile | Asp | Tyr | Ile | Val | Val | Thr | Asp | Ser |
| | | | | | 195 | | | 200 | | | 205 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Glu | Gly | Ile | Leu | Gly | Ile | Gly | Asp | Gln | Gly | Val | Gly | Gly | Ile | Gly | Ile |
| | | | | | 210 | | | 215 | | | 220 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ser | Ile | Ala | Lys | Leu | Ala | Leu | Met | Thr | Leu | Cys | Ala | Gly | Val | Asn | Pro |
| | | | | | 225 | | | 230 | | | 235 | | | 240 | |

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Ser Arg Val Ile Pro Val Val Leu Asp Thr Gly Thr Asn Asn Gln Glu
 245 250 255
 Leu Leu His Asp Pro Leu Tyr Leu Gly Arg Arg Met Pro Arg Val Arg
 260 265 270
 Gly Lys Gln Tyr Asp Asp Phe Ile Asp Asn Phe Val Gln Ser Ala Arg
 275 280 285
 Arg Leu Tyr Pro Lys Ala Val Ile His Phe Glu Asp Phe Gly Leu Ala
 290 295 300
 Asn Ala His Lys Ile Leu Asp Lys Tyr Arg Pro Glu Ile Pro Cys Phe
 305 310 315 320
 Asn Asp Asp Ile Gln Gly Thr Gly Ala Val Thr Leu Ala Ser Ile Thr
 325 330 335
 Ala Ala Leu Lys Val Leu Gly Lys Asn Ile Thr Asp Thr Arg Ile Leu
 340 345 350
 Val Tyr Gly Ala Gly Ser Ala Gly Met Gly Ile Ala Glu Gln Val Tyr
 355 360 365
 Asp Asn Leu Val Ala Gln Gly Leu Asp Asp Lys Thr Ala Arg Gln Asn
 370 375 380
 Ile Phe Leu Met Asp Arg Pro Gly Leu Leu Thr Thr Ala Leu Thr Asp
 385 390 395 400
 Glu Gln Met Ser Asp Val Gln Lys Pro Phe Ala Lys Asp Lys Ala Asn
 405 410 415
 Tyr Glu Gly Val Asp Thr Lys Thr Leu Glu His Val Val Ala Ala Val
 420 425 430
 Lys Pro His Ile Leu Ile Gly Cys Ser Thr Gln Pro Gly Ala Phe Asn
 435 440 445
 Glu Lys Val Val Lys Glu Met Leu Lys His Thr Pro Arg Pro Ile Ile
 450 455 460
 Leu Pro Leu Ser Asn Pro Thr Arg Leu His Glu Ala Val Pro Ala Asp
 465 470 475 480
 Leu Tyr Lys Trp Thr Asp Gly Lys Ala Leu Val Ala Thr Gly Ser Pro
 485 490 495
 Phe Asp Pro Val Asn Gly Lys Glu Thr Ser Glu Asn Asn Asn Cys Phe
 500 505 510
 Val Phe Pro Gly Ile Gly Leu Gly Ala Ile Leu Ser Arg Ser Lys Leu
 515 520 525
 Ile Thr Asn Thr Met Ile Ala Ala Ala Ile Glu Cys Leu Ala Glu Gln
 530 535 540
 Ala Pro Ile Leu Lys Asn His Asp Glu Gly Val Leu Pro Asp Val Ala
 545 550 555 560
 Leu Ile Gln Ile Ile Ser Ala Arg Val Ala Thr Ala Val Val Leu Gln
 565 570 575
 Ala Lys Ala Glu Gly Leu Ala Thr Val Glu Glu Glu Leu Lys Pro Gly
 580 585 590
 Thr Lys Glu His Val Gln Ile Pro Asp Asn Phe Asp Glu Cys Leu Ala
 595 600 605
 Trp Val Glu Thr Gln Met Trp Arg Pro Val Tyr Arg Pro Leu Ile His
 610 615 620
 Val Arg Asp Tyr Asp
 625

<210> SEQ ID NO 45
 <211> LENGTH: 1545
 <212> TYPE: DNA

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<213> ORGANISM: *Yarrowia lipolytica*

<400> SEQUENCE: 45

atgactatcg actcacaata ctacaagtcg cgagacaaaa acgacacggc accaaaatc
gegggaatcc gatatgcccc gctatcgaca ccattactca accgatgtga gacttctct
ctggctggc acatttcag cattccact ttcttcacaa ttttcatgtct atgctcgca
atcccactgc tctggccatt tgtgattgctg tatgttagtgt acgctgttaa agacgactcc
ccgtccaacg gaggagtgtt caagcgatac tcgccttattt caagaaactt cttcatctgg
aagcttttgc gcccataact ctgcacaaga cggtggtct ggagccacg
cacacatact accctctgga cgccaggag tatcaccgtt tgctgagag atactggccg
cagaacaagt acctccgagc aatcatctcc accatcgagt actttctgcc cgccatcgatg
aaacggcttc tttctatcaa cgaggcaggag cagcctggccg agcggatcc tctccgtct
cccggttctc ccagctctcc gggttctcaa cctgacaagt ggattaacca cgacacgaga
tatagccgtt gagaatcata tggctccaa ggcacgcctt cgggctccga acttaacggc
aacggcaaca atggcaccac taaccgacga ctttgcgtt ccgcctctgc tggctccact
gcatctgatt ccacgcttct taacgggtcc ctcaactctt acgccaacca gatcattggc
gaaaacgacc cacagctgtc gcccacaaaa ctcaagccca ctggcagaaa atacatcttc
ggctaccacc cccacggcat tatcgccatg ggagccttg tggttgc caccgaggaa
gctggatgtt ccaagcttcc tccggccatc cctgtttctc ttatgactctt caccacaaac
ttccgagtc ctctctacag agagtacctt atgagttctgg gagtcgttcc tgcgttccaaag
aagtctgtca agggcccttca caagcgaaac cagtctatctt gcattgtgt tggttggc
caggaaatgc ttctggccag acccggtgtc atggacctgg tgctactcaa gcaaggggt
tttggcgatc ttggatggaa ggtcgaaat gtgccttgc ttcccatcat ggccttgg
gagaacgacc tctatgacca ggttagcaac gacaagtcgtt ccaagctgtt ccgatccag
cagttgtca agaacttcc tggattcacc ctcttgc tgcgttcccg agggttcc
aactacgatg tcggtttgtt cccctacagg cgaccgtca acattgtgtt tggttcccc
attgacttgc ttatctccc acacccacc gacgaaaga gttccggataa ccacgaccga
tacatcgccg agctgcagcg aatctacaac gggcacaagg atgaatattt catcgatgg
accqaggagg qcaaaaggqac cccqaggttc cgaatgttgc agttaa

<210> SEQ ID NO 46

<211> LENGTH: 514

<212> TYPE: PRT

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 46

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Thr | Ile | Asp | Ser | Gln | Tyr | Tyr | Lys | Ser | Arg | Asp | Lys | Asn | Asp | Thr |
| 1 | | | | 5 | | | | 10 | | | | | | 15 | |

Ala Pro Lys Ile Ala Gly Ile Arg Tyr Ala Pro Leu Ser Thr Pro Leu
 20 25 30

Leu Asn Arg Cys Glu Thr Phe Ser Leu Val Trp His Ile Phe Ser Ile
35 40 45

Pro Thr Phe Leu Thr Ile Phe Met Leu Cys Cys Ala Ile Pro Leu Leu
50 55 60

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| | | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Pro | Ser | Asn | Gly | Gly | Val | Val | Lys | Arg | Tyr | Ser | Pro | Ile | Ser | Arg | Asn | |
| | | | | | 85 | | | | 90 | | | | | | 95 | |
| Phe | Phe | Ile | Trp | Lys | Leu | Phe | Gly | Arg | Tyr | Phe | Pro | Ile | Thr | Leu | His | |
| | | | | | 100 | | | | 105 | | | | | 110 | | |
| Lys | Thr | Val | Asp | Leu | Glu | Pro | Thr | His | Thr | Tyr | Tyr | Pro | Leu | Asp | Val | |
| | | | | | 115 | | | | 120 | | | | 125 | | | |
| Gln | Glu | Tyr | His | Leu | Ile | Ala | Glu | Arg | Tyr | Trp | Pro | Gln | Asn | Lys | Tyr | |
| | | | | | 130 | | | | 135 | | | | 140 | | | |
| Leu | Arg | Ala | Ile | Ile | Ser | Thr | Ile | Glu | Tyr | Phe | Leu | Pro | Ala | Phe | Met | |
| | | | | | 145 | | | | 150 | | | | 155 | | 160 | |
| Lys | Arg | Ser | Leu | Ser | Ile | Asn | Glu | Gln | Glu | Gln | Pro | Ala | Glu | Arg | Asp | |
| | | | | | 165 | | | | 170 | | | | | 175 | | |
| Pro | Leu | Leu | Ser | Pro | Val | Ser | Pro | Ser | Ser | Pro | Gly | Ser | Gln | Pro | Asp | |
| | | | | | 180 | | | | 185 | | | | | 190 | | |
| Lys | Trp | Ile | Asn | His | Asp | Ser | Arg | Tyr | Ser | Arg | Gly | Glu | Ser | Ser | Gly | |
| | | | | | 195 | | | | 200 | | | | 205 | | | |
| Ser | Asn | Gly | His | Ala | Ser | Gly | Ser | Glu | Leu | Asn | Gly | Asn | Gly | Asn | Asn | |
| | | | | | 210 | | | | 215 | | | | 220 | | | |
| Gly | Thr | Thr | Asn | Arg | Arg | Pro | Leu | Ser | Ser | Ala | Ser | Ala | Gly | Ser | Thr | |
| | | | | | 225 | | | | 230 | | | | 235 | | 240 | |
| Ala | Ser | Asp | Ser | Thr | Leu | Leu | Asn | Gly | Ser | Leu | Asn | Ser | Tyr | Ala | Asn | |
| | | | | | 245 | | | | 250 | | | | | 255 | | |
| Gln | Ile | Ile | Gly | Glu | Asn | Asp | Pro | Gln | Leu | Ser | Pro | Thr | Lys | Leu | Lys | |
| | | | | | 260 | | | | 265 | | | | | 270 | | |
| Pro | Thr | Gly | Arg | Lys | Tyr | Ile | Phe | Gly | Tyr | His | Pro | His | Gly | Ile | Ile | |
| | | | | | 275 | | | | 280 | | | | 285 | | | |
| Gly | Met | Gly | Ala | Phe | Gly | Gly | Ile | Ala | Thr | Glu | Gly | Ala | Gly | Trp | Ser | |
| | | | | | 290 | | | | 295 | | | | 300 | | | |
| Lys | Leu | Phe | Pro | Gly | Ile | Pro | Val | Ser | Leu | Met | Thr | Leu | Thr | Asn | Asn | |
| | | | | | 305 | | | | 310 | | | | 315 | | 320 | |
| Phe | Arg | Val | Pro | Leu | Tyr | Arg | Glu | Tyr | Leu | Met | Ser | Leu | Gly | Val | Ala | |
| | | | | | 325 | | | | 330 | | | | | 335 | | |
| Ser | Val | Ser | Lys | Lys | Ser | Cys | Lys | Ala | Leu | Leu | Lys | Arg | Asn | Gln | Ser | |
| | | | | | 340 | | | | 345 | | | | | 350 | | |
| Ile | Cys | Ile | Val | Val | Gly | Gly | Ala | Gln | Glu | Ser | Leu | Leu | Ala | Arg | Pro | |
| | | | | | 355 | | | | 360 | | | | 365 | | | |
| Gly | Val | Met | Asp | Leu | Val | Leu | Leu | Lys | Arg | Lys | Gly | Phe | Val | Arg | Leu | |
| | | | | | 370 | | | | 375 | | | | 380 | | | |
| Gly | Met | Glu | Val | Gly | Asn | Val | Ala | Leu | Val | Pro | Ile | Met | Ala | Phe | Gly | |
| | | | | | 385 | | | | 390 | | | | 395 | | 400 | |
| Glu | Asn | Asp | Leu | Tyr | Asp | Gln | Val | Ser | Asn | Asp | Lys | Ser | Ser | Lys | Leu | |
| | | | | | 405 | | | | 410 | | | | | 415 | | |
| Tyr | Arg | Phe | Gln | Gln | Phe | Val | Lys | Asn | Phe | Leu | Gly | Phe | Thr | Leu | Pro | |
| | | | | | 420 | | | | 425 | | | | | 430 | | |
| Leu | Met | His | Ala | Arg | Gly | Val | Phe | Asn | Tyr | Asp | Val | Gly | Leu | Val | Pro | |
| | | | | | 435 | | | | 440 | | | | 445 | | | |
| Tyr | Arg | Arg | Pro | Val | Asn | Ile | Val | Val | Gly | Ser | Pro | Ile | Asp | Leu | Pro | |
| | | | | | 450 | | | | 455 | | | | 460 | | | |
| Tyr | Leu | Pro | His | Pro | Thr | Asp | Glu | Glu | Val | Ser | Glu | Tyr | His | Asp | Arg | |
| | | | | | 465 | | | | 470 | | | | 475 | | 480 | |
| Tyr | Ile | Ala | Glu | Leu | Gln | Arg | Ile | Tyr | Asn | Glu | Gly | His | Lys | Asp | Glu | Tyr |
| | | | | | 485 | | | | 490 | | | | | 495 | | |
| Phe | Ile | Asp | Trp | Thr | Glu | Glu | Gly | Lys | Gly | Ala | Pro | Glu | Phe | Arg | Met | |

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201

202

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500

505

510

Ile Glu

<210> SEQ ID NO 47
<211> LENGTH: 1581
<212> TYPE: DNA
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 47

| | |
|---|------|
| atggaagtcc gacgacgaaa aatcgacgtg ctcaaggccc agaaaaacgg ctacgaatcg | 60 |
| ggccaccat ctcgacaatc gtcgcagccc tcctcaagag catcgtccag aacccgcaac | 120 |
| aaacactcct cgtccaccct gtcgctcagc ggactgacca tgaaagtcca gaagaaacct | 180 |
| gccccggcccc cggcgaaactc caaacacgcca ttccctacaca tcaagccctg gcacacgtgc | 240 |
| tgctccacat caatgcttgc gcgcgattat gacggctcca accccagctt caagggcttc | 300 |
| aaaaacatcg gcatgatcat tctcattgtg ggaatctac ggctcgatt cgaaaactac | 360 |
| ctcaaatacg gcatttccaa cccgttcttc gaccccaaaa ttactcttc cgagtggcag | 420 |
| ctctcaggct tgctcatagt cgtggcctac gcacatatcc tcatggccta cgctatttag | 480 |
| agcgctgcca agctgctgtt cctctctaga aaacaccact acatggccgt ggggcttctg | 540 |
| cataccatga acactttgtc gtccatctcg ttgctgtcct acgtcgtcta ctactacgt | 600 |
| cccaaccccg tggcaggcac aatagtcgag tttgtggccg ttattctgtc tctcaaactc | 660 |
| gcctcatacg ccctcactaa ctccggatctc cgaaaagccg caattcatgc ccagaagctc | 720 |
| gacaagacgc aagacgataa cgaaaaggaa tccacctcgt cttcccttcc ttcaagatgac | 780 |
| gcagagactt tggcagacat tgacgtcatt cctgcatact acgcacagct gccctacccc | 840 |
| cagaatgtga cgctgtcgaa cctgctgtac ttctggtttgc ctcccacact ggtctaccag | 900 |
| cccggttacc ccaagacgga gcgtattcga cccaaagcac tgatccgaaa cctgttttag | 960 |
| ctcgtctctc tgtgtcatgtc tattcagttt ctcatcttcc agtacgocca ccccatcatg | 1020 |
| cagtcgtgtc tggctctgtt ctccagccc aagctcgatt atgccaacat ctccgagcgc | 1080 |
| ctcatgaagt tggcctccgt gtctatgtat gtctggccta ttggattcta cgcttttttc | 1140 |
| cagaacggtc tcaatcttat tgccgagctc acctgtttt gaaacagaac cttctaccag | 1200 |
| cagtgggtga attcccgctc cattggccag tactggactc tatggaaacaa gccagtcaac | 1260 |
| cagtaactta gacaccacgt ctacgtgcct cttctcgctc ggggcatgtc gcggttcaat | 1320 |
| gcttcgggtgg tgggtttctt ttctccggcc gtcattccatg aactgttgttgcggcatcccc | 1380 |
| actcacaaca tcatcgaggc cgccttcttc ggcattatgtt cgcagggtgcc tctgatcatg | 1440 |
| getactgaga accttcagca tattaactcc tctctggcc cttcccttgg caactgtgca | 1500 |
| ttctggttca ctttttccct gggacaaccc acttgtgtcat tcttttattat tctggcttac | 1560 |
| aactacaagc agaaccagta g | 1581 |

<210> SEQ ID NO 48
<211> LENGTH: 526
<212> TYPE: PRT
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 48

| | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Glu | Val | Arg | Arg | Lys | Ile | Asp | Val | Leu | Lys | Ala | Gln | Lys | Asn |
| 1 | | | | | | | | | | | | | | |
| | | | | | | | 5 | | 10 | | | | 15 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gly | Tyr | Glu | Ser | Gly | Pro | Pro | Ser | Arg | Gln | Ser | Ser | Gln | Pro | Ser | Ser |
| 20 | | | | | | | 25 | | | | | | 30 | | |

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Arg Ala Ser Ser Arg Thr Arg Asn Lys His Ser Ser Ser Thr Leu Ser
 35 40 45
 Leu Ser Gly Leu Thr Met Lys Val Gln Lys Lys Pro Ala Gly Pro Pro
 50 55 60
 Ala Asn Ser Lys Thr Pro Phe Leu His Ile Lys Pro Val His Thr Cys
 65 70 75 80
 Cys Ser Thr Ser Met Leu Ser Arg Asp Tyr Asp Gly Ser Asn Pro Ser
 85 90 95
 Phe Lys Gly Phe Lys Asn Ile Gly Met Ile Ile Leu Ile Val Gly Asn
 100 105 110
 Leu Arg Leu Ala Phe Glu Asn Tyr Leu Lys Tyr Gly Ile Ser Asn Pro
 115 120 125
 Phe Phe Asp Pro Lys Ile Thr Pro Ser Glu Trp Gln Leu Ser Gly Leu
 130 135 140
 Leu Ile Val Val Ala Tyr Ala His Ile Leu Met Ala Tyr Ala Ile Glu
 145 150 155 160
 Ser Ala Ala Lys Leu Leu Phe Leu Ser Ser Lys His His Tyr Met Ala
 165 170 175
 Val Gly Leu Leu His Thr Met Asn Thr Leu Ser Ser Ile Ser Leu Leu
 180 185 190
 Ser Tyr Val Val Tyr Tyr Tyr Leu Pro Asn Pro Val Ala Gly Thr Ile
 195 200 205
 Val Glu Phe Val Ala Val Ile Leu Ser Leu Lys Leu Ala Ser Tyr Ala
 210 215 220
 Leu Thr Asn Ser Asp Leu Arg Lys Ala Ala Ile His Ala Gln Lys Leu
 225 230 235 240
 Asp Lys Thr Gln Asp Asp Asn Glu Lys Glu Ser Thr Ser Ser Ser Ser
 245 250 255
 Ser Ser Asp Asp Ala Glu Thr Leu Ala Asp Ile Asp Val Ile Pro Ala
 260 265 270
 Tyr Tyr Ala Gln Leu Pro Tyr Pro Gln Asn Val Thr Leu Ser Asn Leu
 275 280 285
 Leu Tyr Phe Trp Phe Ala Pro Thr Leu Val Tyr Gln Pro Val Tyr Pro
 290 295 300
 Lys Thr Glu Arg Ile Arg Pro Lys His Val Ile Arg Asn Leu Phe Glu
 305 310 315 320
 Leu Val Ser Leu Cys Met Leu Ile Gln Phe Leu Ile Phe Gln Tyr Ala
 325 330 335
 Tyr Pro Ile Met Gln Ser Cys Leu Ala Leu Phe Phe Gln Pro Lys Leu
 340 345 350
 Asp Tyr Ala Asn Ile Ser Glu Arg Leu Met Lys Leu Ala Ser Val Ser
 355 360 365
 Met Met Val Trp Leu Ile Gly Phe Tyr Ala Phe Phe Gln Asn Gly Leu
 370 375 380
 Asn Leu Ile Ala Glu Leu Thr Cys Phe Gly Asn Arg Thr Phe Tyr Gln
 385 390 395 400
 Gln Trp Trp Asn Ser Arg Ser Ile Gly Gln Tyr Trp Thr Leu Trp Asn
 405 410 415
 Lys Pro Val Asn Gln Tyr Phe Arg His His Val Tyr Val Pro Leu Leu
 420 425 430
 Ala Arg Gly Met Ser Arg Phe Asn Ala Ser Val Val Val Phe Phe Phe
 435 440 445

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Ser Ala Val Ile His Glu Leu Leu Val Gly Ile Pro Thr His Asn Ile
450 455 460

Ile Gly Ala Ala Phe Phe Gly Met Met Ser Gln Val Pro Leu Ile Met
465 470 475 480

Ala Thr Glu Asn Leu Gln His Ile Asn Ser Ser Leu Gly Pro Phe Leu
485 490 495

Gly Asn Cys Ala Phe Trp Phe Thr Phe Phe Leu Gly Gln Pro Thr Cys
500 505 510

Ala Phe Leu Tyr Tyr Leu Ala Tyr Asn Tyr Lys Gln Asn Gln
515 520 525

<210> SEQ ID NO 49

<211> LENGTH: 3810

<212> TYPE: DNA

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 49

| | |
|--|------|
| atggcttaag acaaggaaat cgactttgac tacacgggag aactggtgat ggacgattc | 60 |
| gagttcccca tcgacgacat gctccacaac gacggagatg actttgtcaa gaaggaaacg | 120 |
| tgggacgagg gttttggttt cggaacaaat ggcgccgtgg gtgcgcagat ggacgtccag | 180 |
| accageccat tttagcgtacc ttgttttggc ggctgtggag caggccctga catgtatggt | 240 |
| ctcatggata caaacatgaa ccacatcaac ggttagtcaca acatgaacag cgtcgtcaag | 300 |
| caggaggact actacacacc gtccatggc actccccatga acccccaaca gcaacagtcc | 360 |
| atgaccctc aacagcagca tcacatgaa cacaaccagg cctcteagct ccaatcttg | 420 |
| catcaacagt cccagaaggc tcaaccacag cagcaacaac aacageccaca tcagtcgaca | 480 |
| ggagtgcata gcataatcac aaaggcatac accagggcag caggagaccc accgtacgg | 540 |
| cgaaagtact cacgacaact caacaagtac cccgaggacg tggagtattc atcttcgac | 600 |
| ccatcgctat ggagcaattt gctgaccaac tcggaaactc cgtaccaata ccagatacat | 660 |
| gtccattcca tgcccgaaa atcacgttg gagacccaa tcaaattgtgc attatcaatc | 720 |
| taccctccgc ctccacagca gtccgttca cttccgacag acaccatttc gcgtcccaag | 780 |
| ttccagctca agcagggcca cattccagac tctgtctct ctttggaaatc atacattgtg | 840 |
| ggcgagcaga acccccagaa gcccgtcaat ttgtgttcta gatgcataa acgagaacag | 900 |
| aagcgagcct gtcgaaagaa actctttgac gagtcggagg agctgtcgat ggtcgagact | 960 |
| cgtcaacgc gtctggctgt cttcaactgc tccgagggtc ttgagttcaa ggatgtggaa | 1020 |
| cgccgagttt acatccccga gtccggcact acagttaccc ccaagcagct ggttctgcc | 1080 |
| ctgcgtctgg cttgtactg tagacaccac ggggagaaaa agggatttcg aatcccttt | 1140 |
| tgtcttagag acgagggagg ccagatttg ggtgtggcc agagtggaaac gaccgtcatg | 1200 |
| atcaactgacg accacaaggc tttggggagac ggggttgcca tgccgactac agccactgct | 1260 |
| cctgccacccg ctggctcttc acaacccccc acccaggttc ctacccccc tgcacatctcg | 1320 |
| tcgacgagct atcgctctcg aaactcgctt cctctatcgct tctactccat ggaagactct | 1380 |
| tctgtcgaggat tcacctcgga ccattctcat tactccaaact atggttctaa acgacgacga | 1440 |
| gacggcttcc ccatcagcga ttggagcggc atgatgaacg tgcgaggcat ggatagacag | 1500 |
| gcttccattt ccagcattcc cggaaatgggtt ggtggcatgt cgaacatgac tggccactgt | 1560 |
| gcttcgggta gcgccactaa tctggctgtc cacaacatga acaacccgc agacgaaaac | 1620 |
| ctggccgtca tcaagcgaat catccccctcg cagggttcca ttggaggccg cattgaagta | 1680 |

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| | |
|--|------|
| accctgcttg gatctggctt caagtccaat ctggggctg tttcggtga caacaaggcc | 1740 |
| gtgggcaccc actgtggc tgattcgacc atcgtgaccc atctggccc ttgcgaccatc | 1800 |
| gtgggtcccg ttgtgggtgc tttcaaggt tttgtgtcg acaagectca gattttacc | 1860 |
| tattttacg acacagacgg ccagttgatt gagttggcgc tccaggttgt gggtctcaag | 1920 |
| atgaacggac ggctggaaga cggccgaaac attgcatgc gaatcgtgg caacaatgga | 1980 |
| ggcggtcgcc ggcacaagg cgccatggca ggccggaaaca tgtctaacgg agacggttgg | 2040 |
| atggaaagtgc tgctgcaga cagttcggtt caacccgtat cgccctccac agaccacgaa | 2100 |
| gtatgtggttc tgcgatgtct ggctctcaca gacattcctg gaggccgaat tgccaactgg | 2160 |
| caactcacca acgcccgggg acagaccatg gttcatctgg ccagtattct gggttactcg | 2220 |
| cgtgttctgg tggctctgt ggctcgagga gctcgtgtgg atgttccga caatgggttgg | 2280 |
| ttcacttc ttcatttcgc tgctctctt ggccgtcgaa agattgccaa gaaactactt | 2340 |
| cggtgcaacg ctgaccctta caaacgtAAC cgaattggcg aaaccgtgtt tgatgttgct | 2400 |
| tgtcctcaca ttctcgatct tctggtcggg cctcaggcga tgcctatggc cggtcagacg | 2460 |
| tcgtatactc ccgattacca tcgtcagcgt cgatcttcat cttcttccac tctggcttcc | 2520 |
| attgcatcca tccaggattc gcgtgagttac ggtttctatg accatggaaat gatttccaa | 2580 |
| ctgtcgatca ttccgtccac gtgctccatt cgatcatcgat cttctcagtt tgacgctgaa | 2640 |
| gacgagtggg acgagcgaga tgaggaggat ggagactttg acgacgattc agatgaggac | 2700 |
| tcagacgatg actcagacgc gctcttcatg tctgttagaa agcacgccaa ggccaagtct | 2760 |
| gtggaatctc ctctctctga ggaggaagag cgacttgc gacacattga ggccgaagac | 2820 |
| caggctgtgg aggccctgtt ggctgccgga atcgtcagta gcaatgtacc cgacgtgg | 2880 |
| tcttcaatg actcggatca cgtgagatct gacacttcca ctgagaacaa gtcctttca | 2940 |
| cggtaacttgc accgtactct cagcatggca tcttggacg atgttctggc ttacatttac | 3000 |
| agacccaaggc gagctactgt gcccaacaag cggcttctgg gagctctcc ttcaagtca | 3060 |
| tccacaagat cgccttttc ggaccatccc atcacgttctt cgggagacga gtccgaccga | 3120 |
| accatttctg cacaatcccc ttccggcggt gcccgtcgag gcccgtctca ttctgtccatc | 3180 |
| tcgcgaatgt ggcgataacct gaagaactcg tctggatcg aggccacccg gtctcgatct | 3240 |
| cgagatgcac acggagccgg tgctccccct gcctacgaa aatcttccc tggccatgg | 3300 |
| gttgtccacg acaagaaggt tgtgcagatg gcccgtgtt ctgctgccgaa gactcgatct | 3360 |
| gggcctgttg ggccttcatc ttctggatcg tgcgtccactt ctggggctgc cgctgtgg | 3420 |
| ccctccccac tagccccat tgtggaggac gaggagcgc tggtagaggc ctggagacga | 3480 |
| cagcgcacat ccatggctaa cgtatcgatg ttatgtccat tctggatcg tgcgtgtc | 3540 |
| atggctatttgc ttatgtccat catcaaggcg ttgggtctgt tccctgacca ggtctctgg | 3600 |
| gttgagtctg tggctgagac tgggggtgtc cactggatcg gaggcgttgc caagatgtgg | 3660 |
| ttcaaggacttca ccggggccag ccactcaagg acacctgttc atttgaggccc | 3720 |
| aacagtctgg tagatcgatc tttcgatcg atgaatgggt ggtccgaccc ggaggccc | 3780 |
| attcatcaag cccaggccccca ggctgcatga | 3810 |

<210> SEQ ID NO 50

<211> LENGTH: 1269

<212> TYPE: PRT

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 50

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Met Ala Lys Asp Lys Glu Ile Asp Phe Asp Tyr Thr Gly Glu Leu Val
1 5 10 15

Met Asp Asp Phe Glu Phe Pro Ile Asp Asp Met Leu His Asn Asp Gly
20 25 30

Asp Asp Phe Val Lys Lys Glu Thr Trp Asp Glu Gly Phe Gly Phe Gly
35 40 45

Thr Asn Gly Ala Val Gly Ala Gln Met Asp Val Gln Thr Ser Pro Phe
50 55 60

Ser Asp Pro Val Phe Gly Gly Val Gly Ala Gly Pro Asp Met Met Gly
65 70 75 80

Leu Met Asp Thr Asn Met Asn His Ile Asn Gly Ser His Asn Met Asn
85 90 95

Ser Val Val Lys Gln Glu Asp Tyr Tyr Thr Pro Ser Met Gly Thr Pro
100 105 110

Met Asn Pro Gln Gln Gln Ser Met Thr Pro Gln Gln Gln His His
115 120 125

Met Asn His Asn Gln Pro Ser Gln Leu Gln Ser Leu His Gln Gln Ser
130 135 140

Gln Lys Ala Gln Pro Gln Gln Gln Gln Pro His Gln Ser Thr
145 150 155 160

Gly Val Asp Ser Ile Ile Thr Lys Ala Tyr Thr Arg Ala Ala Gly Asp
165 170 175

Leu Pro Tyr Gly Arg Lys Tyr Ser Arg Gln Leu Asn Lys Tyr Pro Glu
180 185 190

Asp Val Glu Tyr Ser Ser Phe Asp Pro Ser Leu Trp Ser Asn Leu Leu
195 200 205

Thr Asn Ser Glu Thr Pro Tyr Gln Tyr Gln Ile His Val His Ser Met
210 215 220

Pro Gly Lys Ser Arg Val Glu Thr Gln Ile Lys Cys Ala Leu Ser Ile
225 230 235 240

Tyr Pro Pro Pro Gln Gln Ser Val Arg Leu Pro Thr Asp Thr Ile
245 250 255

Ser Arg Pro Lys Phe Gln Leu Lys Gln Gly His Ile Pro Asp Ser Cys
260 265 270

Leu Ser Leu Glu Val Tyr Ile Val Gly Glu Gln Asn Pro Ser Lys Pro
275 280 285

Val Asn Leu Cys Ser Arg Cys Ile Lys Arg Glu Gln Lys Arg Ala Cys
290 295 300

Arg Lys Lys Leu Phe Asp Glu Ser Glu Glu Leu Ser Trp Val Glu Thr
305 310 315 320

Arg Gln Arg Arg Leu Ala Val Phe Asn Cys Ser Glu Val Leu Glu Phe
325 330 335

Lys Asp Val Glu Arg Arg Val Tyr Ile Pro Glu Ser Gly Thr Thr Val
340 345 350

Thr Ala Lys Gln Leu Val Leu Pro Leu Arg Leu Ala Cys Tyr Cys Arg
355 360 365

His His Gly Glu Lys Lys Gly Phe Arg Ile Leu Phe Cys Leu Arg Asp
370 375 380

Glu Gly Gly Gln Ile Val Gly Val Gly Gln Ser Gly Thr Thr Val Met
385 390 395 400

Ile Thr Asp Asp His Lys Val Val Gly Asp Ala Val Ala Met Pro Thr
405 410 415

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Thr Ala Thr Ala Pro Ala Thr Ala Gly Ser Ser Gln Pro Pro Thr Gln
420 425 430

Val Pro Thr Pro Ala Ala Ser Ser Ser Thr Ser Tyr Arg Pro Arg Asn
435 440 445

Ser Leu Pro Leu Ser Pro Thr Ser Met Glu Asp Ser Ser Ser Glu Phe
450 455 460

Thr Ser Asp His Ser His Tyr Ser Asn Tyr Gly Ser Lys Arg Arg Arg
465 470 475 480

Asp Gly Ser Ser Ile Ser Asp Trp Ser Gly Met Met Asn Val Arg Gly
485 490 495

Met Asp Arg Gln Ala Ser Ile Thr Ser Ile Pro Glu Met Val Gly Gly
500 505 510

Met Ser Asn Met Thr Val Ala Ser Ala Ser Gly Ser Ala Thr Asn Leu
515 520 525

Ala Ala His Asn Met Asn Asn Pro Ala Asp Glu Asn Leu Pro Val Ile
530 535 540

Lys Arg Ile Ile Pro Ser Gln Gly Ser Ile Arg Gly Gly Ile Glu Val
545 550 555 560

Thr Leu Leu Gly Ser Gly Phe Lys Ser Asn Leu Val Ala Val Phe Gly
565 570 575

Asp Asn Lys Ala Val Gly Thr His Cys Trp Ser Asp Ser Thr Ile Val
580 585 590

Thr His Leu Pro Pro Ser Thr Ile Val Gly Pro Val Val Val Ser Phe
595 600 605

Glu Gly Phe Val Leu Asp Lys Pro Gln Ile Phe Thr Tyr Phe Asp Asp
610 615 620

Thr Asp Gly Gln Leu Ile Glu Leu Ala Leu Gln Val Val Gly Leu Lys
625 630 635 640

Met Asn Gly Arg Leu Glu Asp Ala Arg Asn Ile Ala Met Arg Ile Val
645 650 655

Gly Asn Asn Gly Gly Val Ala Gly Ala Gln Gly Ala Met Ala Gly Gly
660 665 670

Asn Met Ser Asn Gly Asp Val Gly Met Glu Ser Ala Ala Ala Asp Ser
675 680 685

Ser Val Gln Pro Val Ser Pro Pro Thr Asp His Glu Asp Val Val Leu
690 695 700

Arg Cys Leu Ala Leu Thr Asp Ile Pro Gly Gly Arg Ile Ala Asn Trp
705 710 715 720

Gln Leu Thr Asn Ala Glu Gly Gln Thr Met Val His Leu Ala Ser Ile
725 730 735

Leu Gly Tyr Ser Arg Val Leu Val Ala Leu Val Ala Arg Gly Ala Arg
740 745 750

Val Asp Val Ser Asp Asn Gly Gly Phe Thr Pro Leu His Phe Ala Ala
755 760 765

Leu Phe Gly Arg Arg Lys Ile Ala Lys Lys Leu Leu Arg Cys Asn Ala
770 775 780

Asp Pro Tyr Lys Arg Asn Arg Ile Gly Glu Thr Val Phe Asp Val Ala
785 790 795 800

Cys Pro His Ile Leu Asp Leu Leu Val Gly Pro Gln Gly Met Pro Met
805 810 815

Ala Val Gln Thr Ser Tyr Thr Pro Asp Tyr His Arg Gln Arg Arg Ser
820 825 830

Ser Ser Ser Ser Thr Leu Ala Ser Ile Ala Ser Ile Gln Asp Ser Arg

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| | | |
|---|------|------|
| 835 | 840 | 845 |
| Glu Tyr Gly Phe Tyr Asp His Gly Met Ile Ser Asn Leu Ser His Ile | | |
| 850 | 855 | 860 |
| Pro Ser Thr Cys Ser Ile Arg Ser Ser Thr Ser Gln Phe Asp Ala Glu | | |
| 865 | 870 | 875 |
| Asp Glu Trp Asp Glu Arg Asp Glu Asp Gly Asp Phe Asp Asp Asp | | |
| 885 | 890 | 895 |
| Ser Asp Glu Asp Ser Asp Asp Ser Asp Ala Leu Phe Met Ser Val | | |
| 900 | 905 | 910 |
| Arg Lys His Ala Lys Ala Lys Ser Val Glu Ser Pro Leu Ser Glu Glu | | |
| 915 | 920 | 925 |
| Glu Glu Arg Leu Val Arg His Ile Glu Ala Glu Asp Gln Ala Val Glu | | |
| 930 | 935 | 940 |
| Ala Arg Val Ala Ala Gly Ile Val Ser Ser Asn Val Pro Asp Val Val | | |
| 945 | 950 | 955 |
| Ser Ser Asn Asp Ser Asp His Val Arg Ser Asp Thr Ser Thr Glu Asn | | |
| 965 | 970 | 975 |
| Lys Ser Phe Ser Arg Tyr Phe Asp Arg Thr Leu Ser Met Ala Ser Trp | | |
| 980 | 985 | 990 |
| Asp Asp Val Leu Ala Tyr Ile Tyr Arg Pro Lys Arg Ala Thr Val Pro | | |
| 995 | 1000 | 1005 |
| Asn Lys Arg Ser Ser Gly Ala Pro Pro Ser Val Arg Ser Thr Arg | | |
| 1010 | 1015 | 1020 |
| Ser Pro Leu Ser Asp His Pro Ile Thr Ser Ser Gly Asp Glu Ser | | |
| 1025 | 1030 | 1035 |
| Asp Arg Thr Ile Ser Ala His Ala Pro Ser Gly Gly Ala Gly Arg | | |
| 1040 | 1045 | 1050 |
| Gly Arg Ser His Ser Ser Ile Ser Arg Met Trp Arg Tyr Leu Lys | | |
| 1055 | 1060 | 1065 |
| Asn Ser Ser Ala Asp Glu Ala Thr Arg Ser Arg Ser Arg Asp Ala | | |
| 1070 | 1075 | 1080 |
| Asn Gly Ala Gly Ala Pro Pro Ala Tyr Glu Glu Ile Phe Pro Gly | | |
| 1085 | 1090 | 1095 |
| His Gly Val Val His Asp Lys Lys Val Val Gln Met Ala Ala Ala | | |
| 1100 | 1105 | 1110 |
| Ser Ala Ala Glu Asn Ser Ser Gly Pro Val Gly Ala Ser Ser Ser | | |
| 1115 | 1120 | 1125 |
| Ala Val Ala Ser Thr Ser Ala Ala Ala Val Val Pro Ser Pro | | |
| 1130 | 1135 | 1140 |
| Leu Ala Pro Ile Val Glu Asp Glu Glu Gln Leu Val Glu Ala Trp | | |
| 1145 | 1150 | 1155 |
| Arg Arg Gln Arg Arg Ser Met Ala Asn Asp Arg Met Leu Phe Ala | | |
| 1160 | 1165 | 1170 |
| Phe Trp Leu Pro Val Leu Leu Met Ala Ile Gly Tyr Met Val Ile | | |
| 1175 | 1180 | 1185 |
| Lys Ala Phe Gly Leu Phe Pro Asp Gln Val Ser Ala Val Glu Ser | | |
| 1190 | 1195 | 1200 |
| Val Ala Glu Thr Val Gly Val His Cys Arg Gly Ala Val Ala Lys | | |
| 1205 | 1210 | 1215 |
| Leu Trp Phe Lys Gln Tyr Pro Val His Arg Gly Gln Pro Leu Lys | | |
| 1220 | 1225 | 1230 |
| Asp Thr Cys Ser Phe Glu Pro Asn Ser Leu Val Glu Ser Ala Leu | | |
| 1235 | 1240 | 1245 |

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| | | | | | | | | | | | | | | |
|------|-----|-----|-----|------|-----|-----|-----|------|-----|-----|-----|-----|-----|-----|
| Arg | Gln | Met | Asn | Gly | Trp | Ser | Asp | Arg | Glu | Val | Pro | Ile | His | Gln |
| 1250 | | | | 1255 | | | | 1260 | | | | | | |

| | | | | | |
|------|-----|-----|-----|-----|-----|
| Ala | Gln | Ala | Gln | Ala | Ala |
| 1265 | | | | | |

<210> SEQ ID NO 51
<211> LENGTH: 3810
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 51

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|--|------|
| atggctaaag acaaggaaat cgactttgac tacacgggag aactggtgat ggacgattc | 60 |
| gagttcccca tcgacgacat gctccacaac gacggagatg actttgtcaa gaaggaaacg | 120 |
| tgggacgagg gttttggttt cggaacaaat ggccgcgtgg gtgcgcagat ggacgtccag | 180 |
| accagccccat ttagegaccc ttttttggc ggcgtgggag caggccctga catgtgggt | 240 |
| ctcatggata caaacatgaa ccacatcaac ggttagtcaca acatgaacag cgtcgtaag | 300 |
| caggaggact actacacacc gtccatgggc actccatgaa acccccaaca gcaacagtcc | 360 |
| atgacccctc aacagcagca tcacatgaa cacaaccagg cctctcagct ccaatcttg | 420 |
| catcaacagt cccagaaggc tcaaccacag cagcaacaac aacageccaca tcagtcgaca | 480 |
| ggagtcgata gcataatcac aaaggcatac accagggcag caggagacct accgtacgga | 540 |
| cgaaagtact cacgacaact caacaagtac cccgaggacg tggagtattc atcttcgac | 600 |
| ccatcgctat ggagcaattt gctgaccaac tcggaaactc cgtaccaata ccagatacat | 660 |
| gtccattcca tgcccgaaa atcacgtgtg gagacccaaa tcaaatgtgc attatcaatc | 720 |
| taccctccgc ctccacagca gtccgttca cttccgacag acaccatttc gcgtcccaag | 780 |
| ttccagtc a gacaggcaca cattccagac tctgtctct ctttggaaat atacatttg | 840 |
| ggcgagcaga accccagcaa gcccgtaat ttgtgttcta gatgcataa acgagaacag | 900 |
| aacgcgaccc gtcgaaagaa actctttgac gagtcggagg agctgtcg ggtcgagact | 960 |
| cgtcaacgac gtctggctgt cttcaactgc tccgagggtgc ttgagttcaa ggatgtggaa | 1020 |
| cgccgagtat acatccccga gtccggact acagttaccc ccaagcagct ggttctgcc | 1080 |
| ctgcgtctgg ttgtctactg tagacaccac gggagaaaa agggatttcg aatcctctt | 1140 |
| tgtcttagag acgaggaggcc ctagattgtg ggtgtggcc agagtgaaac gaccgtcatg | 1200 |
| atcaactgacg accacaaggt tgtggagac gcggttgc tggcgactac agccactgct | 1260 |
| cctggccaccc ctggctcttc acaacccccc acccaggttc ctacccccc tgcacatctcg | 1320 |
| tcgacgagct atcgctctcg aaactcgctt cctctatcg ctacttccat ggaagactct | 1380 |
| tcgtcgaggat tcacctcgga ccattctcat tactccaact atggttctaa acgacgacga | 1440 |
| gacggcttccat ccattcgca ttggagcggc atgatgaacg tgcgaggcat ggatagacag | 1500 |
| gttccattt ccacgttcc cggaaatggtt ggtggcatgt cgaacatgac tggccact | 1560 |
| gttccggta gcgccactaa tctggctgt cacaacatgaa acaacccccc agacgaaaac | 1620 |
| ctggccgtca tcaagcgaat catccccctcg cagggttcca ttcgaggccg cattgaagta | 1680 |
| accctgtttg gatctggctt caagtccaat ctgggtggctg ttttgggtga caacaaggcc | 1740 |
| gtggggcaccctt actgctggtc tgattcgacc atcgtgaccc atctggcc ttcgaccatc | 1800 |
| gtgggtcccg ttgtgggtgc ttgcgaaggt tttgtgtcg acaagctca gattttacc | 1860 |

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tatgttttgacg acacagacagg ccagtgttatttggatc gatgtggcgcc tccagggttgttggggctcaag
atgaacagac ggcttggaaaga cgccccaaac attgcccatttcgc gaatctgttgg caacaatggaa
ggcggttgcgg qcgccacaagg cggcatggca ggcgaaaaaca tgtctaacgg agacgttggaa
atggaaaatgt ctgctgcaga cagttcggtt caacccgtat cgcctccac agaccacgaa
gatgtggtttc tgccatgttctt ggctctcaca gacattcttgg gaggccgaat tgccaacttgg
caactcacca acggcgaggg acagaccatg ttcatcttgg ccagtattctt gggttacttgc
cgtgttctgg tggctcttgg ggctcgagga gctcggttgg atgtttccga caatggtggaa
ttcaactccctc ttcatcttgc tgctcttggt ggccgtcgaat agatttccaa gaaactactt
cggtgcaacg ctgaccctta caaacgttaac cgaatttggcg aaaccgttgg ttagtggct
tgtccctcaca ttctcgatct tctggctcggtt cctcaggccaa tgccatcttgc cgttccagac
tcgtataactc ccgatttacca tcgttccatcg cgtatcttcat cttcttccac tctgggttcc
atggcatcca tccaggatcc tcgttccatcg ggttctatgg accatggaaat gatttccaaac
ctgttccatca ttccgttccac gtgttccattt cgtatcttgc cttcttccat ttttttttcc
gacgagttggg acggcgagaa tgaggaggat ggagacttttgc acgacgatcc agatggggac
tcagacgatg acttcacacgc gcttccatcg tctgttagaa agcacccaa ggccaaatct
gtggaaatctc ctctctctga ggagggaaatcg cgttccatcg gacacatttgc ggccggaaac
caggctgtgg aggccctgtgtt ggcttggcgatc atcgatcgatc gcaatgttacc cgttccatcg
tcttccatcg acttcggatca cgttccatcg gacacccaaatcg ttttttttcc
cggttacttttgc accgttacttgc cggatggca ttttttttgc atgttttgc ttatcttcc
agacccaaacg gagactactgtt gcccacaaacg cggatcttgc gacacccaaatcg ttttttttcc
tccacaagat cggatcttgc gacacccaaatcg atcgttccatcg gacacccaaatcg ttttttttcc
accatcttgc cccatggcc ttcggccgtt gccggatcgatc gacacccaaatcg ttttttttcc
tcgttccatcg ggcgttacttgc cggatcttgc gacacccaaatcg ttttttttcc
cgagatgcaa acggcgccgg tgctccatcg gacacccaaatcg ttttttttcc
gttggatccacg acaagaaggtt gtcgttccatcg gacacccaaatcg ttttttttcc
ggccctgttg gacccatcg ttcggatcgatc gacacccaaatcg ttttttttcc
ccctccatcg tagccatcgatc ttttttttcc
cagcgacatcg cccatggccatcg ttttttttcc
atggatccatcg gttatcgatc ttttttttcc
gttggatccatcg ttttttttcc
ttcaaggatcg accctgttca cggaggccatcg ttttttttcc
aacatcgatcg ttttttttcc
attcatcgatcg cccatggccatcg ttttttttcc
atggatccatcg ttttttttcc

<210> SEQ ID NO 52
<211> LENGTH: 1269
<212> TYPE: PRT
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Synthetic polypeptide

<400> SEQUENCE: 52

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Ala | Lys | Asp | Lys | Glu | Ile | Asp | Phe | Asp | Tyr | Thr | Gly | Glu | Leu | Val |
| 1 | | | | 5 | | | | | 10 | | | | | 15 | |

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219**220**

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Met Asp Asp Phe Glu Phe Pro Ile Asp Asp Met Leu His Asn Asp Gly
 20 25 30

Asp Asp Phe Val Lys Lys Glu Thr Trp Asp Glu Gly Phe Gly Phe Gly
 35 40 45

Thr Asn Gly Ala Val Gly Ala Gln Met Asp Val Gln Thr Ser Pro Phe
 50 55 60

Ser Asp Pro Val Phe Gly Gly Val Gly Ala Gly Pro Asp Met Met Gly
 65 70 75 80

Leu Met Asp Thr Asn Met Asn His Ile Asn Gly Ser His Asn Met Asn
 85 90 95

Ser Val Val Lys Gln Glu Asp Tyr Tyr Thr Pro Ser Met Gly Thr Pro
 100 105 110

Met Asn Pro Gln Gln Gln Ser Met Thr Pro Gln Gln Gln His His
 115 120 125

Met Asn His Asn Gln Pro Ser Gln Leu Gln Ser Leu His Gln Gln Ser
 130 135 140

Gln Lys Ala Gln Pro Gln Gln Gln Gln Pro His Gln Ser Thr
 145 150 155 160

Gly Val Asp Ser Ile Ile Thr Lys Ala Tyr Thr Arg Ala Ala Gly Asp
 165 170 175

Leu Pro Tyr Gly Arg Lys Tyr Ser Arg Gln Leu Asn Lys Tyr Pro Glu
 180 185 190

Asp Val Glu Tyr Ser Ser Phe Asp Pro Ser Leu Trp Ser Asn Leu Leu
 195 200 205

Thr Asn Ser Glu Thr Pro Tyr Gln Tyr Gln Ile His Val His Ser Met
 210 215 220

Pro Gly Lys Ser Arg Val Glu Thr Gln Ile Lys Cys Ala Leu Ser Ile
 225 230 235 240

Tyr Pro Pro Pro Gln Gln Ser Val Arg Leu Pro Thr Asp Thr Ile
 245 250 255

Ser Arg Pro Lys Phe Gln Leu Lys Gln Gly His Ile Pro Asp Ser Cys
 260 265 270

Leu Ser Leu Glu Val Tyr Ile Val Gly Glu Gln Asn Pro Ser Lys Pro
 275 280 285

Val Asn Leu Cys Ser Arg Cys Ile Lys Arg Glu Gln Lys Arg Ala Cys
 290 295 300

Arg Lys Lys Leu Phe Asp Glu Ser Glu Glu Leu Ser Trp Val Glu Thr
 305 310 315 320

Arg Gln Arg Arg Leu Ala Val Phe Asn Cys Ser Glu Val Leu Glu Phe
 325 330 335

Lys Asp Val Glu Arg Arg Val Tyr Ile Pro Glu Ser Gly Thr Thr Val
 340 345 350

Thr Ala Lys Gln Leu Val Leu Pro Leu Arg Leu Ala Cys Tyr Cys Arg
 355 360 365

His His Gly Glu Lys Lys Gly Phe Arg Ile Leu Phe Cys Leu Arg Asp
 370 375 380

Glu Gly Gly Gln Ile Val Gly Val Gly Gln Ser Gly Thr Thr Val Met
 385 390 395 400

Ile Thr Asp Asp His Lys Val Val Gly Asp Ala Val Ala Met Pro Thr
 405 410 415

Thr Ala Thr Ala Pro Ala Thr Ala Gly Ser Ser Gln Pro Pro Thr Gln
 420 425 430

Val Pro Thr Pro Ala Ala Ser Ser Thr Ser Tyr Arg Pro Arg Asn

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221**222**

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435 440 445

Ser Leu Pro Leu Ser Pro Thr Ser Met Glu Asp Ser Ser Ser Glu Phe
 450 455 460

Thr Ser Asp His Ser His Tyr Ser Asn Tyr Gly Ser Lys Arg Arg Arg
 465 470 475 480

Asp Gly Ser Ser Ile Ser Asp Trp Ser Gly Met Met Asn Val Arg Gly
 485 490 495

Met Asp Arg Gln Ala Ser Ile Thr Ser Ile Pro Glu Met Val Gly Gly
 500 505 510

Met Ser Asn Met Thr Val Ala Ser Ala Ser Gly Ser Ala Thr Asn Leu
 515 520 525

Ala Ala His Asn Met Asn Asn Pro Ala Asp Glu Asn Leu Pro Val Ile
 530 535 540

Lys Arg Ile Ile Pro Ser Gln Gly Ser Ile Arg Gly Gly Ile Glu Val
 545 550 555 560

Thr Leu Leu Gly Ser Gly Phe Lys Ser Asn Leu Val Ala Val Phe Gly
 565 570 575

Asp Asn Lys Ala Val Gly Thr His Cys Trp Ser Asp Ser Thr Ile Val
 580 585 590

Thr His Leu Pro Pro Ser Thr Ile Val Gly Pro Val Val Val Ser Phe
 595 600 605

Glu Gly Phe Val Leu Asp Lys Pro Gln Ile Phe Thr Tyr Phe Asp Asp
 610 615 620

Thr Asp Gly Gln Leu Ile Glu Leu Ala Leu Gln Val Val Gly Leu Lys
 625 630 635 640

Met Asn Arg Arg Leu Glu Asp Ala Arg Asn Ile Ala Met Arg Ile Val
 645 650 655

Gly Asn Asn Gly Gly Val Ala Gly Ala Gln Gly Ala Met Ala Gly Gly
 660 665 670

Asn Met Ser Asn Gly Asp Val Gly Met Glu Ser Ala Ala Asp Ser
 675 680 685

Ser Val Gln Pro Val Ser Pro Pro Thr Asp His Glu Asp Val Val Leu
 690 695 700

Arg Cys Leu Ala Leu Thr Asp Ile Pro Gly Gly Arg Ile Ala Asn Trp
 705 710 715 720

Gln Leu Thr Asn Ala Glu Gly Gln Thr Met Val His Leu Ala Ser Ile
 725 730 735

Leu Gly Tyr Ser Arg Val Leu Val Ala Leu Val Ala Arg Gly Ala Arg
 740 745 750

Val Asp Val Ser Asp Asn Gly Gly Phe Thr Pro Leu His Phe Ala Ala
 755 760 765

Leu Phe Gly Arg Arg Lys Ile Ala Lys Lys Leu Leu Arg Cys Asn Ala
 770 775 780

Asp Pro Tyr Lys Arg Asn Arg Ile Gly Glu Thr Val Phe Asp Val Ala
 785 790 795 800

Cys Pro His Ile Leu Asp Leu Leu Val Gly Pro Gln Gly Met Pro Met
 805 810 815

Ala Val Gln Thr Ser Tyr Thr Pro Asp Tyr His Arg Gln Arg Arg Ser
 820 825 830

Ser Ser Ser Ser Thr Leu Ala Ser Ile Ala Ser Ile Gln Asp Ser Arg
 835 840 845

Glu Tyr Gly Phe Tyr Asp His Gly Met Ile Ser Asn Leu Ser His Ile
 850 855 860

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Pro Ser Thr Cys Ser Ile Arg Ser Ser Thr Ser Gln Phe Asp Ala Glu
 865 870 875 880
 Asp Glu Trp Asp Glu Arg Asp Glu Glu Asp Gly Asp Phe Asp Asp Asp
 885 890 895
 Ser Asp Glu Asp Ser Asp Asp Ser Asp Ala Leu Phe Met Ser Val
 900 905 910
 Arg Lys His Ala Lys Ala Lys Ser Val Glu Ser Pro Leu Ser Glu Glu
 915 920 925
 Glu Glu Arg Leu Val Arg His Ile Glu Ala Glu Asp Gln Ala Val Glu
 930 935 940
 Ala Arg Val Ala Ala Gly Ile Val Ser Ser Asn Val Pro Asp Val Val
 945 950 955 960
 Ser Ser Asn Asp Ser Asp His Val Arg Ser Asp Thr Ser Thr Glu Asn
 965 970 975
 Lys Ser Phe Ser Arg Tyr Phe Asp Arg Thr Leu Ser Met Ala Ser Trp
 980 985 990
 Asp Asp Val Leu Ala Tyr Ile Tyr Arg Pro Lys Arg Ala Thr Val Pro
 995 1000 1005
 Asn Lys Arg Ser Ser Gly Ala Pro Pro Ser Val Arg Ser Thr Arg
 1010 1015 1020
 Ser Pro Leu Ser Asp His Pro Ile Thr Ser Ser Gly Asp Glu Ser
 1025 1030 1035
 Asp Arg Thr Ile Ser Ala His Ala Pro Ser Gly Gly Ala Gly Arg
 1040 1045 1050
 Gly Arg Ser His Ser Ser Ile Ser Arg Met Trp Arg Tyr Leu Lys
 1055 1060 1065
 Asn Ser Ser Ala Asp Glu Ala Thr Arg Ser Arg Ser Arg Asp Ala
 1070 1075 1080
 Asn Gly Ala Gly Ala Pro Pro Ala Tyr Glu Glu Ile Phe Pro Gly
 1085 1090 1095
 His Gly Val Val His Asp Lys Lys Val Val Gln Met Ala Ala Ala
 1100 1105 1110
 Ser Ala Ala Glu Asn Ser Ser Gly Pro Val Gly Ala Ser Ser Ser
 1115 1120 1125
 Ala Val Ala Ser Thr Ser Ala Ala Ala Ala Val Val Pro Ser Pro
 1130 1135 1140
 Leu Ala Pro Ile Val Glu Asp Glu Glu Gln Leu Val Glu Ala Trp
 1145 1150 1155
 Arg Arg Gln Arg Arg Ser Met Ala Asn Asp Arg Met Leu Phe Ala
 1160 1165 1170
 Phe Trp Leu Pro Val Leu Leu Met Ala Ile Gly Tyr Met Val Ile
 1175 1180 1185
 Lys Ala Phe Gly Leu Phe Pro Asp Gln Val Ser Ala Val Glu Ser
 1190 1195 1200
 Val Ala Glu Thr Val Gly Val His Cys Arg Gly Ala Val Ala Lys
 1205 1210 1215
 Leu Trp Phe Lys Gln Tyr Pro Val His Arg Gly Gln Pro Leu Lys
 1220 1225 1230
 Asp Thr Cys Ser Phe Glu Pro Asn Ser Leu Val Glu Ser Ala Leu
 1235 1240 1245
 Arg Gln Met Asn Gly Trp Ser Asp Arg Glu Val Pro Ile His Gln
 1250 1255 1260

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Ala Gln Ala Gln Ala Ala
1265

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<210> SEQ ID NO 53
<211> LENGTH: 3003
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 53

atggctaaag acaaggaaat cgacttgac tacacgggag aactggtgat ggacgattc      60
gagttcccca tcgacgacat gctccacaac gacggagatg actttgtcaa gaaggaaacg     120
tgggacgagg gtttggtt cggAACAAAT ggcccgtgg gtgcgcagat ggacgtccag     180
accAGCCAT ttagegaccc tggTTTGGC ggCGTGGGAG caggccctga catgtgggt     240
ctcatggata caaacatgaa ccacatcaac ggtagtcaca acatgaacag cgtcgtaaag   300
caggaggact actcacacacc gtccatgggc actccatgaa accccccaca gcaacagtcc   360
atgacccctc aacagcagca tcacatgaa cacaaccagg cctctcagct ccaatcttg   420
catcaacagt occagaaggc tcaaccacag cagcaacaac aacagccaca tcagtcgaca   480
ggagtgcata gcataatcac aaaggcatac accagggcag caggagact accgtacgga   540
cgaaagtact cacgacaact caacaagtac cccgaggacg tggagtattc atcttcgac   600
ccatcgctat ggagcaattt gctgaccaac tcggaaactc cgtaccaata ccagatacat   660
gtccattcca tgccggaaa atcacgtgt gagacccaaa tcaaattgtgc attatcaatc   720
taccctccgc ctccacagca gtccgttca cttccgacag acaccatttc gcgtcccaag   780
ttccagctca agcaggccca cattccagac tcgtgtctc ctttggaaatc atacatttg   840
ggcgagcaga accccagcaa gcccgtaat ttgtgttcta gatgcatcaa acgagaacag   900
aagcgagcct gtcgaaagaa actctttgac gagtcggagg agctgtcg agtcgagact   960
cgtaacgcac gtctggctgt cttaactgc tccgagggtgc ttgagttcaa ggatgtgaa   1020
cgccgagtagt acatccccga gtccggact acagttaccg ccaagcagct ggttctgcc   1080
ctgcgtctgg ctgtctactg tagacaccac ggggagaaaa agggatttcg aatcctctt   1140
tgtcttagag acgaggaggcc ccagattgtg ggtgtggcc agagtggAAC gaccgtcatg   1200
atcaactgacg accacaaggt tgtggagac ggggttgc tgcgtactac agccactgct   1260
ctggccaccc ctggctcttc acaacccccc acccagggttc ctacccccc tgcacatctcg   1320
tcgacgagct atcgctctcg aaactcgctt cctctatcgct ctacttccat ggaagactct   1380
tcgtcgaggt tcacctcgga ccattctcat tactccaact atggttctaa acgacgacga   1440
gacggcttcc ccatcagcga ttggagccgc atgatgaacg tgcgaggcat ggatagacag   1500
gcttccattt ccagcattcc cggAAATGGTT ggtggcatgt cgaacatgac tggccactgt   1560
gcttcgggta gcccactaa tctggctgtc cacaacatgaa acaacccccc agacgaaaac   1620
ctggccgtca tcaagcgaat catccccctcg cagggttcca ttcgaggccg cattgaagta   1680
accctgtttg gatctggctt caagtccat ctgggtggat ttttgggtga caacaaggcc   1740
gtgggcaccc actgtggtc tgattcgacc atcgtgaccc atctggccgc ttgcaccatc   1800
gtgggtcccg ttgtgggtgc ttgtggatgt tttgtgtcg acaagcctca gattttacc   1860
tattttgacg acacagacgg ccagttgatt gagttggcgc tccaggttgtt ggggtctcaag   1920
atgaacggac ggctgaaaga cggccgaaac attgcccattgc gaatcgtggg caacaatgga   1980

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| | | | | | | |
|-------------|--------------|-------------|-------------|-------------|-------------|------|
| ggcggttgcgg | gchgacacaagg | cggccatggca | ggcgaaaaaca | tgtctaacgg | agacgttggaa | 2040 |
| atggaaaatgt | ctgctgcaga | cagttcggtt | caacccgtat | cgcctccac | agaccacgaa | 2100 |
| gatgtggttc | tgcgatgtct | ggctctcaca | gacattcctg | gaggccgaat | tgccaactgg | 2160 |
| caactcacca | acgcccgggg | acagaccatg | gttcatctgg | ccagtattct | gggttactcg | 2220 |
| cgtgttctgg | tggctcttgt | ggctcgagga | gctcgtgtgg | atgtttccga | caatggtgga | 2280 |
| ttcactcctc | ttcatttcgc | tgctctcttt | ggccgtcgaa | agattgccaa | gaaactactt | 2340 |
| cggtgcaacg | ctgaccctta | caaacgtaac | cgaattggcg | aaaccgtgtt | tgtatgttgct | 2400 |
| tgtcctcaca | ttctcgatct | tctggtcggt | cctcaggcga | tgcctatggc | cgttcagacg | 2460 |
| tcgtatactc | ccgattacca | tcgtcagcgt | cgatcttcat | cttcttccac | tctggcttcc | 2520 |
| attgcatcca | tccaggattc | gcgtgagttac | ggtttctatg | accatggaaat | gatttccaaac | 2580 |
| ctgtcgcata | ttccgtccac | gtgctccatt | cgatcatcga | cttctcagg | tgacgctgaa | 2640 |
| gacgagtggg | acgagcggaa | tgaggaggat | ggagactttg | acgacgatc | agatgaggac | 2700 |
| tcagacatgt | actcagacgc | gcttcatgt | tctgttagaa | agcacgcacaa | ggccaagtct | 2760 |
| gtgaaatctc | ctctctctga | ggaggaagag | cgacttgtgc | gacacattga | ggccgaaagac | 2820 |
| caggctgtgg | aggcccgtgt | ggctgcccga | atcgtcagta | gcaatgtacc | cgacgtggtg | 2880 |
| tcttccaatg | actcggatca | cgtgagatct | gacacttcca | ctgagaacaa | gtcctttca | 2940 |
| cggtactttg | accgtactct | cagcatggca | tcttgggacg | atgttctggc | ttacatttac | 3000 |
| tga | | | | | | 3003 |

<210> SEQ_ID NO 54

<211> LENGTH: 1000

<212> TYPE: PRT

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Synthetic polypeptide

<400> SEQUENCE: 54

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Ala | Lys | Asp | Lys | Glu | Ile | Asp | Phe | Asp | Tyr | Thr | Gly | Glu | Leu | Val |
| 1 | | | | | | 5 | | | 10 | | | | | 15 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Asp | Asp | Phe | Glu | Phe | Pro | Ile | Asp | Asp | Met | Leu | His | Asn | Asp | Gly |
| | | | | | | 20 | | | 25 | | | | 30 | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Asp | Asp | Phe | Val | Lys | Lys | Glu | Thr | Trp | Asp | Glu | Gly | Phe | Gly | Phe | Gly |
| | | | | 35 | | 40 | | | | 45 | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Thr | Asn | Gly | Ala | Val | Gly | Ala | Gln | Met | Asp | Val | Gln | Thr | Ser | Pro | Phe |
| | | | | | | 50 | | | 55 | | | 60 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ser | Asp | Pro | Val | Phe | Gly | Gly | Val | Gly | Ala | Gly | Pro | Asp | Met | Met | Gly |
| | | | | | | 65 | | | 70 | | | 75 | | 80 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Met | Asp | Thr | Asn | Met | Asn | His | Ile | Asn | Gly | Ser | His | Asn | Met | Asn |
| | | | | | | 85 | | | 90 | | | 95 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ser | Val | Val | Lys | Gln | Glu | Asp | Tyr | Tyr | Thr | Pro | Ser | Met | Gly | Thr | Pro |
| | | | | | | 100 | | | 105 | | | 110 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|--|
| Met | Asn | Pro | Gln | Gln | Gln | Ser | Met | Thr | Pro | Gln | Gln | Gln | His | His | |
| | | | | | | 115 | | | 120 | | | 125 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Asn | His | Asn | Gln | Pro | Ser | Gln | Lys | Leu | Gln | Ser | Leu | His | Gln | Gln |
| | | | | | | 130 | | | 135 | | | 140 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|--|
| Gln | Lys | Ala | Gln | Pro | Gln | Gln | Gln | Gln | Gln | Pro | His | Gln | Ser | Thr | |
| | | | | | | 145 | | | 150 | | | 155 | | 160 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gly | Val | Asp | Ser | Ile | Ile | Thr | Lys | Ala | Tyr | Thr | Arg | Ala | Ala | Gly | Asp |
| | | | | | | 165 | | | 170 | | | 175 | | | |

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Leu Pro Tyr Gly Arg Lys Tyr Ser Arg Gln Leu Asn Lys Tyr Pro Glu
 180 185 190
 Asp Val Glu Tyr Ser Ser Phe Asp Pro Ser Leu Trp Ser Asn Leu Leu
 195 200 205
 Thr Asn Ser Glu Thr Pro Tyr Gln Tyr Gln Ile His Val His Ser Met
 210 215 220
 Pro Gly Lys Ser Arg Val Glu Thr Gln Ile Lys Cys Ala Leu Ser Ile
 225 230 235 240
 Tyr Pro Pro Pro Gln Gln Ser Val Arg Leu Pro Thr Asp Thr Ile
 245 250 255
 Ser Arg Pro Lys Phe Gln Leu Lys Gln Gly His Ile Pro Asp Ser Cys
 260 265 270
 Leu Ser Leu Glu Val Tyr Ile Val Gly Glu Gln Asn Pro Ser Lys Pro
 275 280 285
 Val Asn Leu Cys Ser Arg Cys Ile Lys Arg Glu Gln Lys Arg Ala Cys
 290 295 300
 Arg Lys Lys Leu Phe Asp Glu Ser Glu Glu Leu Ser Trp Val Glu Thr
 305 310 315 320
 Arg Gln Arg Arg Leu Ala Val Phe Asn Cys Ser Glu Val Leu Glu Phe
 325 330 335
 Lys Asp Val Glu Arg Arg Val Tyr Ile Pro Glu Ser Gly Thr Thr Val
 340 345 350
 Thr Ala Lys Gln Leu Val Leu Pro Leu Arg Leu Ala Cys Tyr Cys Arg
 355 360 365
 His His Gly Glu Lys Lys Gly Phe Arg Ile Leu Phe Cys Leu Arg Asp
 370 375 380
 Glu Gly Gly Gln Ile Val Gly Val Gly Gln Ser Gly Thr Thr Val Met
 385 390 395 400
 Ile Thr Asp Asp His Lys Val Val Gly Asp Ala Val Ala Met Pro Thr
 405 410 415
 Thr Ala Thr Ala Pro Ala Thr Ala Gly Ser Ser Gln Pro Pro Thr Gln
 420 425 430
 Val Pro Thr Pro Ala Ala Ser Ser Ser Thr Ser Tyr Arg Pro Arg Asn
 435 440 445
 Ser Leu Pro Leu Ser Pro Thr Ser Met Glu Asp Ser Ser Ser Glu Phe
 450 455 460
 Thr Ser Asp His Ser His Tyr Ser Asn Tyr Gly Ser Lys Arg Arg Arg
 465 470 475 480
 Asp Gly Ser Ser Ile Ser Asp Trp Ser Gly Met Met Asn Val Arg Gly
 485 490 495
 Met Asp Arg Gln Ala Ser Ile Thr Ser Ile Pro Glu Met Val Gly Gly
 500 505 510
 Met Ser Asn Met Thr Val Ala Ser Ala Ser Gly Ser Ala Thr Asn Leu
 515 520 525
 Ala Ala His Asn Met Asn Asn Pro Ala Asp Glu Asn Leu Pro Val Ile
 530 535 540
 Lys Arg Ile Ile Pro Ser Gln Gly Ser Ile Arg Gly Gly Ile Glu Val
 545 550 555 560
 Thr Leu Leu Gly Ser Gly Phe Lys Ser Asn Leu Val Ala Val Phe Gly
 565 570 575
 Asp Asn Lys Ala Val Gly Thr His Cys Trp Ser Asp Ser Thr Ile Val
 580 585 590
 Thr His Leu Pro Pro Ser Thr Ile Val Gly Pro Val Val Val Ser Phe

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231

232

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| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|------|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| 595 | 600 | 605 | | | | | | | | | | | | | |
| Glu | Gly | Phe | Val | Leu | Asp | Lys | Pro | Gln | Ile | Phe | Thr | Tyr | Phe | Asp | Asp |
| 610 | | | | | | 615 | | | | | | | 620 | | |
| Thr | Asp | Gly | Gln | Leu | Ile | Glu | Leu | Ala | Leu | Gln | Val | Val | Gly | Leu | Lys |
| 625 | | | | | | 630 | | | | | 635 | | | 640 | |
| Met | Asn | Gly | Arg | Leu | Glu | Asp | Ala | Arg | Asn | Ile | Ala | Met | Arg | Ile | Val |
| | 645 | | | | | | | | 650 | | | | 655 | | |
| Gly | Asn | Asn | Gly | Gly | Val | Ala | Gly | Ala | Gln | Gly | Ala | Met | Ala | Gly | Gly |
| | 660 | | | | | 665 | | | | | 670 | | | | |
| Asn | Met | Ser | Asn | Gly | Asp | Val | Gly | Met | Glu | Ser | Ala | Ala | Asp | Ser | |
| | 675 | | | | | 680 | | | | | 685 | | | | |
| Ser | Val | Gln | Pro | Val | Ser | Pro | Pro | Thr | Asp | His | Glu | Asp | Val | Val | Leu |
| | 690 | | | | | 695 | | | | | 700 | | | | |
| Arg | Cys | Leu | Ala | Leu | Thr | Asp | Ile | Pro | Gly | Gly | Arg | Ile | Ala | Asn | Trp |
| | 705 | | | | | 710 | | | | | 715 | | | 720 | |
| Gln | Leu | Thr | Asn | Ala | Glu | Gly | Gln | Thr | Met | Val | His | Leu | Ala | Ser | Ile |
| | 725 | | | | | 730 | | | | | 735 | | | | |
| Leu | Gly | Tyr | Ser | Arg | Val | Leu | Val | Ala | Leu | Val | Ala | Arg | Gly | Ala | Arg |
| | 740 | | | | | 745 | | | | | 750 | | | | |
| Val | Asp | Val | Ser | Asp | Asn | Gly | Gly | Phe | Thr | Pro | Leu | His | Phe | Ala | Ala |
| | 755 | | | | | 760 | | | | | 765 | | | | |
| Leu | Phe | Gly | Arg | Arg | Lys | Ile | Ala | Lys | Lys | Leu | Leu | Arg | Cys | Asn | Ala |
| | 770 | | | | | 775 | | | | | 780 | | | | |
| Asp | Pro | Tyr | Lys | Arg | Asn | Arg | Ile | Gly | Glu | Thr | Val | Phe | Asp | Val | Ala |
| | 785 | | | | | 790 | | | | | 795 | | | 800 | |
| Cys | Pro | His | Ile | Leu | Asp | Leu | Leu | Val | Gly | Pro | Gln | Gly | Met | Pro | Met |
| | 805 | | | | | 810 | | | | | 815 | | | | |
| Ala | Val | Gln | Thr | Ser | Tyr | Thr | Pro | Asp | Tyr | His | Arg | Gln | Arg | Arg | Ser |
| | 820 | | | | | 825 | | | | | 830 | | | | |
| Ser | Ser | Ser | Ser | Thr | Leu | Ala | Ser | Ile | Ala | Ser | Ile | Gln | Asp | Ser | Arg |
| | 835 | | | | | 840 | | | | | 845 | | | | |
| Glu | Tyr | Gly | Phe | Tyr | Asp | His | Gly | Met | Ile | Ser | Asn | Leu | Ser | His | Ile |
| | 850 | | | | | 855 | | | | | 860 | | | | |
| Pro | Ser | Thr | Cys | Ser | Ile | Arg | Ser | Ser | Thr | Ser | Gln | Phe | Asp | Ala | Glu |
| | 865 | | | | | 870 | | | | | 875 | | | 880 | |
| Asp | Glu | Trp | Asp | Glu | Arg | Asp | Glu | Asp | Gly | Asp | Phe | Asp | Asp | Asp | |
| | 885 | | | | | 890 | | | | | 895 | | | | |
| Ser | Asp | Glu | Asp | Ser | Asp | Asp | Ser | Asp | Ala | Leu | Phe | Met | Ser | Val | |
| | 900 | | | | | 905 | | | | | 910 | | | | |
| Arg | Lys | His | Ala | Lys | Ala | Lys | Ser | Val | Glu | Ser | Pro | Leu | Ser | Glu | Glu |
| | 915 | | | | | 920 | | | | | 925 | | | | |
| Glu | Glu | Arg | Leu | Val | Arg | His | Ile | Glu | Ala | Glu | Asp | Gln | Ala | Val | Glu |
| | 930 | | | | | 935 | | | | | 940 | | | | |
| Ala | Arg | Val | Ala | Ala | Gly | Ile | Val | Ser | Ser | Asn | Val | Pro | Asp | Val | Val |
| | 945 | | | | | 950 | | | | | 955 | | | 960 | |
| Ser | Ser | Asn | Asp | Ser | Asp | His | Val | Arg | Ser | Asp | Thr | Ser | Thr | Glu | Asn |
| | 965 | | | | | 970 | | | | | 975 | | | | |
| Lys | Ser | Phe | Ser | Arg | Tyr | Phe | Asp | Arg | Thr | Leu | Ser | Met | Ala | Ser | Trp |
| | 980 | | | | | 985 | | | | | 990 | | | | |
| Asp | Asp | Val | Leu | Ala | Tyr | Ile | Tyr | | | | | | | | |
| | 995 | | | | | 1000 | | | | | | | | | |

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<211> LENGTH: 772
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 55

| | | | | | | |
|--------------|-------------|------------|-------------|-------------|-------------|-----|
| atgtctggac | cttccacccct | cggcacggga | ctgcacccctc | tccccacaga | gaccggaaag | 60 |
| ttccccacca | acatcatgga | ccgattctcc | ctcaagggtt | agggtgcctc | cgtcaccggc | 120 |
| tcctcgtag | gtatcggtta | ctgcgtggcc | gaggcctacg | cccaggccgg | tgccgacgtg | 180 |
| gccccatctgtt | acaactccca | ccccggccac | gcaaaggctg | agcacccgc | taagacctac | 240 |
| ggcggtcaagg | ccaaggccctt | caagtgcctt | gtcaccggac | ccggccgcgt | ggagtccacc | 300 |
| atccagcaga | tcgagaagga | cattggcacc | attgacatct | tcgtcgccaa | cgctgggtgc | 360 |
| ccctggaccg | ccggccccat | gatcgacgtg | cccgacaaca | aggagtggga | caagggtcatc | 420 |
| aacctggatc | tcaacgggtc | ctactactgc | gccaagttacg | ccggccagat | cttcaagaag | 480 |
| aaggggcaagg | gatccttcat | cttcacccgc | tccatgtccg | gccacattgt | caacatcccc | 540 |
| cagatgcagg | cctgttacaa | cgccggcaag | gccgctctgc | tgcacctgtc | tcgatcgctg | 600 |
| gcccgtcgagt | ggggccggctt | tgcccgatgc | aacacagtct | ccccctggctt | catggccacc | 660 |
| gagatctccg | actttgtccc | caaggagacc | aaggagaagt | ggtggcagct | cattccatg | 720 |
| ggcccgagagg | gagacccctc | cgagctctag | cctacctcta | cattggctct | ga | 772 |

<210> SEQ ID NO 56
 <211> LENGTH: 200
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 56

| | | | | | | |
|--------------|--------------|--------------|------------|------------|-------------|-----|
| cacaaatatt | tttgattttac | tttgggttttgc | ccctattcgg | aaattttatt | gataatctaat | 60 |
| agaagtatta | aagtaaaaat | gtactaatac | ttaattgtaa | tgtcatcaga | aataacattt | 120 |
| gaggaaaata | tttcaaaccctt | aattgatata | tatattagag | atgtcccgct | tctctgtcat | 180 |
| taatatatattc | aagcaatcga | | | | | 200 |

<210> SEQ ID NO 57
 <211> LENGTH: 840
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 57

| | | | | | | |
|--------------|-------------|-------------|-------------|-------------|------------|-----|
| atgaagttca | cctccgttac | tcttcctcgcc | cttggccgccc | ttgtcggtgc | cgacaacgcc | 60 |
| gttgtctctc | agatcaacga | tggccagatc | caggctcttc | cggctgggtgg | tgagggtgcc | 120 |
| aaagccggccc | ctgctcccttc | tggagctgcc | cccggtgccc | ccgggtgtgg | tgctccggc | 180 |
| gtgtgggtctc | ccggcgctgg | tggccctggc | gctggcgagg | gtgctaagcc | ctctggagct | 240 |
| gccccccgggtt | ccccccggcc | tggtgctccc | ggtgctgggtg | agggtgttaa | gccttctggc | 300 |
| ggtgcccccg | gtgctggcgc | tcctggtgct | ggcgagggtg | ctaagccctc | tggtggtgcc | 360 |
| cctgggtcccc | ccggcgctgg | tgtcccccgt | gctgggtgagg | gtgctaagcc | ctctgggtgt | 420 |
| gccccccgggtt | ccccccggcc | tggtgagggt | gccaaggccct | ccggctctgc | tcccggtgt | 480 |
| cctggcgctg | gtgagggtgc | caagccctcc | ggctctgtctc | ccgggtgtcc | tggtggctgt | 540 |

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| | |
|--|-----|
| gagggtgcca agccctctgg ctctgctccc ggtgctcctg gtgctggta gggtgccaag | 600 |
| ccctctggct ctgctcccg tgctcctgga gctggtcaagcc ctccgctgga | 660 |
| ggtgagcacc ccgctgctga ggccactggt gtcgtcaactc agatccacga cggccagatc | 720 |
| caggctcccg agcagaccca gccccccgt gccggccctg cccaggctaa cggtgctgcc | 780 |
| accctcggtg cccagatcgt tgccgggttt gtcggcgctg cgggtgtcgc tctttctaa | 840 |

<210> SEQ ID NO 58
<211> LENGTH: 1542
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 58

| | |
|--|------|
| atggccgaca acaagcctct gtgcacgatt accacgcccc acccgtaacc caagcgta | 60 |
| aagatctctg ccgaggagaa agaaaagatg cgacttgaaa aggaacagat caagaaggcag | 120 |
| aaagaggaag agcgagagca gcttcgaaga cagaaggaag aagagaaaga gctactgaga | 180 |
| aagcagaaag aggaggagaa ggaacaactg aggaaacaga aggaggagga gaagagggct | 240 |
| aaagaggagg agagagggct agagagaggg agaaaaacgac gacgagaaga ggaacgaaag | 300 |
| aaggctgccc aagagaagga gcttgagcga gccaagattg cagaggagaa ggctaagtt | 360 |
| gctgaagaga aggaggccaa gagacttcaa aaagaagctg aactcaagaa gaaggagcaa | 420 |
| gaacagactc gaatcatgtc tttctttaac aagaagacca aaaagaagac caagaaggaa | 480 |
| gctgttaaca gtgacaagtg tttggacttt gataaagact tcctaccctt ccacatcaaa | 540 |
| gataccgtgt gtatggcaga caagacggag tctgaagtga tggatcagga tcctgttgac | 600 |
| tggctcaaca gtctcaacct ttctgtatgc agcaacaccc cggaaacgaga agaaccacct | 660 |
| gttcccgta aaaccatcat tactcacatc cagaccgctg ccactctggg tctcaatcct | 720 |
| gataattaca acggtaactcc ttttagacacg ctggtaatg ctcttcctag acgataactt | 780 |
| cagttctatg gtgaegagcg accegcatac ctgggcacgt actccaagag ctgctcgct | 840 |
| gatctgttgc agaaccctct cttccaggtg cttgggttgg actacgagta cgacagttag | 900 |
| gcagactggg aagatgaagg agaagatatt gaagatgtg aaattagtgg agacgaggag | 960 |
| atggaggacg acgaaatggc cgactttgtg tttctgtatg atgccaagag tcccagcacc | 1020 |
| atgacttcaa aggtcacgac agcccaggaa cttgttgg tctggggctg ctcagatatg | 1080 |
| gttggatga cttttggagg actgattgtc cagggggcaa ttgaccatt caaagactat | 1140 |
| tggactgttgc caaaagttga gcagaagacc gatactaaga gtgacgtgac aatgactatg | 1200 |
| cgacatcg cttctggta acgtattaaa tctactacaa caaaaaccga actcagcccg | 1260 |
| tttgaagtcc tctccaaaac tctgtcacct tccccagcgg ttgcttcagc cacgaaacag | 1320 |
| tttctggctg ctgccaagcc tcagaagctc attgctggag acgacctgac tgcttttg | 1380 |
| aagcgagtag atggatccga cgataacaag acgctgttga ccgagctgct ttgtaagcag | 1440 |
| tatccccagt acacacgaa gatggtcacg gccaccattc agcaactatgc tgagcgacag | 1500 |
| ggtcctaaga gcgacaagcg gtgggttctg aaggatatct ag | 1542 |

<210> SEQ ID NO 59
<211> LENGTH: 1944
<212> TYPE: DNA
<213> ORGANISM: Yarrowia lipolytica

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<400> SEQUENCE: 59

| | |
|--|------|
| atgagcttc cccaacaagt aatagcgccg ggccaaacggc tcaacgagct tctggaggcc | 60 |
| atcaaacagg agttagactc cgtgaccaac gaggcgctcg tctaccggct gcacaaggac | 120 |
| gagtttgacg tcaaggtaa ccagcagacg tcagatctgg gccagattcg acagtcggc | 180 |
| tacgagctag aaatggcgca ccgaaagatg aaggagcgct acgaggaggaa aatcatgcgg | 240 |
| ctcaagagcg agctggaggc ccgaggtgga cccgctgca accccgcaca ctcccagcag | 300 |
| cagcaacagc agcaacagca acagcagcaa cagcagcagc agaaccagca ggcacaggac | 360 |
| caacaagcac gggccgcga acaacaggca gcccagcagc aggccctcgc ccagcagcag | 420 |
| gccgcccagc agcaggctct ggcccaacagc caggcccagg ctcaacagca ggcccaggcc | 480 |
| caggcccacc acatgggtgg tggccccct tcgcaaggac agccccctgc gctgtcgct | 540 |
| ccatcatcca acgtgttcag cggcatcatg tccggtcagc cggcacccctc ttctctggct | 600 |
| cccccgcagg gacagcccg tagccccccag cctggtcagc ccaacacctgg tcaaccccgag | 660 |
| ccctactccg gctacgtggg tgctaaccgc tacacgtctt cgccacataa cggacccccc | 720 |
| gtcatcagcg caatggcctc gcccaacagc aagaagcgac aggtgtcgac ccccggttccc | 780 |
| ggcaaggcgt ctcccccagg gtggcccccgg gagatgcaac agcagcagca acagcaggc | 840 |
| cctccacagc agcagcaacc tccccagcag cagcaacaga gccccgaaga gatggcaac | 900 |
| tacctggcgcc acatggacat tgagcgggta cctccggcgc taaaaaaaaca aaaggccgac | 960 |
| tgggggtcg tttacaacca gcgagcacca cggctgtgg acgtggatat tggcagtcg | 1020 |
| ctggaccaca actctgttagt gtgctgtgtg cggttctccg ctgacggcaa gtacattgccc | 1080 |
| actggctgta accgatctgc ccagatttc gacgtgcaga ctggccagct catctgcgg | 1140 |
| ctgcaggacg actcggcgtcga ccgagaaggc gacctgtaca tccggccgt gtgttctcg | 1200 |
| ccggacggta agtacctggc caccggcgcc gaggacaagc agatccgagt gtggacatt | 1260 |
| aaatctcaga gcatacggca cgtgttcaact ggcacggcagg aggacattta ctgcgtggac | 1320 |
| tttgcgaa acggccgaca cattgcctct ggctctggcg accgcacagt ccgaatgtgg | 1380 |
| gatattgaga gcgccagtg tactctaacc ctgtcgatcg aggacggcgt caccacggtg | 1440 |
| gcacatctcgcc cgacggcaa gtttgtggct gcaggcagct tggacaagtc tgtgcaatc | 1500 |
| tgggacacctt ctaccggggtt cctgggttag cgtctggagg cccctgtatgg acacaaggac | 1560 |
| tccgtctata gtgtagctt caccccaac ggtatggatc ttgttccgg ctgcgtggac | 1620 |
| aagacgatca agctgtggga gctgcaggct cctcgaggca ttcaaggccaa ccagcagga | 1680 |
| ggcgtctgcg tcaagacgct gtgtggacac aaggactttt ttctgagtg gtggacatt | 1740 |
| ctggatggcc agtggattct ttccggctcc aaggacgggg gtgtcaatt ctggacccct | 1800 |
| cgaacgggcc aggtgcaact catgtgcag ggtcatcgaa attcggtcat cagtgtggct | 1860 |
| cctagtcaca tgggggggtt gtttgcactt ggaagtggag attgcaaggc tcgaatctgg | 1920 |
| cgataacttcc ctgtcaacag ataa | 1944 |

<210> SEQ_ID NO 60

<211> LENGTH: 647

<212> TYPE: PRT

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 60

| | | | |
|---|---|----|----|
| Met Ser Phe Pro Gln Gln Val Ile Ala Pro Gly Gln Arg Leu Asn Glu | | | |
| 1 | 5 | 10 | 15 |

-continued

Leu Leu Glu Ala Ile Lys Gln Glu Phe Asp Ser Val Thr Asn Glu Ala
 20 25 30
 Ser Val Tyr Arg Leu His Lys Asp Glu Phe Asp Val Lys Val Asn Gln
 35 40 45
 Gln Thr Ser Asp Leu Gly Gln Ile Arg Gln Ser Val Tyr Glu Leu Glu
 50 55 60
 Met Ala His Arg Lys Met Lys Glu Arg Tyr Glu Glu Ile Met Arg
 65 70 75 80
 Leu Lys Ser Glu Leu Glu Ala Arg Gly Gly Pro Ala Ala Asn Pro Ala
 85 90 95
 His Ser Gln
 100 105 110
 Gln Gln Asn Gln Gln Ala Gln Asp Gln Gln Ala Arg Ala Ala Gln Gln
 115 120 125
 Gln Ala Ala Gln Gln Gln Ala Leu Ala Gln Gln Ala Ala Gln Gln
 130 135 140
 Gln Ala Leu Ala Gln Gln Gln Ala Gln Gln Gln Ala Gln Ala
 145 150 155 160
 Gln Ala His His Met Gly Gly Val Pro Pro Ser Gln Gly Gln Pro Pro
 165 170 175
 Ser Leu Leu Arg Pro Ser Ser Asn Val Phe Ser Gly Ile Met Ser Gly
 180 185 190
 Gln Pro Gly Thr Ser Ser Leu Ala Pro Pro Gln Gly Gln Pro Gly Gln
 195 200 205
 Pro Gln Pro Gly Gln Pro Gln Pro Gly Gln Pro Gln Pro Tyr Ser Gly
 210 215 220
 Tyr Val Gly Ala Asn Gly Tyr Thr Ser Ser Pro His Asn Gly Pro Pro
 225 230 235 240
 Val Ile Ser Ala Met Ala Ser Pro Asn Ser Lys Lys Arg Gln Val Ser
 245 250 255
 Thr Pro Val Pro Gly Lys Ala Ser Pro Gln Val Ala Pro Gln Glu Met
 260 265 270
 Gln Gln Gln Gln Gln Gln Gly Pro Pro Gln Gln Gln Gln Pro Pro
 275 280 285
 Gln Gln Gln Gln Ser Pro Glu Glu Met Gly Asn Tyr Leu Gly Asp
 290 295 300
 Met Asp Ile Glu Arg Val Pro Pro Glu Leu Lys Gln Lys Ala Asp
 305 310 315 320
 Trp Phe Val Val Tyr Asn Gln Arg Ala Pro Arg Leu Leu Asp Val Asp
 325 330 335
 Ile Val Gln Ser Leu Asp His Asn Ser Val Val Cys Cys Val Arg Phe
 340 345 350
 Ser Ala Asp Gly Lys Tyr Ile Ala Thr Gly Cys Asn Arg Ser Ala Gln
 355 360 365
 Ile Phe Asp Val Gln Thr Gly Gln Leu Ile Cys Arg Leu Gln Asp Asp
 370 375 380
 Ser Val Asp Arg Glu Gly Asp Leu Tyr Ile Arg Ser Val Cys Phe Ser
 385 390 395 400
 Pro Asp Gly Lys Tyr Leu Ala Thr Gly Ala Glu Asp Lys Gln Ile Arg
 405 410 415
 Val Trp Asp Ile Lys Ser Gln Ser Ile Arg His Val Phe Thr Gly His
 420 425 430
 Glu Gln Asp Ile Tyr Ser Leu Asp Phe Ser Arg Asn Gly Arg His Ile

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435 440 445

Ala Ser Gly Ser Gly Asp Arg Thr Val Arg Met Trp Asp Ile Glu Ser
 450 455 460

Gly Gln Cys Thr Leu Thr Leu Ser Ile Glu Asp Gly Val Thr Thr Val
 465 470 475 480

Ala Ile Ser Pro Asp Gly Lys Phe Val Ala Ala Gly Ser Leu Asp Lys
 485 490 495

Ser Val Arg Ile Trp Asp Thr Ser Thr Gly Phe Leu Val Glu Arg Leu
 500 505 510

Glu Ala Pro Asp Gly His Lys Asp Ser Val Tyr Ser Val Ala Phe Thr
 515 520 525

Pro Asn Gly Met Asp Leu Val Ser Gly Ser Leu Asp Lys Thr Ile Lys
 530 535 540

Leu Trp Glu Leu Gln Ala Pro Arg Gly Ile Gln Ala Asn Gln Arg Gly
 545 550 555 560

Gly Val Cys Val Lys Thr Leu Cys Gly His Lys Asp Phe Val Leu Ser
 565 570 575

Val Ala Ser Thr Leu Asp Gly Gln Trp Ile Leu Ser Gly Ser Lys Asp
 580 585 590

Arg Gly Val Gln Phe Trp Asp Pro Arg Thr Gly Gln Val Gln Leu Met
 595 600 605

Leu Gln Gly His Arg Asn Ser Val Ile Ser Val Ala Pro Ser Pro Met
 610 615 620

Gly Gly Leu Phe Ala Thr Gly Ser Gly Asp Cys Lys Ala Arg Ile Trp
 625 630 635 640

Arg Tyr Phe Pro Val Asn Arg
 645

<210> SEQ ID NO 61

<211> LENGTH: 900

<212> TYPE: DNA

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 61

```

atgtcttatca agcgagaaga gtccttact cccacccccc aggacctggg atctccctg 60
acagctgatt ctccctggctc tccccagtct ggagacaagg gaaagaagga tctcactctg 120
ccccttccctg ctgggtctct tccccctcga aagagagcta agacagagaa cgaaaaggag 180
cagagacgc tcgagcggat catgcgaaac cggcaggcgg cacatgcgtc tcgagagaag 240
aagcgacgac atttgagga cctggagaag aagtgcgtgg agttgtcgac cgaaaacaac 300
gatctacacc accagggtgac tgagtccaag aagaccaaca tgcacccat ggaacaacac 360
tactcgctgg tggccaagct gcacgcgtc tgcgtcgatc tcaacatggc caagtcttcc 420
ggagctttgg ccggcggttga tgtcccccac atgagcgatc tgtctatggc ccccaagttg 480
gagatgccccca ccggcggttcc ttcccagccc atgggtctcg ccagcgcc caccctttc 540
aaccacgata atgagaccgt cgtccccgac tctccttattt tgaagaccga ggaagtgcac 600
tctacaaact ttctccatca cacggaggcc tctccccccccc cccaaacttgc tgagagact 660
ggctcaggct cgccatcgatc gactctgtcc tggacgaaat ctgattatct tggacccgg 720
gcgcgtcatc cagcgtatc gactgtcgca actactgacc agcagcgatc gcacaagatt 780
tcattttcat caaggacgag cccgttgcacg acgagcttgg actgcgttgc ctgtcgatc 840
acttcaccct gtttgaagac aacaaggcagc ctgcccagca cgactttattt gctgtatct 900

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<210> SEQ ID NO 62
<211> LENGTH: 299
<212> TYPE: PRT
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 62

```

Met Ser Ile Lys Arg Glu Glu Ser Phe Thr Pro Thr Pro Glu Asp Leu
1           5          10          15

Gly Ser Pro Leu Thr Ala Asp Ser Pro Gly Ser Pro Glu Ser Gly Asp
20          25          30

Lys Arg Lys Lys Asp Leu Thr Leu Pro Leu Pro Ala Gly Ala Leu Pro
35          40          45

Pro Arg Lys Arg Ala Lys Thr Glu Asn Glu Lys Glu Gln Arg Arg Ile
50          55          60

Glu Arg Ile Met Arg Asn Arg Gln Ala Ala His Ala Ser Arg Glu Lys
65          70          75          80

Lys Arg Arg His Leu Glu Asp Leu Glu Lys Lys Cys Ser Glu Leu Ser
85          90          95

Ser Glu Asn Asn Asp Leu His His Gln Val Thr Glu Ser Lys Lys Thr
100         105         110

Asn Met His Leu Met Glu Gln His Tyr Ser Leu Val Ala Lys Leu Gln
115         120         125

Gln Leu Ser Ser Leu Val Asn Met Ala Lys Ser Ser Gly Ala Leu Ala
130         135         140

Gly Val Asp Val Pro Asp Met Ser Asp Val Ser Met Ala Pro Lys Leu
145         150         155         160

Glu Met Pro Thr Ala Ala Pro Ser Gln Pro Met Gly Leu Ala Ser Ala
165         170         175

Pro Thr Leu Phe Asn His Asp Asn Glu Thr Val Val Pro Asp Ser Pro
180         185         190

Ile Val Lys Thr Glu Glu Val Asp Ser Thr Asn Phe Leu Leu His Thr
195         200         205

Glu Ser Ser Ser Pro Pro Glu Leu Ala Glu Ser Thr Gly Ser Gly Ser
210         215         220

Pro Ser Ser Thr Leu Ser Cys Asp Glu Thr Asp Tyr Leu Val Asp Arg
225         230         235         240

Ala Arg His Pro Ala Val Met Thr Val Ala Thr Thr Asp Gln Gln Arg
245         250         255

Arg His Lys Ile Ser Phe Ser Ser Arg Thr Ser Pro Leu Thr Thr Ser
260         265         270

Leu Asp Cys Met Asp Cys Arg Met Thr Ser Pro Cys Leu Lys Thr Thr
275         280         285

Ser Ser Leu Pro Ser Thr Thr Leu Leu Leu Ile
290         295

```

<210> SEQ ID NO 63
<211> LENGTH: 738
<212> TYPE: DNA
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 63

```

atgcgccaaa agctgcccgtt caacccgctc cagtcgcttc tccccgcaat ctttgtcg 60
ggcaaaaaac acgatgcgcg cagccgctgg gaaatgcgcc agatgaaaga caagcatgt 120
gccccatggcca aggctgacgg attccggctcg cgagccgcgt acaagctaca ggaactcgac 180

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| | |
|--|-----|
| tccatgttcc ggctgttcaa gcccggcatg acgggtggtg | 240 |
| atttgggctt tgccgcggc | |
| gcatggagtc aagtggctgc tcagcgagtg cggcctggag gcagagttat tggagtggat | 300 |
| atccttcctt gcatttcctcc tccaggagtg tccagcatcc agggaaattt cctgtccaaa | 360 |
| gaaaacacaaa acgagctaa acgtgtgtcg gccgtctcg cgatggagt tcccaaggac | 420 |
| aaggactctg gtggcgccat aggactgtct cctccgtctt atctggacac tgaacgcgag | 480 |
| cttggcagta ttaacagcaa cagcaacgaa ccccaatttg ggcacgacta cccggtagat | 540 |
| atagtgctta gtgacatgtg cgaaacgtta ccccaggAAC acggatttt tcaaagaact | 600 |
| attaatgacc catactatag gatggccaat gtttccggca tagctgtgag ggaccatgt | 660 |
| gccagtttg tgagtgaagg aaggaagcgc attgggtgtg gtgcagccag ctgcgtatgt | 720 |
| gcagaaggaa agccataa | 738 |

<210> SEQ ID NO 64

<211> LENGTH: 245

<212> TYPE: PRT

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 64

| | |
|---|--|
| Met Arg Gln Lys Leu Pro Phe Asn Pro Leu Gln Ser Leu Leu Pro Arg | |
| 1 5 10 15 | |

| | |
|---|--|
| Ile Phe Val Arg Gly Lys Lys His Asp Ala Arg Ser Arg Trp Glu Met | |
| 20 25 30 | |

| | |
|---|--|
| Arg Gln Met Lys Asp Lys His Val Ala Met Ala Lys Ala Asp Gly Phe | |
| 35 40 45 | |

| | |
|---|--|
| Arg Ser Arg Ala Ala Tyr Lys Leu Gln Glu Leu Asp Ser Met Phe Arg | |
| 50 55 60 | |

| | |
|---|--|
| Leu Phe Lys Pro Gly Met Thr Val Val Asp Leu Gly Phe Ala Pro Gly | |
| 65 70 75 80 | |

| | |
|---|--|
| Ala Trp Ser Gln Val Ala Ala Gln Arg Val Arg Pro Gly Gly Arg Val | |
| 85 90 95 | |

| | |
|---|--|
| Ile Gly Val Asp Ile Leu Pro Cys Ile Pro Pro Pro Gly Val Ser Ser | |
| 100 105 110 | |

| | |
|---|--|
| Ile Gln Gly Asn Phe Leu Ser Lys Glu Thr Gln Asn Glu Leu Lys Arg | |
| 115 120 125 | |

| | |
|---|--|
| Val Leu Ala Val Ser Ala Met Gly Val Pro Lys Asp Lys Asp Ser Gly | |
| 130 135 140 | |

| | |
|---|--|
| Gly Ala Ile Gly Thr Ala Pro Pro Ser Tyr Leu Asp Thr Glu Arg Glu | |
| 145 150 155 160 | |

| | |
|---|--|
| Leu Gly Ser Ile Asn Ser Asn Ser Asn Glu Pro Gln Phe Gly Asp Asp | |
| 165 170 175 | |

| | |
|---|--|
| Tyr Pro Val Asp Ile Val Leu Ser Asp Met Cys Glu Thr Leu Pro Gln | |
| 180 185 190 | |

| | |
|---|--|
| Glu His Gly Phe Phe Gln Arg Thr Ile Asn Asp Pro Tyr Tyr Arg Met | |
| 195 200 205 | |

| | |
|---|--|
| Ala Asn Val Ser Gly Ile Ala Val Arg Asp His Ala Ala Ser Ile Val | |
| 210 215 220 | |

| | |
|---|--|
| Ser Glu Gly Arg Lys Arg Ile Gly Cys Gly Ala Ala Ser Phe Asp Val | |
| 225 230 235 240 | |

| | |
|---------------------|--|
| Ala Glu Gly Lys Pro | |
| 245 | |

<210> SEQ ID NO 65

<211> LENGTH: 1590

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<212> TYPE: DNA
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 65

```

atgtttaca ccaagcccga cccgggtggtt gattattccc gcctcaagga catggacatg      60
tacccctgagt acgacaatgg ccagaacatg ggctttcca acatgaacat gaccgatctt      120
tacgacggcg gtcttaacat gtcgtcgatg gcgcaccccg tggcggtgaa ccagatggc      180
agcatgggcc ccatgggctc tttaagtaac atgccccatgg gttttgtgtc ccagaaccag      240
cctcaaactc aggctcaggc ccaggcccag agccagaacc agaatcagaa ccagaaccag      300
aaccagaacc agcctcagaa tcacaacacc catgttatga gcgataacca caaccatacc      360
cacaccaaca atactcacaa caccaacgtc acccacaaca cccccctccat gggtggtcac      420
acaacacctg tcgggggcca cgacaccaat gactcggccc atgttggggg tcacgcccac      480
aatgtcacat cccccacccc ggcaacccct gcctccacat cttccgtacc cgcaaccccg      540
cctcagattc ctttcacggc cgccgcaccc gcaccgtca gcaaatatgt gaccgatgac      600
gagcgatggc aggcaactggt cgaccgagac cccgagggtg acggcgcctt catctactgc      660
gtcaccaggca ccaagggtgtc ctgcggccc acgtgctcggt cccggctcgc gctgggtcc      720
aacatttgtt attttgacac catgaaggag gctgtggccg cccgctacccg cccctgcgca      780
cggtgcaacc ccgacgtgag cgagatgaac tgcgcgegcac ggcggctggg ctccgtgtt      840
aacctcatcc actcgctgga gcccgacaag gtgccacgtg tcaagaagct agccgagttc      900
gtcggectca cgctctggca cttdcacggt ctcttcaagc ggtacacggg cctcacgcct      960
cgacagtaca tcaactgagtt ccacaagcga aagcgccttgg ggtgeccgca gttgeagtc      1020
agcaagggtt taaccaagaa gagctatgag cgacagcgcgt gtcgcccaggc cagcaacggc      1080
tccacgcccc agcagtctcc ccaagtcggc gcctcttcgc cagccggcga ggtggaggcc      1140
atcaagctcg agaccccggt cgaaaccgtc cagccgtat actacgacag caacggcgtt      1200
actcacaacg ctgccaacgt cggggctcac agtccaaatg tcaactcacaa cactagccat      1260
gtcgggaagca acgcaacccctc cgccacgcgc tccattgcca ctcccttttc caacacaacg      1320
tcaacccgaca cctcgacgccc ggcccaggac tccggataaca tcattgcca cggttccaa      1380
gcacagcaacg cccgctctgt ggttgctcg gggcctgcca cccgctctgg cgacaactgg      1440
atcaagacgg agccctcgat ggattttatg cctcggtacg agccgeggta cgaccagtct      1500
atctccattg acgcccccat gtttattctt gatggtaacg agtatacatca caacggggag      1560
atgttgggtg acatgtgggg gactctctaa                                         1590

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<210> SEQ ID NO 66
<211> LENGTH: 529
<212> TYPE: PRT
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 66

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Phe | Tyr | Thr | Lys | Pro | Asp | Pro | Val | Val | Asp | Tyr | Ser | Arg | Leu | Lys |
| 1 | | | | | 5 | | | 10 | | | 15 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Asp | Met | Asp | Met | Tyr | Pro | Glu | Tyr | Asp | Asn | Gly | Gln | Asn | Met | Gly | Phe |
| | | | 20 | | | | | 25 | | | | | 30 | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ser | Asn | Met | Asn | Met | Thr | Asp | Leu | Tyr | Asp | Gly | Gly | Leu | Asn | Met | Ser |
| | | | | | 35 | | | | 40 | | | 45 | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ser | Met | Ala | Gln | Pro | Val | Ala | Leu | Asn | Gln | Met | Gly | Ser | Met | Gly | Pro |
| | | | | 50 | | | | 55 | | | 60 | | | | |

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Met Gly Ser Leu Ser Asn Met Pro Met Gly Phe Val Ser Gln Asn Gln
 65 70 75 80

Pro Gln Thr Gln Ala Gln Ala Gln Ser Gln Asn Gln Asn Gln
 85 90 95

Asn Gln Asn Gln Asn Gln Pro Gln Asn His Asn Thr His Val
 100 105 110

Met Ser Asp Asn His Asn His Thr His Asn Asn Thr His Asn Thr
 115 120 125

Asn Val Thr His Asn Thr Pro Ser Met Gly Gly His Thr Thr Ser Val
 130 135 140

Gly Gly His Asp Thr Asn Asp Ser Ala His Val Gly Gly His Ala Ser
 145 150 155 160

Asn Val Thr Ser Pro Thr Pro Ala Thr Pro Ala Ser Thr Ser Val
 165 170 175

Pro Ala Thr Ser Pro Gln Ile Pro Phe Thr Val Ala Pro Pro Ala Pro
 180 185 190

Ser Gly Lys Tyr Val Thr Asp Asp Glu Arg Trp Gln Ala Leu Val Asp
 195 200 205

Arg Asp Pro Glu Ala Asp Gly Ala Phe Ile Tyr Cys Val Thr Ser Thr
 210 215 220

Lys Val Tyr Cys Arg Pro Thr Cys Ser Ala Arg Leu Ala Leu Arg Ser
 225 230 235 240

Asn Ile Val Tyr Phe Asp Thr Met Lys Glu Ala Val Ala Ala Gly Tyr
 245 250 255

Arg Pro Cys Arg Arg Cys Asn Pro Asp Val Ser Glu Met Asn Ser Gln
 260 265 270

Arg Arg Ala Val Gly Ser Val Cys Asn Leu Ile His Ser Leu Glu Pro
 275 280 285

Asp Lys Val Pro Arg Val Lys Lys Leu Ala Glu Ser Val Gly Leu Thr
 290 295 300

Leu Trp His Phe His Arg Leu Phe Lys Arg Tyr Thr Gly Leu Thr Pro
 305 310 315 320

Arg Gln Tyr Ile Thr Glu Phe His Lys Arg Lys Arg Leu Gly Leu Pro
 325 330 335

Gln Leu Gln Val Ser Lys Val Val Thr Lys Lys Ser Tyr Glu Arg Gln
 340 345 350

Gln Arg Arg Gln Gly Ser Asn Gly Ser Thr Pro Gln Gln Ser Pro Gln
 355 360 365

Val Gly Ala Ser Ser Pro Ala Gly Glu Val Glu Ala Ile Lys Leu Glu
 370 375 380

Thr Pro Val Glu Thr Val Gln Pro Leu Tyr Tyr Asp Ser Asn Gly Val
 385 390 395 400

Thr His Asn Ala Ala Asn Val Gly Ala His Ser Ser Asn Val Thr His
 405 410 415

Asn Thr Ser His Val Gly Ser Asn Ala Thr Ser Ala Thr Ser Ser Ile
 420 425 430

Ala Thr Pro Leu Ser Asn Thr Thr Ser Pro Asp Thr Ser Thr Pro Ala
 435 440 445

Gln Asp Ser Ala Tyr Ile Ala His Gly Ser Asn Ala Ser Asn Ala
 450 455 460

Ala Pro Val Val Ala Pro Gly Pro Ala Thr Gly Ser Gly Asp Asn Trp
 465 470 475 480

Ile Lys Thr Glu Pro Ser Met Asp Phe Met Pro Arg Tyr Glu Pro Arg

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| | | |
|-----|-----|-----|
| 485 | 490 | 495 |
|-----|-----|-----|

Tyr Asp Gln Ser Ile Ser Ile Asp Ala Pro Met Phe Ile Pro Asp Gly
500 505 510

Asn Glu Tyr His His Asn Gly Glu Met Leu Gly Asp Met Trp Gly Thr
515 520 525

Leu

<210> SEQ ID NO 67

<211> LENGTH: 1709

<212> TYPE: DNA

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 67

| | |
|---|------|
| atgatttctg ctattcgccc cggccgttca tcttccgttc gtgttgcccc tatggccaaac | 60 |
| accgccttcc gggccctactc tacccaggat gtgagttttt ctttttttc atcaatttgt | 120 |
| tgcgtgtcgca cggatttcgt tgcgctcagcc tgattgcaac agccttaggc cccatttcg | 180 |
| acctgttctt gcctcggcaa aagttttcc gaatgcattgt gacacgtcga atgtgggtct | 240 |
| ttcaaggcgc acgagcagca taaaatatgg aatgtgttgt gtgcagaagt cgacattaca | 300 |
| taaccccgcg gcaaccatac gagatggcag tcataacaat tgcaatttag caatacaaac | 360 |
| cacactgcaa cccactaaaa agaaaacacga ctaacaaata gggctttaag gagcgattcg | 420 |
| ccgagctcat ccccgagaac gtcgagaaga tcaagaagct ccgaaaggag aagggttaaca | 480 |
| ccgtcatcg cgaggtcatc ctcgaccagg cttacgggtg tatgcgaggt attaagggtc | 540 |
| tgcgtctggaa gggatccgtc ctcgaccccg aggagggtat ccgattccga ggtctgacta | 600 |
| tcccccaccc ccagaagcag ctccccacg cccctggcg aaaggagcct ctcccgagg | 660 |
| gtctttctg gtcctgttc accggcgaga tccccactga tgctcaggc aagggtctgt | 720 |
| ccgctgactg ggcctctcgca gccgagatcc ccaagcatgt tgaggagctc atcgaccgt | 780 |
| gccccccac cctccacccc atggctcage tcggatttgc cgtcaacgct ctggagtccg | 840 |
| agtctcagtt caccaaggct tacgagaagg gtgttaacaa gaaggagtagc tggcagtaca | 900 |
| cctacgagga ttccatgaac ctcattgcca agctccccgt cattgtttct cgaatctacc | 960 |
| gaaacctttt caaggacgga aagattttt gtcatttgc caactcttt gactactctg | 1020 |
| ctaacttcgc ctctctgtc ggcttggcg acaacaaggaa gttcatttgc cttctgac | 1080 |
| tctacctcac catccacgct gaccacgagg gagtaacgt ctctgeccac accaccaagc | 1140 |
| ttgttggttc tgctctctcc tctcccttcc tctctctgtc cgctgggttc aacggcttgc | 1200 |
| ccggctctct ccacggccga gctaaccagg aggtcatttgc gtggatttgc gagatgaagt | 1260 |
| ccaagatgg ctctgtatgtc accaaggagg acattgagaa gtacctctgg gataccctta | 1320 |
| aggccggtcg agtcgtcccc ggtaacggac acggccgttcc cggaaagacc gatcctcgat | 1380 |
| acaccggcca gcgagatgtc gccctcgac acatggccga ctacgaccctc ttccacccgt | 1440 |
| tttccaccat ctacgaggtt gcccccaagg ttctcaccga gcacggcaag accaagaacc | 1500 |
| ctggcccaa tgtggactcc cactccgggtg tcttcctccca gtactacggt ctcactgagc | 1560 |
| agtcttacta cactgttctc ttccgggttt cccgagctat cgggtgtccctg ccccgactca | 1620 |
| tcatggaccg agcttacgggt gctcccatcg agcgacccaa gtccttctct accgagaagt | 1680 |
| acgctgagct cgttggctc aagctctaa | 1709 |

<210> SEQ ID NO 68

<211> LENGTH: 465

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<212> TYPE: PRT
 <213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 68

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Met Ile Ser Ala Ile Arg Pro Ala Val Arg Ser Ser Val Arg Val Ala
1           5          10          15

Pro Met Ala Asn Thr Ala Phe Arg Ala Tyr Ser Thr Gln Asp Gly Leu
20          25          30

Lys Glu Arg Phe Ala Glu Leu Ile Pro Glu Asn Val Glu Lys Ile Lys
35          40          45

Lys Leu Arg Lys Glu Gly Asn Thr Val Ile Gly Glu Val Ile Leu
50          55          60

Asp Gln Ala Tyr Gly Gly Met Arg Gly Ile Lys Gly Leu Val Trp Glu
65          70          75          80

Gly Ser Val Leu Asp Pro Glu Glu Gly Ile Arg Phe Arg Gly Leu Thr
85          90          95

Ile Pro Asp Leu Gln Lys Gln Leu Pro His Ala Pro Gly Gly Lys Glu
100         105         110

Pro Leu Pro Glu Gly Leu Phe Trp Leu Leu Leu Thr Gly Glu Ile Pro
115         120         125

Thr Asp Ala Gln Val Lys Gly Leu Ser Ala Asp Trp Ala Ser Arg Ala
130         135         140

Glu Ile Pro Lys His Val Glu Glu Leu Ile Asp Arg Cys Pro Pro Thr
145         150         155         160

Leu His Pro Met Ala Gln Leu Gly Ile Ala Val Asn Ala Leu Glu Ser
165         170         175

Glu Ser Gln Phe Thr Lys Ala Tyr Glu Lys Gly Val Asn Lys Lys Glu
180         185         190

Tyr Trp Gln Tyr Thr Tyr Glu Asp Ser Met Asn Leu Ile Ala Lys Leu
195         200         205

Pro Val Ile Ala Ser Arg Ile Tyr Arg Asn Leu Phe Lys Asp Gly Lys
210         215         220

Ile Val Gly Ser Ile Asp Asn Ser Leu Asp Tyr Ser Ala Asn Phe Ala
225         230         235         240

Ser Leu Leu Gly Phe Gly Asp Asn Lys Glu Phe Ile Glu Leu Leu Arg
245         250         255

Leu Tyr Leu Thr Ile His Ala Asp His Glu Gly Asn Val Ser Ala
260         265         270

His Thr Thr Lys Leu Val Gly Ser Ala Leu Ser Ser Pro Phe Leu Ser
275         280         285

Leu Ser Ala Gly Leu Asn Gly Leu Ala Gly Pro Leu His Gly Arg Ala
290         295         300

Asn Gln Glu Val Leu Glu Trp Ile Leu Glu Met Lys Ser Lys Ile Gly
305         310         315         320

Ser Asp Val Thr Lys Glu Asp Ile Glu Lys Tyr Leu Trp Asp Thr Leu
325         330         335

Lys Ala Gly Arg Val Val Pro Gly Tyr Gly His Ala Val Leu Arg Lys
340         345         350

Thr Asp Pro Arg Tyr Thr Ala Gln Arg Glu Phe Ala Leu Glu His Met
355         360         365

Pro Asp Tyr Asp Leu Phe His Leu Val Ser Thr Ile Tyr Glu Val Ala
370         375         380

Pro Lys Val Leu Thr Glu His Gly Lys Thr Lys Asn Pro Trp Pro Asn
385         390         395         400

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Val Asp Ser His Ser Gly Val Leu Leu Gln Tyr Tyr Gly Leu Thr Glu
405 410 415

Gln Ser Tyr Tyr Thr Val Leu Phe Gly Val Ser Arg Ala Ile Gly Val
420 425 430

Leu Pro Gln Leu Ile Met Asp Arg Ala Tyr Gly Ala Pro Ile Glu Arg
435 440 445

Pro Lys Ser Phe Ser Thr Glu Lys Tyr Ala Glu Leu Val Gly Leu Lys
450 455 460

Leu
465

<210> SEQ ID NO 69
<211> LENGTH: 7270
<212> TYPE: DNA
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 69

| | |
|---|------|
| atgcgactgc aattgaggac actaacacgt cggttttca ggtgagtaaa cgacgggtggc | 60 |
| cgtggccacg acagccgagg cgtcacgtat ggccagacga gcacattctc gcccaccaa | 120 |
| cctcgccacg acaagaaaact aaccaggatgg ggcttcagga tcttcaacgc cagatgtggc | 180 |
| tcccttggtg gaccccaaca ttccacaaagg tctcgccctc catttcttg gactcaattc | 240 |
| tgtccacaca gccaaggccct caaaagtcaa ggagtttg tgcttcacgc gaggtcatac | 300 |
| agtttatcaac aagggtgatgg tttgacgttt agactgtata acaggcgccc gcagtgcac | 360 |
| aacgaccaaa aagggtcgaa aaagggtcgaa aaacggacac aaaagctgga aaacaagagt | 420 |
| gtaatacatt cttacacgtc caattgttag acaaacaacgg ctgttcggtc cccaaaccac | 480 |
| cagtatcacc tattttccac ttgtgtctcg gatctgatca taatctgatc tcaagatgaa | 540 |
| atttacgcca ccgacatgtat atttgatgatt tcggattctc cagaccgagc agattccacg | 600 |
| aataccacca cttggccacc ttcaaggccgct tctcggcgcg attcggccact ttccccaaacg | 660 |
| agtgttacta acccagggtcc tcatacgctaa caacggattt gcccggatgg aggagatccg | 720 |
| ttcagtgatcgaa aatggggccct acgagacattt tggcgacgag cgagcaatct cggttacccgt | 780 |
| catggccacc cccgaagatc tcgctgcca cggcgactac attagaatgg ccgatcgat | 840 |
| cgtcgagggtg cccggaggaa ccaacaacaa caactacgccc aacgtcgagc tgattgtcgaa | 900 |
| cgtggctgag cgattcggcg tcgatgccgt gtggggccatg ccagtggaaaa | 960 |
| tccccctgctc cccgagtcgc tagcgccctc tccccgcaag attgtttca tcggccctcc | 1020 |
| cggagctgcc atgagatctc tggggagacaa aatttcttct accattgtgg cccagcacgc | 1080 |
| aaagggtcccg tgtatccgt ggtctggaaac cggagtgccgt gagggttggg ttgacaagag | 1140 |
| caccaacctc gtgtccgtgt ccgaggaggt gtacaccaag ggctgcacca cccgtccaa | 1200 |
| gcagggtctg gagaaggctc agcagattgg attccccgtg atgatcaagg cttccgagg | 1260 |
| aggaggagga aagggtattt gaaagggtgaa gcgagaggag gacttcgagg ctgttacca | 1320 |
| ccagggtcgcg ggagagatcc cccgctcgcc catcttcatt atgcagctg caggcaatgc | 1380 |
| ccggcatttg gaggtgcagc ttctggctga tcagtaacggc aacaatattt cactgtttgg | 1440 |
| tcgagattgt tcggttcagc gacggcatca aaagattatt gaggaggctc ctgtgactgt | 1500 |
| ggctggccag cagacccatca ctgcacatggaa gaaggctgcc gtgcgactcg gtaagctgt | 1560 |
| cgatgatgtc tctgcaggta ccgttgaata tctgtattcc catgaggacg acaagttcta | 1620 |
| cttcttggag ctgaatccctc gtcttcagggt cgaacatccct accaccgaga tggtcaccgg | 1680 |

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| | |
|--|------|
| tgtcaacctg cccgcgtgcc agcttcagat cgccatgggt atccccctcg atcgaatcaa | 1740 |
| ggacattcgt ctcttttacg gtgttaaccc tcacaccacc actccaaatg atttcgactt | 1800 |
| ctcggggcag gatgctgata agacacagcg acgtcccgtc ccccgaggtc acaccactgc | 1860 |
| ttgccgaatc acatccgagg accctggaga gggtttcaag ccctccggag gtactatgca | 1920 |
| cgagctcaac ttccgatcct cgtccaacgt gtggggttac ttctccgtt gtaaccagg | 1980 |
| aggtatccat tcgttctcggtt attcgcagtt tggcacatc ttccgttgcgt gtagaaaccg | 2040 |
| aagtgcgtct cgaaaggcaca tgggttgtgc tttgaaggaa ctatctatc gaggtgactt | 2100 |
| ccgaaccacc gtcgagttacc tcatcaagct gctggagaca cccgacttcg aggacaacac | 2160 |
| catcaccacc ggctggctgg atgagcttat ctccaaacaag ctgactgccc agcgaccg | 2220 |
| ctcggttctc gctgttggtt gtgggtgtgc taccaaggcc catcgagctt ccgaggactc | 2280 |
| tattgcacc tacatggctt cgcttagagaa gggccaggtc cctgctcgag acattctcaa | 2340 |
| gacccttttc cccgttgcact tcatctacga gggccagcggg tacaaggttca ccggccacccg | 2400 |
| gtcggtctgag gactcttaca cgctgttcat caacggttct cgtgcgaca ttggagttag | 2460 |
| acctcttttgc acgggtggta ttctgtgtct tggtaggtggg agatcccaca atgtctactg | 2520 |
| gaaggaggag gttggagcca cgcgactgtc tggtgactcc aagacgtgcc ttctcgaggt | 2580 |
| ggagaacgcac cccactcage ttcgatctcc ctctcccggt aagctggta agtctctgg | 2640 |
| cgagaacggc gaccacgtgc gagecaacca gcccattgcc gagattgagg tcatgaagat | 2700 |
| gtacatgact ctcaactgtc aggaggacgg tattgtccag ctgtatgcg agccgggttc | 2760 |
| caccatcgag gctggcgaca tccctcgat cttggccctt gatgatcctt ccaaggtaa | 2820 |
| gcatgccaag cccttgggg gccagcttcc cgagcttggg ccccccactc tcageggtaa | 2880 |
| caaggctcat cagcgatacg agcaactgtca gaacgtgtc cataacattc tgcttggtt | 2940 |
| cgataaccag gtgggtatga agtccactct tcaggagatg gtgggtctgc tccgaaaccc | 3000 |
| ttagcttcct tatctccagt gggctcatca ggtgtcttct ctgcacaccc gaatgagcgc | 3060 |
| caagctggat gctactcttgc tgggtctcat tgacaaggcc aagcagcggag tggegagtt | 3120 |
| tcctgecaag cagcttctgc gagecccttga gaaggaggcg agctctggcg aggtcgatgc | 3180 |
| gtcttcctcag caaactcttgc tccctctgtt tgaccttgcgt cggaggtacc aggaeggtct | 3240 |
| tgctatccac gagcttcagg ttgtgtcagg cttctgcag gcctactacg actctgaggc | 3300 |
| ccgggttctgc ggacccaacg tacgtgacga ggatgtcatt ctcaagcttc gagaggagaa | 3360 |
| ccgagattct ctgcggaaagg ttgtgtatggc ccagctgtct cattctcgag tcggagccaa | 3420 |
| gaacaacctt gtgctggccc ttctcgatga atacaagggtg gccgaccagg ctggcacccg | 3480 |
| ctctctgtcc tccaaacgtgc acgttgcataa gtacttgcga cctgtgtgc gaaagatgt | 3540 |
| ggagctggaa tctcgaggtt ctgccaaggat atctctgaaa gcccggagaga ttctcatcca | 3600 |
| gtgcgtctgc ccctctctaa aggagcgaac tgaccagctt gaggcacatc tgcgatctc | 3660 |
| tgtcggtcgag tctcgatatacg gagagggtgg tctggagcac cgaactcccc gagccgat | 3720 |
| tctcaaggag gttgtcgact ccaagtgatcat tgcgtttgtat gtgcgttgcggcc agtttttgc | 3780 |
| ccacgtatcg ccctggatcg tccttgcgtc cctggagctg tacatccgac gagcttgcac | 3840 |
| ggcctactcc atccctggaca tcaactacca ccaggactcg gacctgcctc ccgtcatctc | 3900 |
| gtggcgattt agactgccta ccatgtcgatc tgctttgtac aactcagtag tgcgttctgg | 3960 |
| ctccaaaacc cccacttccc cctcggtgtc tcgagctgat tccgtctcg actttcgta | 4020 |

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| | | | | | | |
|-------------|-------------|-------------|-------------|---------------|--------------|------|
| caccgtttag | cgagactctg | ctcccgctcg | aaccggagcg | attgttgcgcg | tgcctcatct | 4080 |
| ggatgatctg | gaggatgctc | tgactcgtgt | tctggagaac | ctgccccaaac | ggggcgctgg | 4140 |
| tcttgecate | tctgttggtg | ctagcaacaa | gagtgcgcgt | gcttcgtctc | gtgaegctgc | 4200 |
| tgctgtgcc | gcttcatccg | ttgacactgg | cctgtccaac | atttgcacg | ttatgatgg | 4260 |
| tcgggtttag | gagtcgtatg | acgacgacac | tctgattgcc | cgaaatccc | aggtaattga | 4320 |
| ggactttaag | gaggactttg | aggectgttc | tctgcgacac | atcaccttct | ccttggcaa | 4380 |
| ctcccgaggt | acttatccca | agtatttac | gttccgaggc | cccgataacg | aggaggaccc | 4440 |
| caactatccg | cacattgagc | ctgctctggc | cttccagctg | gagctcgccc | gtctgtccaa | 4500 |
| cttcgacatc | aagcctgtcc | acaccgacaa | ccgaaacatc | cacgtgtacg | aggctactgg | 4560 |
| caagaacgct | gcttccgaca | agcggttctt | cacccgaggt | atcgtacgac | ctggcgtct | 4620 |
| tcgagagaac | atccccacct | cgaggtatct | catttccgag | gctgaccggc | tcatgagcga | 4680 |
| tattnnngac | gctcttagagg | tgatttggaa | caccaactcg | gatctcaacc | acatttcat | 4740 |
| caacttctca | gccgtttttg | ctctgaagcc | cgaggaggtt | gaagctgcct | ttggcggg | 4800 |
| cctggagcga | tttggccgac | gtctgtggcg | acttcgagtc | accgggtccg | agatccgaat | 4860 |
| gatggtatcc | gaccccgaaa | ctggctctgc | tttccctctg | cgagcaatga | tcaacaacgt | 4920 |
| ctctggttac | gttgtcagt | ctgagctgta | cgctgaggcc | aagaacgaca | agggccagt | 4980 |
| gattnncaag | tctctgggca | agccggctc | catgcacatg | cggtctatca | acactcccta | 5040 |
| ccccaccaag | gagtggtctgc | agcccaagcg | gtacaaggcc | catctgatgg | gtaccaccta | 5100 |
| ctgctatgac | ttccccgagc | tggtccgaca | gtccatttag | tcggactgga | agaagtatga | 5160 |
| cggcaaggct | cccgacgatc | tcatgacttg | caacgagctg | attctcgatg | aggactctgg | 5220 |
| c gagctgcag | gagggtgaacc | gagagcccg | cgccaaacac | gtcggatgg | ttgcgtggaa | 5280 |
| gtttgaggcc | aagacccccc | agtaccctcg | aggccgatct | t tc atcg tgg | tggccaa cga | 5340 |
| tatcacccctc | c agattggtt | cg tttggccc | tgctgaggac | c agtttcttct | tcaagg t gac | 5400 |
| ggagctggct | cgaaagctcg | gtatttctcg | aatctatctg | tctgccaact | ctggtgc t c | 5460 |
| aatcggcatt | gctgacgagc | tcg tggca | gtacaagggt | g cgtggaa | acgagactga | 5520 |
| cccttccaag | ggcttcaagt | accttta ctt | cacccctgag | t ct ttc gcca | ccctcaagcc | 5580 |
| cgacacttgtt | gtcaccactg | agattgagga | ggagggtccc | aa cggcgtgg | agaagcgtca | 5640 |
| tgtgatcgac | ta catttgcg | gagagaagga | cggtctcgga | gtcgagtgtc | tg cgggg | 5700 |
| tgg tctcatt | gcaggcgc | cttctcgac | ctacaaggat | atcttca | tcaactctgt | 5760 |
| cacctgtcga | tccgttggta | tcggtgctt | c ttgttctgt | cttgg | tcaac | 5820 |
| gattgaggcc | cagcccatca | ttctca | ttggatgg | tgcccc | ccgatgg | 5880 |
| agaggcttac | tcttccaact | tgca | tggtactcag | atcatgtaca | acaacgg | 5940 |
| gtctcatctg | actgc | ccg | atgatctaa | cggtgtcc | aaagatcatg | 6000 |
| atacatccct | gttctcgag | gttcc | cg | cctcaca | aga | 6060 |
| ggatcgagac | gtgacgttcc | agc | ctg | ccg | acg | 6120 |
| ttctggccga | actctcgagg | atgg | gttctgtt | cg | atgg | 6180 |
| ccaggagact | ctgtctggct | gg | ggca | agg | gg | 6240 |
| catcccttc | ggtgtcattg | gt | gtc | gag | ac | 6300 |
| tcccgccaac | ccggactcta | tt | gagatg | g | ccggccagg | 6360 |
| caactcgcc | ttcaagac | ctc | aggccat | caac | gacttc | 6420 |

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| | | | | | | |
|-------------|-------------|-------------|------------|-------------|-------------|------|
| tctcatgatt | tttgtaact | ggcgaggcgtt | ttctgggtgt | cagcgagaca | tgtacaatga | 6480 |
| ggttctcaag | tacggatctt | tcattgtgt | tgctctggtt | gactacaagc | agcccatcat | 6540 |
| ggtgtacatc | cctcccaccc | gtgagctg | agggtgggtt | ttgggtgtgg | ttgaccccac | 6600 |
| catcaactcg | gacatgtatgg | agatgtacgc | tgacgtcgag | tctcgaggtg | gtgtgtgtgg | 6660 |
| gccccgaggga | atggtcggta | tcaagtaccg | acgagaca | ctactggaca | ccatggctcg | 6720 |
| tctggatccc | gagttactcct | ctctcaagaa | gcagctttag | gagttctcccg | attcttgagga | 6780 |
| getcaagggtc | aagctcagcg | tgcgagagaa | gtctctcatg | cccatctacc | agcagatctc | 6840 |
| cgtgcagttt | gccgacttgc | atgaccgagc | tggccgaatg | gaggccaagg | gtgtcattcg | 6900 |
| tgaggcgtct | gtgtggaaagg | atgtctcg | attcttc | tggcgaatcc | gacgacgatt | 6960 |
| atgtcgaggag | tacctcatta | ccaagatcaa | tagcattcg | ccctcttgca | ctcggttga | 7020 |
| gtgtctggct | cgaatcaagt | cgtggaaagcc | tgcacttctt | gatcagggtct | ctgaccgggg | 7080 |
| tgttgccgag | tggtttgacg | agaactctga | tgccgtctct | gctcgactca | gcgagctcaa | 7140 |
| gaaggacgct | tctgcccagt | cgtttgc | tcaactgaga | aaggaccgac | agggtactct | 7200 |
| ccagggcatg | aagcaggc | tcgttctct | ttctgaggct | gagcggggctg | agctgtctcaa | 7260 |
| qqqqttatqa | | | | | | 7270 |

<210> SEQ ID NO 70

<211> LENGTH: 2266

<212> TYPE: PRT

<213> ORGANISM: *Yarrowia lipolytica*

<400> SEQUENCE: 70

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Arg | Leu | Gln | Leu | Arg | Thr | Leu | Thr | Arg | Arg | Phe | Phe | Ser | Met | Ala |
| 1 | | | | 5 | | | | | 10 | | | | | 15 | |

Ser Gly Ser Ser Thr Pro Asp Val Ala Pro Leu Val Asp Pro Asn Ile
20 25 30

His Lys Gly Leu Ala Ser His Phe Phe Gly Leu Asn Ser Val His Thr
35 40 45

Ala Lys Pro Ser Lys Val Lys Glu Phe Val Ala Ser His Gly Gly His
50 55 60

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Thr | Val | Ile | Asn | Lys | Val | Leu | Ile | Ala | Asn | Asn | Gly | Ile | Ala | Ala | Val |
| 65 | | | | | 70 | | | | | 75 | | | | | 80 |

Lys Glu Ile Arg Ser Val Arg Lys Trp Ala Tyr Glu Thr Phe Gly Asp
85 90 95

Glu Arg Ala Ile Ser Phe Thr Val Met Ala Thr Pro Glu Asp Leu Ala
100 105 110

Ala Asn Ala Asp Tyr Ile Arg Met Ala Asp Gln Tyr Val Glu Val Pro
115 120 125

Gly Gly Thr Asn Asn Asn Tyr Ala Asn Val Glu Leu Ile Val Asp
130 135 140

Val Ala Glu Arg Phe Gly Val Asp Ala Val Trp Ala Gly Trp Gly His
145 150 155 160

Ala Ser Glu Asn Pro Leu Leu Pro Glu Ser Leu Ala Ala Ser Pro Arg
165 170 175

Lys Ile Val Phe Ile Gly Pro Pro Gly Ala Ala Met Arg Ser Leu Gly
180 185 190

Asp Lys Ile Ser Ser Thr Ile Val Ala Gln His Ala Lys Val Pro Cys
195 200 205

Ile Pro Trp Ser Gly Thr Gly Val Asp Glu Val Val Val Val Asp Lys Ser

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| | | |
|---|-----|-----|
| 210 | 215 | 220 |
| Thr Asn Leu Val Ser Val Ser Glu Glu Val Tyr Thr Lys Gly Cys Thr | | |
| 225 | 230 | 235 |
| 240 | | |
| Thr Gly Pro Lys Gln Gly Leu Glu Lys Ala Lys Gln Ile Gly Phe Pro | | |
| 245 | 250 | 255 |
| Val Met Ile Lys Ala Ser Glu Gly Gly Lys Gly Ile Arg Lys | | |
| 260 | 265 | 270 |
| Val Glu Arg Glu Glu Asp Phe Glu Ala Ala Tyr His Gln Val Glu Gly | | |
| 275 | 280 | 285 |
| Glu Ile Pro Gly Ser Pro Ile Phe Ile Met Gln Leu Ala Gly Asn Ala | | |
| 290 | 295 | 300 |
| Arg His Leu Glu Val Gln Leu Leu Ala Asp Gln Tyr Gly Asn Asn Ile | | |
| 305 | 310 | 315 |
| 320 | | |
| Ser Leu Phe Gly Arg Asp Cys Ser Val Gln Arg Arg His Gln Lys Ile | | |
| 325 | 330 | 335 |
| Ile Glu Glu Ala Pro Val Thr Val Ala Gly Gln Gln Thr Phe Thr Ala | | |
| 340 | 345 | 350 |
| Met Glu Lys Ala Ala Val Arg Leu Gly Lys Leu Val Gly Tyr Val Ser | | |
| 355 | 360 | 365 |
| Ala Gly Thr Val Glu Tyr Leu Tyr Ser His Glu Asp Asp Lys Phe Tyr | | |
| 370 | 375 | 380 |
| Phe Leu Glu Leu Asn Pro Arg Leu Gln Val Glu His Pro Thr Thr Glu | | |
| 385 | 390 | 395 |
| 400 | | |
| Met Val Thr Gly Val Asn Leu Pro Ala Ala Gln Leu Gln Ile Ala Met | | |
| 405 | 410 | 415 |
| Gly Ile Pro Leu Asp Arg Ile Lys Asp Ile Arg Leu Phe Tyr Gly Val | | |
| 420 | 425 | 430 |
| Asn Pro His Thr Thr Pro Ile Asp Phe Asp Phe Ser Gly Glu Asp | | |
| 435 | 440 | 445 |
| Ala Asp Lys Thr Gln Arg Arg Pro Val Pro Arg Gly His Thr Thr Ala | | |
| 450 | 455 | 460 |
| Cys Arg Ile Thr Ser Glu Asp Pro Gly Glu Gly Phe Lys Pro Ser Gly | | |
| 465 | 470 | 475 |
| 480 | | |
| Gly Thr Met His Glu Leu Asn Phe Arg Ser Ser Ser Asn Val Trp Gly | | |
| 485 | 490 | 495 |
| Tyr Phe Ser Val Gly Asn Gln Gly Ile His Ser Phe Ser Asp Ser | | |
| 500 | 505 | 510 |
| Gln Phe Gly His Ile Phe Ala Phe Gly Glu Asn Arg Ser Ala Ser Arg | | |
| 515 | 520 | 525 |
| Lys His Met Val Val Ala Leu Lys Glu Leu Ser Ile Arg Gly Asp Phe | | |
| 530 | 535 | 540 |
| Arg Thr Thr Val Glu Tyr Leu Ile Lys Leu Leu Glu Thr Pro Asp Phe | | |
| 545 | 550 | 555 |
| 560 | | |
| Glu Asp Asn Thr Ile Thr Thr Gly Trp Leu Asp Glu Leu Ile Ser Asn | | |
| 565 | 570 | 575 |
| Lys Leu Thr Ala Glu Arg Pro Asp Ser Phe Leu Ala Val Val Cys Gly | | |
| 580 | 585 | 590 |
| Ala Ala Thr Lys Ala His Arg Ala Ser Glu Asp Ser Ile Ala Thr Tyr | | |
| 595 | 600 | 605 |
| Met Ala Ser Leu Glu Lys Gly Gln Val Pro Ala Arg Asp Ile Leu Lys | | |
| 610 | 615 | 620 |
| Thr Leu Phe Pro Val Asp Phe Ile Tyr Glu Gly Gln Arg Tyr Lys Phe | | |
| 625 | 630 | 635 |
| 640 | | |

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Thr Ala Thr Arg Ser Ser Glu Asp Ser Tyr Thr Leu Phe Ile Asn Gly
 645 650 655
 Ser Arg Cys Asp Ile Gly Val Arg Pro Leu Ser Asp Gly Gly Ile Leu
 660 665 670
 Cys Leu Val Gly Gly Arg Ser His Asn Val Tyr Trp Lys Glu Glu Val
 675 680 685
 Gly Ala Thr Arg Leu Ser Val Asp Ser Lys Thr Cys Leu Leu Glu Val
 690 695 700
 Glu Asn Asp Pro Thr Gln Leu Arg Ser Pro Ser Pro Gly Lys Leu Val
 705 710 715 720
 Lys Phe Leu Val Glu Asn Gly Asp His Val Arg Ala Asn Gln Pro Tyr
 725 730 735
 Ala Glu Ile Glu Val Met Lys Met Tyr Met Thr Leu Thr Ala Gln Glu
 740 745 750
 Asp Gly Ile Val Gln Leu Met Lys Gln Pro Gly Ser Thr Ile Glu Ala
 755 760 765
 Gly Asp Ile Leu Gly Ile Leu Ala Leu Asp Asp Pro Ser Lys Val Lys
 770 775 780
 His Ala Lys Pro Phe Glu Gln Leu Pro Glu Leu Gly Pro Pro Thr
 785 790 795 800
 Leu Ser Gly Asn Lys Pro His Gln Arg Tyr Glu His Cys Gln Asn Val
 805 810 815
 Leu His Asn Ile Leu Leu Gly Phe Asp Asn Gln Val Val Met Lys Ser
 820 825 830
 Thr Leu Gln Glu Met Val Gly Leu Leu Arg Asn Pro Glu Leu Pro Tyr
 835 840 845
 Leu Gln Trp Ala His Gln Val Ser Ser Leu His Thr Arg Met Ser Ala
 850 855 860
 Lys Leu Asp Ala Thr Leu Ala Gly Leu Ile Asp Lys Ala Lys Gln Arg
 865 870 875 880
 Gly Gly Glu Phe Pro Ala Lys Gln Leu Leu Arg Ala Leu Glu Lys Glu
 885 890 895
 Ala Ser Ser Gly Glu Val Asp Ala Leu Phe Gln Gln Thr Leu Ala Pro
 900 905 910
 Leu Phe Asp Leu Ala Arg Glu Tyr Gln Asp Gly Leu Ala Ile His Glu
 915 920 925
 Leu Gln Val Ala Ala Gly Leu Leu Gln Ala Tyr Tyr Asp Ser Glu Ala
 930 935 940
 Arg Phe Cys Gly Pro Asn Val Arg Asp Glu Asp Val Ile Leu Lys Leu
 945 950 955 960
 Arg Glu Glu Asn Arg Asp Ser Leu Arg Lys Val Val Met Ala Gln Leu
 965 970 975
 Ser His Ser Arg Val Gly Ala Lys Asn Asn Leu Val Leu Ala Leu Leu
 980 985 990
 Asp Glu Tyr Lys Val Ala Asp Gln Ala Gly Thr Asp Ser Pro Ala Ser
 995 1000 1005
 Asn Val His Val Ala Lys Tyr Leu Arg Pro Val Leu Arg Lys Ile
 1010 1015 1020
 Val Glu Leu Glu Ser Arg Ala Ser Ala Lys Val Ser Leu Lys Ala
 1025 1030 1035
 Arg Glu Ile Leu Ile Gln Cys Ala Leu Pro Ser Leu Lys Glu Arg
 1040 1045 1050

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Thr Asp Gln Leu Glu His Ile Leu Arg Ser Ser Val Val Glu Ser
 1055 1060 1065
 Arg Tyr Gly Glu Val Gly Leu Glu His Arg Thr Pro Arg Ala Asp
 1070 1075 1080
 Ile Leu Lys Glu Val Val Asp Ser Lys Tyr Ile Val Phe Asp Val
 1085 1090 1095
 Leu Ala Gln Phe Phe Ala His Asp Asp Pro Trp Ile Val Leu Ala
 1100 1105 1110
 Ala Leu Glu Leu Tyr Ile Arg Arg Ala Cys Lys Ala Tyr Ser Ile
 1115 1120 1125
 Leu Asp Ile Asn Tyr His Gln Asp Ser Asp Leu Pro Pro Val Ile
 1130 1135 1140
 Ser Trp Arg Phe Arg Leu Pro Thr Met Ser Ser Ala Leu Tyr Asn
 1145 1150 1155
 Ser Val Val Ser Ser Gly Ser Lys Thr Pro Thr Ser Pro Ser Val
 1160 1165 1170
 Ser Arg Ala Asp Ser Val Ser Asp Phe Ser Tyr Thr Val Glu Arg
 1175 1180 1185
 Asp Ser Ala Pro Ala Arg Thr Gly Ala Ile Val Ala Val Pro His
 1190 1195 1200
 Leu Asp Asp Leu Glu Asp Ala Leu Thr Arg Val Leu Glu Asn Leu
 1205 1210 1215
 Pro Lys Arg Gly Ala Gly Leu Ala Ile Ser Val Gly Ala Ser Asn
 1220 1225 1230
 Lys Ser Ala Ala Ala Ser Ala Arg Asp Ala Ala Ala Ala Ala Ala
 1235 1240 1245
 Ser Ser Val Asp Thr Gly Leu Ser Asn Ile Cys Asn Val Met Ile
 1250 1255 1260
 Gly Arg Val Asp Glu Ser Asp Asp Asp Asp Thr Leu Ile Ala Arg
 1265 1270 1275
 Ile Ser Gln Val Ile Glu Asp Phe Lys Glu Asp Phe Glu Ala Cys
 1280 1285 1290
 Ser Leu Arg Arg Ile Thr Phe Ser Phe Gly Asn Ser Arg Gly Thr
 1295 1300 1305
 Tyr Pro Lys Tyr Phe Thr Phe Arg Gly Pro Ala Tyr Glu Glu Asp
 1310 1315 1320
 Pro Thr Ile Arg His Ile Glu Pro Ala Leu Ala Phe Gln Leu Glu
 1325 1330 1335
 Leu Ala Arg Leu Ser Asn Phe Asp Ile Lys Pro Val His Thr Asp
 1340 1345 1350
 Asn Arg Asn Ile His Val Tyr Glu Ala Thr Gly Lys Asn Ala Ala
 1355 1360 1365
 Ser Asp Lys Arg Phe Phe Thr Arg Gly Ile Val Arg Pro Gly Arg
 1370 1375 1380
 Leu Arg Glu Asn Ile Pro Thr Ser Glu Tyr Leu Ile Ser Glu Ala
 1385 1390 1395
 Asp Arg Leu Met Ser Asp Ile Leu Asp Ala Leu Glu Val Ile Gly
 1400 1405 1410
 Thr Thr Asn Ser Asp Leu Asn His Ile Phe Ile Asn Phe Ser Ala
 1415 1420 1425
 Val Phe Ala Leu Lys Pro Glu Glu Val Glu Ala Ala Phe Gly Gly
 1430 1435 1440
 Phe Leu Glu Arg Phe Gly Arg Arg Leu Trp Arg Leu Arg Val Thr

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| | | |
|---|------|------|
| 1445 | 1450 | 1455 |
| Gly Ala Glu Ile Arg Met Met Val Ser Asp Pro Glu Thr Gly Ser | | |
| 1460 | 1465 | 1470 |
| Ala Phe Pro Leu Arg Ala Met Ile Asn Asn Val Ser Gly Tyr Val | | |
| 1475 | 1480 | 1485 |
| Val Gln Ser Glu Leu Tyr Ala Glu Ala Lys Asn Asp Lys Gly Gln | | |
| 1490 | 1495 | 1500 |
| Trp Ile Phe Lys Ser Leu Gly Lys Pro Gly Ser Met His Met Arg | | |
| 1505 | 1510 | 1515 |
| Ser Ile Asn Thr Pro Tyr Pro Thr Lys Glu Trp Leu Gln Pro Lys | | |
| 1520 | 1525 | 1530 |
| Arg Tyr Lys Ala His Leu Met Gly Thr Thr Tyr Cys Tyr Asp Phe | | |
| 1535 | 1540 | 1545 |
| Pro Glu Leu Phe Arg Gln Ser Ile Glu Ser Asp Trp Lys Lys Tyr | | |
| 1550 | 1555 | 1560 |
| Asp Gly Lys Ala Pro Asp Asp Leu Met Thr Cys Asn Glu Leu Ile | | |
| 1565 | 1570 | 1575 |
| Leu Asp Glu Asp Ser Gly Glu Leu Gln Glu Val Asn Arg Glu Pro | | |
| 1580 | 1585 | 1590 |
| Gly Ala Asn Asn Val Gly Met Val Ala Trp Lys Phe Glu Ala Lys | | |
| 1595 | 1600 | 1605 |
| Thr Pro Glu Tyr Pro Arg Gly Arg Ser Phe Ile Val Val Ala Asn | | |
| 1610 | 1615 | 1620 |
| Asp Ile Thr Phe Gln Ile Gly Ser Phe Gly Pro Ala Glu Asp Gln | | |
| 1625 | 1630 | 1635 |
| Phe Phe Phe Lys Val Thr Glu Leu Ala Arg Lys Leu Gly Ile Pro | | |
| 1640 | 1645 | 1650 |
| Arg Ile Tyr Leu Ser Ala Asn Ser Gly Ala Arg Ile Gly Ile Ala | | |
| 1655 | 1660 | 1665 |
| Asp Glu Leu Val Gly Lys Tyr Lys Val Ala Trp Asn Asp Glu Thr | | |
| 1670 | 1675 | 1680 |
| Asp Pro Ser Lys Gly Phe Lys Tyr Leu Tyr Phe Thr Pro Glu Ser | | |
| 1685 | 1690 | 1695 |
| Leu Ala Thr Leu Lys Pro Asp Thr Val Val Thr Glu Ile Glu | | |
| 1700 | 1705 | 1710 |
| Glu Glu Gly Pro Asn Gly Val Glu Lys Arg His Val Ile Asp Tyr | | |
| 1715 | 1720 | 1725 |
| Ile Val Gly Glu Lys Asp Gly Leu Gly Val Glu Cys Leu Arg Gly | | |
| 1730 | 1735 | 1740 |
| Ser Gly Leu Ile Ala Gly Ala Thr Ser Arg Ala Tyr Lys Asp Ile | | |
| 1745 | 1750 | 1755 |
| Phe Thr Leu Thr Leu Val Thr Cys Arg Ser Val Gly Ile Gly Ala | | |
| 1760 | 1765 | 1770 |
| Tyr Leu Val Arg Leu Gly Gln Arg Ala Ile Gln Ile Glu Gly Gln | | |
| 1775 | 1780 | 1785 |
| Pro Ile Ile Leu Thr Gly Ala Pro Ala Ile Asn Lys Leu Leu Gly | | |
| 1790 | 1795 | 1800 |
| Arg Glu Val Tyr Ser Ser Asn Leu Gln Leu Gly Gly Thr Gln Ile | | |
| 1805 | 1810 | 1815 |
| Met Tyr Asn Asn Gly Val Ser His Leu Thr Ala Arg Asp Asp Leu | | |
| 1820 | 1825 | 1830 |
| Asn Gly Val His Lys Ile Met Gln Trp Leu Ser Tyr Ile Pro Ala | | |
| 1835 | 1840 | 1845 |

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Ser Arg Gly Leu Pro Val Pro Val Leu Pro His Lys Thr Asp Val
 1850 1855 1860
 Trp Asp Arg Asp Val Thr Phe Gln Pro Val Arg Gly Glu Gln Tyr
 1865 1870 1875
 Asp Val Arg Trp Leu Ile Ser Gly Arg Thr Leu Glu Asp Gly Ala
 1880 1885 1890
 Phe Glu Ser Gly Leu Phe Asp Lys Asp Ser Phe Gln Glu Thr Leu
 1895 1900 1905
 Ser Gly Trp Ala Lys Gly Val Val Val Gly Arg Ala Arg Leu Gly
 1910 1915 1920
 Gly Ile Pro Phe Gly Val Ile Gly Val Glu Thr Ala Thr Val Asp
 1925 1930 1935
 Asn Thr Thr Pro Ala Asp Pro Ala Asn Pro Asp Ser Ile Glu Met
 1940 1945 1950
 Ser Thr Ser Glu Ala Gly Gln Val Trp Tyr Pro Asn Ser Ala Phe
 1955 1960 1965
 Lys Thr Ser Gln Ala Ile Asn Asp Phe Asn His Gly Glu Ala Leu
 1970 1975 1980
 Pro Leu Met Ile Leu Ala Asn Trp Arg Gly Phe Ser Gly Gly Gln
 1985 1990 1995
 Arg Asp Met Tyr Asn Glu Val Leu Lys Tyr Gly Ser Phe Ile Val
 2000 2005 2010
 Asp Ala Leu Val Asp Tyr Lys Gln Pro Ile Met Val Tyr Ile Pro
 2015 2020 2025
 Pro Thr Gly Glu Leu Arg Gly Gly Ser Trp Val Val Val Asp Pro
 2030 2035 2040
 Thr Ile Asn Ser Asp Met Met Glu Met Tyr Ala Asp Val Glu Ser
 2045 2050 2055
 Arg Gly Gly Val Leu Glu Pro Glu Gly Met Val Gly Ile Lys Tyr
 2060 2065 2070
 Arg Arg Asp Lys Leu Leu Asp Thr Met Ala Arg Leu Asp Pro Glu
 2075 2080 2085
 Tyr Ser Ser Leu Lys Lys Gln Leu Glu Glu Ser Pro Asp Ser Glu
 2090 2095 2100
 Glu Leu Lys Val Lys Leu Ser Val Arg Glu Lys Ser Leu Met Pro
 2105 2110 2115
 Ile Tyr Gln Gln Ile Ser Val Gln Phe Ala Asp Leu His Asp Arg
 2120 2125 2130
 Ala Gly Arg Met Glu Ala Lys Gly Val Ile Arg Glu Ala Leu Val
 2135 2140 2145
 Trp Lys Asp Ala Arg Arg Phe Phe Phe Trp Arg Ile Arg Arg Arg
 2150 2155 2160
 Leu Val Glu Glu Tyr Leu Ile Thr Lys Ile Asn Ser Ile Leu Pro
 2165 2170 2175
 Ser Cys Thr Arg Leu Glu Cys Leu Ala Arg Ile Lys Ser Trp Lys
 2180 2185 2190
 Pro Ala Thr Leu Asp Gln Gly Ser Asp Arg Gly Val Ala Glu Trp
 2195 2200 2205
 Phe Asp Glu Asn Ser Asp Ala Val Ser Ala Arg Leu Ser Glu Leu
 2210 2215 2220
 Lys Lys Asp Ala Ser Ala Gln Ser Phe Ala Ser Gln Leu Arg Lys
 2225 2230 2235

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| | | | | | | | | | | | | | | |
|------|-----|-----|-----|-----|------|-----|-----|-----|-----|------|-----|-----|-----|-----|
| Asp | Arg | Gln | Gly | Thr | Leu | Gln | Gly | Met | Lys | Gln | Ala | Leu | Ala | Ser |
| 2240 | | | | | 2245 | | | | | 2250 | | | | |

| | | | | | | | | | | | | | | |
|------|-----|-----|-----|-----|------|-----|-----|-----|-----|------|-----|-----|--|--|
| Leu | Ser | Glu | Ala | Glu | Arg | Ala | Glu | Leu | Leu | Lys | Gly | Leu | | |
| 2255 | | | | | 2260 | | | | | 2265 | | | | |

<210> SEQ ID NO 71

<211> LENGTH: 1134

<212> TYPE: DNA

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 71

| | | | | | | |
|-------------|-------------|------------|------------|------------|-------------|------|
| atgtggggaa | gttcacatgc | attcgctgg | aatctgatc | tgacactaca | actacacacc | 60 |
| aggtccaaca | tgagcgacaa | tacgacaatc | aaaaagccga | tccgacccaa | accgatccgg | 120 |
| acggaacgcc | tgccttacgc | tggggccgca | aaaatcatcc | gagccaacca | gaaagaccac | 180 |
| tactttgagt | ccgtgcttga | acagcatctc | gtcacgttcc | tgcagaaatg | gaagggagta | 240 |
| cgatttatcc | accagtacaa | ggaggagctg | gagacggcgt | ccaagttgc | atatctcggt | 300 |
| tttgtgtacgc | tttgtggctc | caagactctc | ggagaagagt | acaccaatct | catgtacact | 360 |
| atcagagacc | aaacagactct | accgggggtg | gtgagacgg | ttggctacgt | gccttccaa | 420 |
| actctgttcc | catacctgtt | tgtgcgtac | atgggcaagt | tgcgcgccaa | actgtatgcgc | 480 |
| gagtatcccc | atctgggtgg | gtacgacgaa | gatgagcctg | tgcgcgtccc | ggaaacatgg | 540 |
| aaggagcggg | tcatcaagac | gtttgtgaa | aagtttgaca | agttcacggc | gctggaggg | 600 |
| tttaccgcga | tccacttggc | gatttctac | gtctacggc | cgtactacca | gctcagtaag | 660 |
| cggatctggg | gcatgcgtt | tgtatgg | caccgactgg | acaagaatga | gcctcgaatc | 720 |
| ggttacgaga | tgctcggct | gctgatttc | gcccggttt | ccacgtcatt | tgtgcagacg | 780 |
| ggaagagagt | acctcggagc | gctgctggaa | aagagcgtgg | agaaagaggc | agggggagaag | 840 |
| gaagatgaaa | aggaagcggt | tgtgccaaa | aagaagtct | caattccgtt | cattgaggat | 900 |
| acagaagggg | agacggaaga | caagatcgat | ctggaggacc | ctcgacagct | caagttcatt | 960 |
| cctgaggcgt | ccagagcgt | cactctgt | ctgtcataca | ttagtgcgcc | ggcatgtacg | 1020 |
| ccatgtggac | acttttctg | ttggactgt | atttccgaat | gggtgagaga | gaagcccgag | 1080 |
| tgcccttgt | gtcggcaggg | tgtgagagag | cagaacttgt | tgcctatcag | ataaa | 1134 |

<210> SEQ ID NO 72

<211> LENGTH: 377

<212> TYPE: PRT

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 72

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Trp | Gly | Ser | Ser | His | Ala | Phe | Ala | Gly | Glu | Ser | Asp | Leu | Thr | Leu |
| 1 | | | | | 5 | | | | 10 | | | | 15 | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gln | Leu | His | Thr | Arg | Ser | Asn | Met | Ser | Asp | Asn | Thr | Thr | Ile | Lys | Lys |
| | | | | | 20 | | | 25 | | | 30 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Pro | Ile | Arg | Pro | Lys | Pro | Ile | Arg | Thr | Glu | Arg | Leu | Pro | Tyr | Ala | Gly |
| | | | | | 35 | | | 40 | | | 45 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ala | Ala | Glu | Ile | Ile | Arg | Ala | Asn | Gln | Lys | Asp | His | Tyr | Phe | Glu | Ser |
| | | | | | 50 | | | 55 | | | 60 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Val | Leu | Glu | Gln | His | Leu | Val | Thr | Phe | Leu | Gln | Lys | Trp | Lys | Gly | Val |
| | | | | | 65 | | | 70 | | | 75 | | | 80 | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Arg | Phe | Ile | His | Gln | Tyr | Lys | Glu | Glu | Leu | Glu | Thr | Ala | Ser | Lys | Phe |
| | | | | | 85 | | | 90 | | | 95 | | | | |

Ala Tyr Leu Gly Leu Cys Thr Leu Val Gly Ser Lys Thr Leu Gly Glu

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| 100 | 105 | 110 | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Glu | Tyr | Thr | Asn | Leu | Met | Tyr | Thr | Ile | Arg | Asp | Arg | Thr | Ala | Leu | Pro |
| 115 | | | | | | | | | | | | | | | 125 |
| Gly | Val | Val | Arg | Arg | Phe | Gly | Tyr | Val | Leu | Ser | Asn | Thr | Leu | Phe | Pro |
| 130 | | | | | | | | | | | | | | | 140 |
| Tyr | Leu | Phe | Val | Arg | Tyr | Met | Gly | Lys | Leu | Arg | Ala | Lys | Leu | Met | Arg |
| 145 | | | | | | | | | | | | | | | 160 |
| Glu | Tyr | Pro | His | Leu | Val | Glu | Tyr | Asp | Glu | Asp | Glu | Pro | Val | Pro | Ser |
| 165 | | | | | | | | | | | | | | | 175 |
| Pro | Glu | Thr | Trp | Lys | Glu | Arg | Val | Ile | Lys | Thr | Phe | Val | Asn | Lys | Phe |
| 180 | | | | | | | | | | | | | | | 190 |
| Asp | Lys | Phe | Thr | Ala | Leu | Glu | Gly | Phe | Thr | Ala | Ile | His | Leu | Ala | Ile |
| 195 | | | | | | | | | | | | | | | 205 |
| Phe | Tyr | Val | Tyr | Gly | Ser | Tyr | Tyr | Gln | Leu | Ser | Lys | Arg | Ile | Trp | Gly |
| 210 | | | | | | | | | | | | | | | 220 |
| Met | Arg | Tyr | Val | Phe | Gly | His | Arg | Leu | Asp | Lys | Asn | Glu | Pro | Arg | Ile |
| 225 | | | | | | | | | | | | | | | 240 |
| Gly | Tyr | Glu | Met | Leu | Gly | Leu | Leu | Ile | Phe | Ala | Arg | Phe | Ala | Thr | Ser |
| 245 | | | | | | | | | | | | | | | 255 |
| Phe | Val | Gln | Thr | Gly | Arg | Glu | Tyr | Leu | Gly | Ala | Leu | Leu | Glu | Lys | Ser |
| 260 | | | | | | | | | | | | | | | 270 |
| Val | Glu | Lys | Glu | Ala | Gly | Glu | Lys | Glu | Asp | Glu | Lys | Glu | Ala | Val | Val |
| 275 | | | | | | | | | | | | | | | 285 |
| Pro | Lys | Lys | Lys | Ser | Ser | Ile | Pro | Phe | Ile | Glu | Asp | Thr | Glu | Gly | Glu |
| 290 | | | | | | | | | | | | | | | 300 |
| Thr | Glu | Asp | Lys | Ile | Asp | Leu | Glu | Asp | Pro | Arg | Gln | Leu | Lys | Phe | Ile |
| 305 | | | | | | | | | | | | | | | 320 |
| Pro | Glu | Ala | Ser | Arg | Ala | Cys | Thr | Leu | Cys | Leu | Ser | Tyr | Ile | Ser | Ala |
| 325 | | | | | | | | | | | | | | | 335 |
| Pro | Ala | Cys | Thr | Pro | Cys | Gly | His | Phe | Phe | Cys | Trp | Asp | Cys | Ile | Ser |
| 340 | | | | | | | | | | | | | | | 350 |
| Glu | Trp | Val | Arg | Glu | Lys | Pro | Glu | Cys | Pro | Leu | Cys | Arg | Gln | Gly | Val |
| 355 | | | | | | | | | | | | | | | 365 |
| Arg | Glu | Gln | Asn | Leu | Leu | Pro | Ile | Arg | | | | | | | |
| 370 | | | | | | | | | | | | | | | 375 |

<210> SEQ ID NO 73
<211> LENGTH: 2364
<212> TYPE: DNA
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 73

| | | | | | | |
|-------------|-------------|-------------|-------------|------------|-------------|-----|
| atgaccgaca | aggactggga | tcttgtctac | aagggtccacg | ttttcggtgc | ctacaagggtt | 60 |
| accccgagctg | cctggcctta | cttccgaaag | cagaagtacg | gtcgagttat | ctctacacct | 120 |
| tccgctgctg | gtctttacgg | aaacttcggc | cagaccaact | actccgctgc | caagctcgcc | 180 |
| ctgggtggtt | tccggtagac | tctcgccaag | gagggtgcca | agtacaacat | tacttccaac | 240 |
| gtcatcgctc | ctcttgctgc | ttccccgaatg | accgagacag | tcatgcccga | ggatatcctc | 300 |
| aagctcctca | agcctgagta | cgttggttct | ctgggtcggt | acctcaccca | cgactctgtc | 360 |
| accgagtctt | atgggtatata | cgagggtcggt | gctggttaca | tggctaaaat | ccgatgggag | 420 |
| cgaggcaacg | gtgctgtttt | caagggcgcac | gacacttca | ccccgtctgc | tattctgaag | 480 |
| cgatggatg | aggcaccc | ttttgagac | cccacctacc | ctaacggccc | tgctgacttc | 540 |

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| | | | | | | |
|-----------------------|-------------------|----------------------|-------------|-------------|-------------|------|
| ttcaaaatacgttgaggagtc | tgttaagcgaccggaga | aacccccagggacc | caccgtctcc | 600 | | |
| ttcaaggacc | agggttgtcat | tgtcaactggacccgggt | gcattggccg | agcttactct | 660 | |
| cacctccttgc | ctaagcttgg | tgccaagggttgatgtcgat | gttggtaacg | attcggtaa | ccctcagaag | 720 |
| aaattaaggc | cctcggtgg | atcgccgtcg | ctgacaagaa | caacgtcata | cacggtgaga | 780 |
| aggttgttca | gaccgctatc | gacgcctcg | gtgctgtcca | cgccgttgc | aacaacgctg | 840 |
| gtattctccg | agacaagtct | ttcgccaaca | tggatgatga | gatgtggcag | gtatcttgc | 900 |
| atgtccacct | caacggtaact | tactccgtta | ccaaggccgc | gtggccccac | ctgatcttgc | 960 |
| ttcctaaggc | agaagtacgg | ccgtgtcata | aacaccacat | caacttctgg | tatctacgg | 1020 |
| aggccaaacta | ctctgccccc | aaggctggta | tcctcggttt | ctcccgagct | aacttcggcc | 1080 |
| cttgctcgag | agggtgagaa | gtacaacatt | cttgcataaca | ccattgcccc | taacgctgg | 1140 |
| actgcccata | ctgcttctgt | cttcaactgag | gagatgtcg | agcttctcaa | gcccgttcc | 1200 |
| atcgcaccca | tcaccgtcct | gcttgcttcc | gatcaggctc | cegtcaccgg | tgatctgttt | 1260 |
| gagactgggtt | ctgcttggat | cgacagact | cgatggcagc | gagctgggtgg | taaggccttc | 1320 |
| aacccaaga | agggtgtcac | ccccgaaatg | gttcgagaca | gttgggttaa | gatgtcgac | 1380 |
| ttcgatgtat | gtactccac | ccatcccacc | actccctcg | agtctactac | tcagatttt | 1440 |
| gagaacatct | tcaacgtgcc | tgtgaggag | gttggaggaga | ctgctctcg | tgctggcccc | 1500 |
| gggtggteccg | gtatctcaa | caaggaggc | gaaccttgc | actacactta | cacttaccga | 1560 |
| gacctcattt | tttacaacact | ttgtctcggt | gccaaggcta | atgagctcaa | gtatgtttc | 1620 |
| gagggtgtat | atgacttcca | gaccgtgccc | actttcggt | ttatccctta | catgggtggc | 1680 |
| ctcatcaact | ccaactatgg | cgacttcgtt | cctaacttca | accctatgtat | gtttctccac | 1740 |
| ggtgagcgt | accttgaat | ccgacagttgg | cctatttcta | ccaatgtcac | attggagaac | 1800 |
| aaggctaagg | tcatcgatgt | cggtgacaag | ggcaaggctg | ccctccttgc | cactgtacc | 1860 |
| accaccacga | acaaggagac | tggtgaggag | gttttctaca | acgagtttcc | tcttttcata | 1920 |
| cgaggctctg | gtggtttcgg | tggtaagtct | accggtaactg | accgtggcgc | tgccactgt | 1980 |
| gccaacaaggc | ccctcgctcg | agcttctgcac | ttcgttaagg | agatcaagat | ccaggaggac | 2040 |
| caggctgocat | tttaccgact | ttctgggtat | tacaaccctc | ttcacatcga | ccctgtttt | 2100 |
| gtctgtgttgc | gtactttga | ccgacctatt | ctccacggtc | tctgtctttt | tggtgtctcc | 2160 |
| ggtaagggttc | tttacgatca | gtttgggtcc | ttcaagaacg | ctaagggtccg | atttgcgtgtt | 2220 |
| cacgtcttcc | ctggtgagac | cctgaagggtt | gagggctgga | aggaggccaa | caaggtcatt | 2280 |
| ttccagacca | aggttgttga | gctgaggtact | accggccatca | gcaatggccgc | cattgagctc | 2340 |
| ttccccaaagg | atqctaqaqt | cttaa | | | | 2364 |

<210> SEQ ID NO 74

<211> LENGTH: 787

<212> TYPE: PRT

<213> ORGANISM: *Yarrowia lipolytica*

<400> SEQUENCE: 74

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Thr | Asp | Lys | Asp | Trp | Asp | Leu | Val | Tyr | Lys | Val | His | Val | Phe | Gly |
| 1 | | | | 5 | | | | | 10 | | | | | | 15 |

Ala Tyr Lys Val Thr Arg Ala Ala Trp Pro Tyr Phe Arg Lys Gln Lys
 20 25 30

Tyr Gly Arg Val Ile Ser Thr Ser Ser Ala Ala Gly Leu Tyr Gly Asn
35 40 45

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Phe Gly Gln Thr Asn Tyr Ser Ala Ala Lys Leu Ala Leu Val Gly Phe
50 55 60

Gly Glu Thr Leu Ala Lys Glu Gly Ala Lys Tyr Asn Ile Thr Ser Asn
65 70 75 80

Val Ile Ala Pro Leu Ala Ala Ser Arg Met Thr Glu Thr Val Met Pro
85 90 95

Glu Asp Ile Leu Lys Leu Leu Lys Pro Glu Tyr Val Val Pro Leu Val
100 105 110

Gly Tyr Leu Thr His Asp Ser Val Thr Glu Ser Tyr Gly Ile Tyr Glu
115 120 125

Val Gly Ala Gly Tyr Met Ala Lys Ile Arg Trp Glu Arg Gly Asn Gly
130 135 140

Ala Val Phe Lys Gly Asp Asp Thr Phe Thr Pro Ser Ala Ile Leu Lys
145 150 155 160

Arg Trp Asp Glu Val Thr Ser Phe Glu Ser Pro Thr Tyr Pro Asn Gly
165 170 175

Pro Ala Asp Phe Phe Lys Tyr Ala Glu Glu Ser Val Lys Arg Pro Glu
180 185 190

Asn Pro Gln Gly Pro Thr Val Ser Phe Lys Asp Gln Val Val Ile Val
195 200 205

Thr Gly Ala Gly Ala Gly Ile Gly Arg Ala Tyr Ser His Leu Leu Ala
210 215 220

Lys Leu Gly Ala Lys Val Val Asn Asp Phe Gly Asn Pro Gln Lys
225 230 235 240

Val Val Asp Glu Ile Lys Ala Leu Gly Ile Ala Val Ala Asp Lys
245 250 255

Asn Asn Val Ile His Gly Glu Lys Val Val Gln Thr Ala Ile Asp Ala
260 265 270

Phe Gly Ala Val His Ala Val Val Asn Asn Ala Gly Ile Leu Arg Asp
275 280 285

Lys Ser Phe Ala Asn Met Asp Asp Glu Met Trp Gln Leu Ile Phe Asp
290 295 300

Val His Leu Asn Gly Thr Tyr Ser Val Thr Lys Ala Ala Trp Pro His
305 310 315 320

Phe Leu Lys Gln Lys Tyr Gly Arg Val Ile Asn Thr Thr Ser Thr Ser
325 330 335

Gly Ile Tyr Gly Asn Phe Gly Gln Ala Asn Tyr Ser Ala Ala Lys Ala
340 345 350

Gly Ile Leu Gly Phe Ser Arg Ala Leu Ala Arg Glu Gly Glu Lys Tyr
355 360 365

Asn Ile Leu Val Asn Thr Ile Ala Pro Asn Ala Gly Thr Ala Met Thr
370 375 380

Ala Ser Val Phe Thr Glu Glu Met Leu Glu Leu Phe Lys Pro Asp Phe
385 390 395 400

Ile Ala Pro Ile Thr Val Leu Leu Ala Ser Asp Gln Ala Pro Val Thr
405 410 415

Gly Asp Leu Phe Glu Thr Gly Ser Ala Trp Ile Gly Gln Thr Arg Trp
420 425 430

Gln Arg Ala Gly Gly Lys Ala Phe Asn Thr Lys Lys Gly Val Thr Pro
435 440 445

Glu Met Val Arg Asp Ser Trp Ala Lys Ile Val Asp Phe Asp Asp Gly
450 455 460

Asn Ser Thr His Pro Thr Thr Pro Ser Glu Ser Thr Thr Gln Ile Leu

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| | | | |
|---|-----|-----|-----|
| 465 | 470 | 475 | 480 |
| Glu Asn Ile Phe Asn Val Pro Asp Glu Glu Val Glu Glu Thr Ala Leu | | | |
| 485 | 490 | 495 | |
| Val Ala Gly Pro Gly Gly Pro Gly Ile Leu Asn Lys Glu Gly Glu Pro | | | |
| 500 | 505 | 510 | |
| Phe Asp Tyr Thr Tyr Thr Tyr Arg Asp Leu Ile Leu Tyr Asn Leu Gly | | | |
| 515 | 520 | 525 | |
| Leu Gly Ala Lys Ala Asn Glu Leu Lys Tyr Val Phe Glu Gly Asp Asp | | | |
| 530 | 535 | 540 | |
| Asp Phe Gln Thr Val Pro Thr Phe Gly Val Ile Pro Tyr Met Gly Gly | | | |
| 545 | 550 | 555 | 560 |
| Leu Ile Thr Thr Asn Tyr Gly Asp Phe Val Pro Asn Phe Asn Pro Met | | | |
| 565 | 570 | 575 | |
| Met Leu Leu His Gly Glu Gln Tyr Leu Glu Ile Arg Gln Trp Pro Ile | | | |
| 580 | 585 | 590 | |
| Pro Thr Asn Ala Thr Leu Glu Asn Lys Ala Lys Val Ile Asp Val Val | | | |
| 595 | 600 | 605 | |
| Asp Lys Gly Lys Ala Ala Leu Leu Val Thr Ala Thr Thr Thr Asn | | | |
| 610 | 615 | 620 | |
| Lys Glu Thr Gly Glu Glu Val Phe Tyr Asn Glu Ser Ser Leu Phe Ile | | | |
| 625 | 630 | 635 | 640 |
| Arg Gly Ser Gly Gly Phe Gly Gly Lys Ser Thr Gly Thr Asp Arg Gly | | | |
| 645 | 650 | 655 | |
| Ala Ala Thr Ala Ala Asn Lys Pro Pro Ala Arg Ala Pro Asp Phe Val | | | |
| 660 | 665 | 670 | |
| Lys Glu Ile Lys Ile Gln Glu Asp Gln Ala Ala Ile Tyr Arg Leu Ser | | | |
| 675 | 680 | 685 | |
| Gly Asp Tyr Asn Pro Leu His Ile Asp Pro Ala Phe Ala Ala Val Gly | | | |
| 690 | 695 | 700 | |
| Asn Phe Asp Arg Pro Ile Leu His Gly Leu Cys Ser Phe Gly Val Ser | | | |
| 705 | 710 | 715 | 720 |
| Gly Lys Ala Leu Tyr Asp Gln Phe Gly Pro Phe Lys Asn Ala Lys Val | | | |
| 725 | 730 | 735 | |
| Arg Phe Ala Gly His Val Phe Pro Gly Glu Thr Leu Lys Val Glu Gly | | | |
| 740 | 745 | 750 | |
| Trp Lys Glu Gly Asn Lys Val Ile Phe Gln Thr Lys Val Val Glu Arg | | | |
| 755 | 760 | 765 | |
| Gly Thr Thr Ala Ile Ser Asn Ala Ala Ile Glu Leu Phe Pro Lys Asp | | | |
| 770 | 775 | 780 | |
| Ala Lys Leu | | | |
| 785 | | | |

<210> SEQ ID NO 75

<211> LENGTH: 2694

<212> TYPE: DNA

<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 75

| | |
|---|-----|
| atgctggcct ctcgagttc catcaaggct gtgagtatcg atggtaaga aagacaccga | 60 |
| caatcgccac gttgtgccac agacacagac gcgttctac acacacacac acaagagtgc | 120 |
| acgtgtgggt tagccgaggt atttcgacag ggaggaaaaa cgacaacgaa aggaccgaca | 180 |
| gataccaaag caacccaatc accaccaa tcaatgatcc ccgccccgg gaatgcgaa | 240 |
| aaggcttctg cgacattaca acaaagccaa ctctgttgat ttgttgttg cgacattggc | 300 |

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| | |
|---|------|
| tttgtgcgg tccaaaatt acctcgacca accacacggc ggcaattgaa gacaatgcaa | 360 |
| attnaatagc acataactaac ccagccccgc cttgcacgat ctctcgcgac taccactaat | 420 |
| gcctccctca acttggactc caaggtccga atgaacaact gggaggccaa caacttcctc | 480 |
| aacttcaaga agcacacccga gaacgtccag attgtcaagg agcgactcaa ccgaccctg | 540 |
| acctacgctg agaagattct ctacggccat ctcgacaagg cccatgagca ggagattgtc | 600 |
| cgaggtcagt octactcaa gctgcgaccc gatcgagccg cctgcccagga tgccaccgccc | 660 |
| cagatggcca ttctgcagtt catgtctgcc ggtatccccca ccgtccagac cccaccacc | 720 |
| gtccactgtg accatcttat ccaggcccag gttgggtggt agcaggatct tgctcgagcc | 780 |
| atcgacatca acaaggaggt ctacaacttc cttggcaccc cctccgcacaa gtacgacatt | 840 |
| ggtttctgga aggccggatc cggttattatc caccagatca ttctcgagaa ctacgcctc | 900 |
| cccggtgccc ttctcatttg ttccgactct catacccccac acgcccggtagg tctcggtatg | 960 |
| ctcgccatcg gtgtcggtgg tgccgatgtc gtcgacgtca tggccggctc cccctggag | 1020 |
| cttaaggccc ocaagattat cggtgtcaag ctgaccggta agctctctgg ctggacctcc | 1080 |
| cccaaggata ttatcctgaa ggtagctgtt atcctcaccg tcaagggtgg aaccgggtct | 1140 |
| atcgtcaggt acttcgggtga tgggtgcgtt aacctgtctt gcaactggat gggaaaccatc | 1200 |
| tgttaacatgg gtgcggagat tgggtgcgtacc acctccaccc tccccctcaa cgagcgtatg | 1260 |
| gcgcgactacc ttaacgcccac tggccgaaag gagattggccg actttgctcg actttacaac | 1320 |
| cacttctct ctgccgatga gggttgttag tacgatcagc tcacgtcgat tgacatgaa | 1380 |
| acccttgagc ttacgtcaa cggcccttc actcccgatc ttgccacccc catctccaa | 1440 |
| ctcaaggatg tgcggcgtca gaacggatgg ccccttgagg tcaaggcgttgc tcttatcgcc | 1500 |
| tcttgcacca actcccttta cgaggatatg gagcgatccg cctccattgc caaggacgcc | 1560 |
| atggcccaacg gtcttaagtc caagtccatc tacaccgtca ccccccgggtc cgagcagatc | 1620 |
| cgagccacca ttgagcgttca ggggtgttagt cagacccgttcc tcgacttcgg tggatcgat | 1680 |
| cttgctaaacg ctgtggccc ctgcattgtt cagtggtggacc gacgagacat caagaagggt | 1740 |
| gagaagaaca ccattgtctc ttcttacaac cgaaacttca ctggccgaaa cgattcta | 1800 |
| cctgcccaccc acgctttcgat cacctctccc gatctcgatcc ccccttcgc cattgtgg | 1860 |
| gacctccat tcaaccctct cactgactcc ctgaggattt ctgagggtaa ggagttcaag | 1920 |
| ctcaaggagc ccactggaaa gggctgtccc gaccggatgtt acgaccccccgtt catggacacc | 1980 |
| taccaggctc ccccccggca ccgatctgcgat gtcgagggttg atgtttcccc cacttccgac | 2040 |
| cgactccaga tcctcaagcc cttcaagctt tgggacggca aggacggat tgcacatgccc | 2100 |
| atccctcatca agtctcttgg taagaccacc actgaccata tctctcgatcc cggccctgg | 2160 |
| cttaagtacc gaggccatct ccagaacatc tccaacaact acatgattgg agccatcaac | 2220 |
| gctgagaacg aggaggccaa caacgtccga aaccagatca ctggcgatgtt gggaggagtt | 2280 |
| cccgagactg ccattgttta ccgagacaac ggtatccgat ggggtgttgcgat gggaggatgt | 2340 |
| aacttcgggttgggggttcc tcgagagcac gctgcgtttt agcccccggat cctcggtgtt | 2400 |
| ttcgccatca tcaccaagtcc ttttgcggaa attcacgaga ctaacctgaa gaagcagggt | 2460 |
| ctcctgcccc ttaacttcgtt caacgggtctt gactacgaca agatccagcc ctccgataag | 2520 |
| atctccatcc ttggtcttaa ggacccgttcc cccggcaaga acgtccatccat tgaggatacc | 2580 |
| cccaaggacg gtgccaagtg gaccaccgag gtttctcaca cctacaactc tgagcagtc | 2640 |

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gagtggttca agtacggctc tgccctcaac aagatggctg cctccaagaa ataa 2694

<210> SEQ ID NO 76
<211> LENGTH: 779
<212> TYPE: PRT
<213> ORGANISM: Yarrowia lipolytica

<400> SEQUENCE: 76

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Leu | Ala | Ser | Arg | Val | Ser | Ile | Lys | Ala | Pro | Arg | Leu | Ala | Arg | Ser |
| 1 | | | | | 5 | | | 10 | | | 15 | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Ala | Thr | Thr | Thr | Asn | Ala | Ser | Leu | Asn | Leu | Asp | Ser | Lys | Val | Arg |
| | | 20 | | | | 25 | | | 30 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Asn | Asn | Trp | Glu | Ala | Asn | Asn | Phe | Leu | Asn | Phe | Lys | Lys | His | Thr |
| | 35 | | | 40 | | | | 45 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Glu | Asn | Val | Gln | Ile | Val | Lys | Glu | Arg | Leu | Asn | Arg | Pro | Leu | Thr | Tyr |
| | 50 | | | | 55 | | | 60 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ala | Glu | Lys | Ile | Leu | Tyr | Gly | His | Leu | Asp | Lys | Pro | His | Glu | Gln | Glu |
| 65 | | | | 70 | | | 75 | | 80 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ile | Val | Arg | Gly | Gln | Ser | Tyr | Leu | Lys | Leu | Arg | Pro | Asp | Arg | Ala | Ala |
| | 85 | | | | 90 | | | 95 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Cys | Gln | Asp | Ala | Thr | Ala | Gln | Met | Ala | Ile | Leu | Gln | Phe | Met | Ser | Ala |
| | 100 | | | | 105 | | | 110 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gly | Ile | Pro | Thr | Val | Gln | Thr | Pro | Thr | Thr | Val | His | Cys | Asp | His | Leu |
| | 115 | | | | 120 | | | 125 | | | | | | | |

| | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ile | Gln | Ala | Gln | Val | Gly | Glu | Gln | Asp | Leu | Ala | Arg | Ala | Ile | Asp |
| | 130 | | | 135 | | | 140 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ile | Asn | Lys | Glu | Val | Tyr | Asn | Phe | Leu | Gly | Thr | Ala | Ser | Ala | Lys | Tyr |
| 145 | | | | 150 | | | 155 | | 160 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Asp | Ile | Gly | Phe | Trp | Lys | Ala | Gly | Ser | Gly | Ile | Ile | His | Gln | Ile | Ile |
| | 165 | | | | 170 | | | 175 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Glu | Asn | Tyr | Ala | Phe | Pro | Gly | Ala | Leu | Leu | Ile | Gly | Ser | Asp | Ser |
| | 180 | | | | 185 | | | 190 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| His | Thr | Pro | Asn | Ala | Gly | Gly | Leu | Gly | Met | Leu | Ala | Ile | Gly | Val | Gly |
| | 195 | | | | 200 | | | 205 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Gly | Ala | Asp | Val | Val | Asp | Val | Met | Ala | Gly | Leu | Pro | Trp | Glu | Leu | Lys |
| | 210 | | | 215 | | | 220 | | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ala | Pro | Lys | Ile | Ile | Gly | Val | Lys | Leu | Thr | Gly | Lys | Leu | Ser | Gly | Trp |
| 225 | | | | 230 | | | 235 | | 240 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Thr | Ser | Pro | Lys | Asp | Ile | Ile | Leu | Lys | Val | Ala | Gly | Ile | Leu | Thr | Val |
| | 245 | | | | 250 | | | 255 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Lys | Gly | Gly | Thr | Gly | Ala | Ile | Val | Glu | Tyr | Phe | Gly | Asp | Gly | Val | Asp |
| | 260 | | | | 265 | | | 270 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Asn | Leu | Ser | Cys | Thr | Gly | Met | Gly | Thr | Ile | Cys | Asn | Met | Gly | Ala | Glu |
| | 275 | | | | 280 | | | 285 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ile | Gly | Ala | Thr | Thr | Ser | Thr | Phe | Pro | Phe | Asn | Glu | Arg | Met | Ala | Asp |
| | 290 | | | 295 | | | 300 | | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Tyr | Leu | Asn | Ala | Thr | Gly | Arg | Lys | Glu | Ile | Ala | Asp | Phe | Ala | Arg | Leu |
| 305 | | | | 310 | | | 315 | | 320 | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Tyr | Asn | His | Phe | Leu | Ser | Ala | Asp | Glu | Gly | Cys | Glu | Tyr | Asp | Gln | Leu |
| | 325 | | | | 330 | | | 335 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ile | Glu | Ile | Asp | Leu | Asn | Thr | Leu | Glu | Pro | Tyr | Val | Asn | Gly | Pro | Phe |
| | 340 | | | | 345 | | | 350 | | | | | | | |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Thr | Pro | Asp | Leu | Ala | Thr | Pro | Ile | Ser | Lys | Leu | Lys | Asp | Val | Ala | Val |
| | 355 | | | 360 | | | 365 | | | | | | | | |

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Glu Asn Gly Trp Pro Leu Glu Val Lys Val Gly Leu Ile Gly Ser Cys
 370 375 380

Thr Asn Ser Ser Tyr Glu Asp Met Glu Arg Ser Ala Ser Ile Ala Lys
 385 390 395 400

Asp Ala Met Ala His Gly Leu Lys Ser Lys Ser Ile Tyr Thr Val Thr
 405 410 415

Pro Gly Ser Glu Gln Ile Arg Ala Thr Ile Glu Arg Asp Gly Gln Leu
 420 425 430

Gln Thr Phe Leu Asp Phe Gly Gly Ile Val Leu Ala Asn Ala Cys Gly
 435 440 445

Pro Cys Ile Gly Gln Trp Asp Arg Arg Asp Ile Lys Lys Gly Glu Lys
 450 455 460

Asn Thr Ile Val Ser Ser Tyr Asn Arg Asn Phe Thr Gly Arg Asn Asp
 465 470 475 480

Ser Asn Pro Ala Thr His Ala Phe Val Thr Ser Pro Asp Leu Val Thr
 485 490 495

Ala Phe Ala Ile Ala Gly Asp Leu Arg Phe Asn Pro Leu Thr Asp Ser
 500 505 510

Leu Lys Asp Ser Glu Gly Lys Glu Phe Lys Leu Lys Glu Pro Thr Gly
 515 520 525

Lys Gly Leu Pro Asp Arg Gly Tyr Asp Pro Gly Met Asp Thr Tyr Gln
 530 535 540

Ala Pro Pro Ala Asp Arg Ser Ala Val Glu Val Asp Val Ser Pro Thr
 545 550 555 560

Ser Asp Arg Leu Gln Ile Leu Lys Pro Phe Lys Pro Trp Asp Gly Lys
 565 570 575

Asp Gly Ile Asp Met Pro Ile Leu Ile Lys Ser Leu Gly Lys Thr Thr
 580 585 590

Thr Asp His Ile Ser Gln Ala Gly Pro Trp Leu Lys Tyr Arg Gly His
 595 600 605

Leu Gln Asn Ile Ser Asn Asn Tyr Met Ile Gly Ala Ile Asn Ala Glu
 610 615 620

Asn Glu Glu Ala Asn Asn Val Arg Asn Gln Ile Thr Gly Glu Trp Gly
 625 630 635 640

Gly Val Pro Glu Thr Ala Ile Ala Tyr Arg Asp Asn Gly Ile Arg Trp
 645 650 655

Val Val Val Gly Gly Asp Asn Phe Gly Glu Gly Ser Ser Arg Glu His
 660 665 670

Ala Ala Leu Glu Pro Arg Phe Leu Gly Gly Phe Ala Ile Ile Thr Lys
 675 680 685

Ser Phe Ala Arg Ile His Glu Thr Asn Leu Lys Lys Gln Gly Leu Leu
 690 695 700

Pro Leu Asn Phe Val Asn Gly Ala Asp Tyr Asp Lys Ile Gln Pro Ser
 705 710 715 720

Asp Lys Ile Ser Ile Leu Gly Leu Lys Asp Leu Ala Pro Gly Lys Asn
 725 730 735

Val Thr Ile Glu Val Thr Pro Lys Asp Gly Ala Lys Trp Thr Thr Glu
 740 745 750

Val Ser His Thr Tyr Asn Ser Glu Gln Leu Glu Trp Phe Lys Tyr Gly
 755 760 765

Ser Ala Leu Asn Lys Met Ala Ala Ser Lys Lys
 770 775

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<210> SEQ ID NO 77
<211> LENGTH: 1464
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 77

| | | | | | | |
|-------------|-------------|-------------|------------|-------------|--------------|------|
| atggatctgg | cgaaaatcac | cgacggcttc | gtcaagcacg | agacacctgc | gtcgccctct | 60 |
| tcttgcgtcca | ccaccaacac | aggcccacc | ccagacttgt | ctccagtgac | gccctccaag | 120 |
| aatgtgaga | agcggccacg | agaggacac | cctgaagagt | cgcaacacac | gagcgccgc | 180 |
| gecaacacga | acaacaacgc | tagcgtgtct | ctcatgtcca | ccccagagcc | caagtgcgtcg | 240 |
| tctccccccg | gactgtcgca | tttgcacac | ctgatgcaaa | agtcggacac | catgtaccga | 300 |
| cagaacactca | actcggacca | gtacatctac | tcggacgagg | agaaggagaa | ccacaagact | 360 |
| tcgggcaagc | cccacacccc | ccaggtgcct | catacgccct | ccagtggtgc | gacacaacaa | 420 |
| ccccaaatatg | cattttatcc | acattccatc | acctcgtacc | cgtcgaacga | gcctcagatt | 480 |
| gacaacgcac | ggctggcgcg | ccgaaaacga | cgccgaacgt | ctcccaacgga | actcgcgtcg | 540 |
| ctggagcagg | agtttgcgg | caaccagaag | cctcccaacg | acattcgcgt | cgacattgccc | 600 |
| cgccgagtcg | acatgactga | aaaggctgtg | caggtgttgt | tccagaacaa | gcccagagc | 660 |
| gtgcgaaaga | gcatgaacaa | gagcatgacc | gatgacacct | ctttcgccga | ctcttcgttc | 720 |
| gctgaaacta | ccttgacga | gacagacggt | aactccacat | tccgtccaa | ttccaacgtc | 780 |
| agcaccagcg | taagcaacaa | gtcaatcact | tcttccatca | cagacaacaa | gtcgccctcg | 840 |
| gcacagtcaa | ccacccgcga | ctctgggtcc | aacgccaacg | ccaacgccaa | cgccaaacgccc | 900 |
| aacaacaaca | ccgcatccac | ttcctccaca | aacgactccg | aaattgcata | cgtgcccccc | 960 |
| aaaacaaaacg | gcagetcatt | ctctgttttc | gaagataccc | ccgagactcc | cgcgaaaaag | 1020 |
| aaaccccgatg | ctcccgact | gtccatgegt | ggtggaaagg | ctactgttat | ctacgcggc | 1080 |
| aagcccaagg | gtgtcacgct | gtccctggga | agacgtcttg | gggtccctgc | cacaccctcc | 1140 |
| tctcccgcca | acaacaatct | tggccctggga | ggctcgectc | tggccacatc | gtctctatg | 1200 |
| acccagcgga | ccgcgtcgca | actgaaccag | gcatctgcat | cttctccct | atcggtgttt | 1260 |
| aagtccaaatg | cttttggaaac | tgcggaggaa | agcctggctt | cgacgtcaa | gaageggctt | 1320 |
| ccgtccatgc | actacgacct | gcccgtgacc | aacaagacgt | cgtctgtgcg | ccatggcggt | 1380 |
| agctctcccg | tggtcgacgc | cggcggcgt | gaggccgagt | gtatccaa | tctctctct | 1440 |
| cttcgaaacg | gaggacgatg | gtaa | | | | 1464 |

<210> SEQ ID NO 78
<211> LENGTH: 1548
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 78

| | | | | | | |
|-------------|------------|------------|------------|------------|------------|-----|
| atgttgcgag | ccctgaatac | cgtccagcga | cttccagca | cccgagccat | gtccacccct | 60 |
| tccatttcgt | ctctgtttaa | gaacccaaat | cttctgcgaa | accaggccta | tgtcaatgg | 120 |
| cagtggtct | cctccaagac | cggagacact | ttcagcgttg | agaacccacg | cactggcgag | 180 |
| actctggggcc | aggtgcccga | gttctctgtc | gccgaggccg | atgaggctgt | ccagcacgca | 240 |
| cagactgcct | tcaagacctt | caaacatacc | actggacgag | agcgatccaa | gatgctgcga | 300 |

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| | |
|---|------|
| aagtggtacg atctgatgca ggagaatgtc ggtgatctgg ccaccctgg gactctggag | 360 |
| aacggtaagt ccctcgctga cgccaagggc gagattggct acggaggcatc tttcttcgag | 420 |
| tggttctccg aggaagctcc tcgaatctac ggagacatca ttccatccgc caaccccgcc | 480 |
| aaccgaatct acacaatcaa gcagccccatc ggagtctgctg gaatcatcac cccctggAAC | 540 |
| ttccccctcgcc ccatgatcac ccgaaaggct gctgctgtgg ttgctgtgg ctgtaccatg | 600 |
| gtgatcaagc ctgggtccga aacctcctac tctgcccattt ctctggctta cctggctgaa | 660 |
| caggccggca tccctaaggg ttttgtcaac gtggtcacta ctaagaagaa cactcgagct | 720 |
| tttggtaacg ccctgtgcga gaacccgacc gtcaaaaagg tttcttcac gggctccact | 780 |
| ggtgtcggaa agacccttat gggcgcatcg gcctccactc ttaagaagct gtccttgag | 840 |
| ctcgggtggca acgctccctt cattgtgttt gaggacgccc atattgaccc ggctgtcgac | 900 |
| ggagctattt cgtccaagtt ccgaggcact gcccagacct gtgtctgtgc aaaccgaatt | 960 |
| tatgtgcacg agagcatcgc cgagaagttt gctgagccaa tggcagccgt ggtcaaggac | 1020 |
| ttcaagggtt gaaacggctc cgaccctaac accacccatg gcccatttat ccacgaggaa | 1080 |
| gccaaggggca agatccagga gcagggttgcgatgctgtca agaagggagg aaaggtaactc | 1140 |
| attggaggct ccgacgcccc tgagatcgga aaggcctttt tccagectac cgtcatttcc | 1200 |
| ggggccaagt ctgatatgct gattgcctcc gaggagacgt ttggtcccat tgctgccatc | 1260 |
| ttccccctta agaccgacgc tgaggtcatt gagcttgcga acaaggcaga ggtcggtctg | 1320 |
| gcccggctact tctactccaa ggacgtgtac cgaatccaa aggttgcga ggctctcgag | 1380 |
| gtcggaatgg tcgggtttaa caccgggtctg atgacggagt gtgtctgtcc ctttggcggt | 1440 |
| atcaaggagt ctggctttgg ccgagaggc tccaagtagc gcctggatga ctacatggtg | 1500 |
| ctcaagacta ttgttgtgtc tggcgatcgag ccccacattc agccttaa | 1548 |

<210> SEQ ID NO 79

<211> LENGTH: 627

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 79

| | |
|---|-----|
| atgtattcat tcgacttcaa ctttgacacg gcatatccgc cacagactga atattccaaa | 60 |
| caagacgact gtctggata catgcccattc acgcctcattt acctggactg gagctcgctg | 120 |
| acattccccgc cgggttaata cgccacccatc gtcgataacg tgctcccgaa agaaccctcg | 180 |
| gagccctcgcc acgtgttttc ttcttccggaa gaagaaagcc cctactttt cgacgaatac | 240 |
| tgcaccatcc cctctctggc cgaccagtc aaagaaaacc ccaacatttg ggccatggca | 300 |
| aacaccgtca agaaaggaggc ctacgtgtgt agccactgca ctaagcaggc caccggcgtc | 360 |
| aagttcaaaa ccatggtcga ctttgcacc caccctcgact cgcatttcata tgaccgaagc | 420 |
| tgc当地atgcg cccgacacaaa atgtccctgg tccattgtgg gcttcttcac tcgatcgaa | 480 |
| atgc当地gaac acacaaactc ggtccatcgaa caaacaccct tcacatgca aatctgtgac | 540 |
| cgccgggtttg tacgagaaga ctctctcaaa cggcatgtca aactactcca catttctccc | 600 |
| ctcaaaaacca gacgaaagag tacctga | 627 |

<210> SEQ ID NO 80

<211> LENGTH: 1683

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

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<220> FEATURE:

<223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 80

| | | | | | | |
|------------|------------|-------------|------------|------------|-------------|------|
| atgcaccacc | acctaacc | caaggcgctc | tttctggtg | agtatggcg | acagaaatgg | 60 |
| acggaggaac | gtggcagagc | cgattgacca | gccacgcagg | ccgaccaagc | cccattgagt | 120 |
| gagccattgg | acgtccttgg | cccgaataga | cgtctctcc | caggttgcc | ggaaaaacga | 180 |
| gctgttatat | ccgaacgagc | tgttgtgcc | caaaaagcc | cctactaacc | cccaggccga | 240 |
| aaggagagca | cctctcccc | gacacaagcc | gcgtccggct | ccggagccgt | gtctccaggc | 300 |
| cgacacctgg | attcgtccac | caacgtcgaa | gatgtggatg | agcttgacgg | agacggccag | 360 |
| aacatcatca | tggaaattat | cgcgcagctg | cgaccggcg | ctgatctgtc | tcgaatcaca | 420 |
| cttccccact | tcatttcgta | gctaaagtcc | atgctcgagc | gaatcacaaa | ctccctgcag | 480 |
| caccccatat | atgtcattga | ggcccacgcc | accaaggacc | ccatgcagcg | gttcatccaa | 540 |
| gtggtaaagt | ggtaccactc | cggctggcac | atcacccccc | aggccgtcaa | aaagccccctg | 600 |
| aaccccattc | tcggcgagtt | cttcacatgc | tactggact | acgacgacgg | ttccacggg | 660 |
| tactacatct | ccgagcagac | ctccaccac | cctcccaagt | catcctactt | ttatcatgtc | 720 |
| cctgagcaca | acatccgagt | cgacggtaca | ctggctccca | agtcccgtt | cctggtaac | 780 |
| tcagctgctt | ctctcatgga | ggggccacc | attctcaagt | tcctggacat | tgttagatgcc | 840 |
| aagggegctc | ccgaggagta | cgaaatcact | tcgcccata | cctacgcccc | aggtatttctc | 900 |
| tttgaacggc | tcaagtacga | gtactgcac | cactcgatca | tcaagtgtcc | cgctctggac | 960 |
| ctgactctgg | acctggactt | caaggccaag | ggcttcattt | ccggatata | caatgccttc | 1020 |
| gagggccaga | tcaagaagat | ctccaccggc | gagggctttt | acgatgttta | tggaaagtgg | 1080 |
| gatgaaatca | tcgagctcaa | gaacctcaag | accggcgaga | agtccgtgt | gtttgacgtg | 1140 |
| actaaggccg | ccctgcaccc | tcccaaggtg | cgaccatcg | ctgagcaggc | cgccaccgag | 1200 |
| tcccgacgac | tgtggagcc | cgtcacccgac | gctcttgcta | agcgagacca | caccgttgct | 1260 |
| accgacgaaa | agtcaagat | tgaggacaaa | cagcgaacgc | tggcaagga | gcpagaagag | 1320 |
| cacggcgtca | agtccctgcc | caaactgttc | aagccgcccc | cgcctccct | ggacttcatt | 1380 |
| ctgtataagg | atctgcacgg | cactccgaa | gagatcacca | aggagattct | cagcatagtc | 1440 |
| cccattctgc | ccggccaaca | gttcaccaag | gactttgaaa | tgtccggcga | gaagaatac | 1500 |
| aagctggaga | agagcggcca | ggccacgcgc | gagactcagc | ccacccgcac | gaccactgcg | 1560 |
| gtgtcccccc | aagcaggcgc | tgtccccaca | accctgtct | acggccagac | tccctggcc | 1620 |
| aagacttctg | atcttcagga | ggcttcccc | accgaagagg | acgagttcca | cgacgcccag | 1680 |
| tag | | | | | | 1683 |

<210> SEQ_ID NO 81

<211> LENGTH: 5510

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Synthetic polynucleotide

<400> SEQUENCE: 81

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| atgacaagtgt | atgcgataaaa | cgccatggaa | aacgacagta | cgacgggtgt | agaggtggaa | 60 |
| acgacatttg | tgaacgataa | cgtggtccgt | ggcttcctcg | atgttgacag | tgatacgtg | 120 |
| ccagacgtcc | aaggactcct | tccactggtc | caagtgcagc | tggtggcgga | tatctcaaga | 180 |

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| | | | | | | |
|-------------|-------------|-------------|-------------|-------------|-------------|------|
| gagatgctgg | aggcgaaga | agtgcgtggaa | atcacccgatc | cagagtcaca | tggcgtaaaa | 240 |
| aataccgaag | cagggtacga | aacgaactca | cgtgaccccc | tgcgtcgcttc | tgcgcctgct | 300 |
| accctggttc | ctaacgagag | cacatttagag | attcatgtca | cgcggaaagta | caccaccaag | 360 |
| gacaagaaac | gaggccgcaa | aaagaccaag | aaggacgaag | attgggttgg | aacatgcttg | 420 |
| ggtgtgttgc | aactaggaaa | cgtggaaacc | agtgaccacg | tgcttaccgc | tttgaacag | 480 |
| gtctttccg | tagccaaatt | taaccgcgaa | aatcgagtca | gtgtcttttc | agtgaatcct | 540 |
| cacgtcactg | ttaccaaaaa | caatgggtt | tacagcattt | ccatcacttt | tggagtcttt | 600 |
| gcaaggcctt | ttgtatggca | cgtcaaccct | gagatccata | tggcagggtca | cctcaacatt | 660 |
| gtgaatgtca | tccgacagtt | cctggggcgt | actaagataa | aacagctaca | taagaacgac | 720 |
| tatgtgactc | ctgaataactt | ctacgagtgc | ctggaaactca | aggatgatac | cgagggttag | 780 |
| atcaacagag | atcttcagcc | ggaagggtg | agatcaaaac | ttttggat | ccagcttga | 840 |
| actgtgggtt | gggttctgga | tagagaaaag | ggagaatcgc | gtgagaagac | gatagaggga | 900 |
| atcccttcac | catggaaacg | gttcagggtct | catggtatca | actgggttgg | tgatTTTGT | 960 |
| gggtctcaaca | ttggtcctga | gaaggaggtg | atggagattt | tgacacgaga | tacgaaacca | 1020 |
| acaactgagg | accccgagat | tcaaggcgt | tcacgtgacg | cagatTTAA | ggctggat | 1080 |
| ggacttattg | ctgatgaaat | gggtcttgga | aagacagttg | agctactagc | tgttagtcctg | 1140 |
| aataacccca | gacctgaattt | tccaccgc | acacactacg | atctgtactc | tgacagagac | 1200 |
| gtgttaccta | ccaagacgc | tctcatTTA | tgtcctgcca | gtatcgtca | acagtggatt | 1260 |
| gtcgaggta | ctaaacatgc | tcccagtctc | tctgtctttc | tgtacactgg | tcgagcagct | 1320 |
| ttggatgctc | aaagagagaa | ggaaggta | cccgataccg | atattgaggt | tggatttgac | 1380 |
| tcagatactg | attcagaagg | cccttttgg | tcaaaacatg | cacaatttct | ctctcagttc | 1440 |
| gacattgttag | tcacatccta | tgaagttgc | tctcgcgagg | ttgccaacgc | tctttacaac | 1500 |
| cctctgagag | gtcgtgtAAC | tcgcaccaag | acgaagctaa | agtcgaaaga | tacccgagat | 1560 |
| gtcgatctcg | tgcaagaccg | gtttccctc | caatctccac | ttagtcagct | tcagttctgg | 1620 |
| cgtgtgtttc | tggacgagg | tcaatgggt | gaaacacgg | tctccaacgc | agctgttgta | 1680 |
| gtcgttatta | ttccccgg | gcatgcattt | ggagtcagtg | gtactcctat | aaagaaggc | 1740 |
| atgcctgact | tacttggcat | tggtgtgtt | ttgagatgt | aaacccggca | gttttatgga | 1800 |
| agaagtgatt | gtgagtatac | taaaggaaca | gtcagagtgg | catgtgacaa | aaaaacaaaa | 1860 |
| aaccatatgg | ctcaatgaag | ggctacgact | aacacagatg | cgtataacta | ctcttggca | 1920 |
| aacggatacc | ttagcacaac | tatgacagca | tctggagtaa | gtcatcaaa | acactggag | 1980 |
| atgctcatgc | ttgacaagcc | tcgggttgc | gacgttattc | gtcaaatgtc | tattcgacat | 2040 |
| actaaggcgc | aggcagaga | tcaacttagta | ttgcctcctc | aggaaagaca | ccatgtgaga | 2100 |
| ctcagattca | atctagtcga | ggaagaaaac | tacggacacc | tgcgtgaagg | tgttgagagt | 2160 |
| gccgtcagtg | aggcagttgc | tagttcttc | atgagagaag | agagggaaac | tacacgtgag | 2220 |
| gcagctgtgg | tggataggt | tggcggtctg | ccttcaagtg | tcaactcccc | tgtgagcaac | 2280 |
| agacctagag | gcactttcaa | catcgaggc | tctaattccct | atgcttagtat | catggcgaat | 2340 |
| atcaacaaca | cagtcatgt | acctgaaattt | gagattgatc | ccagtatcac | ttcttagtgaa | 2400 |
| gagggtgacg | gcacaacatgt | ctacactacc | tggcggttgc | ctgttagacac | gtatgggtt | 2460 |
| gagtctagcg | gtacagctgc | tagtagcacc | gtgcgtgacg | gcgtatgataa | cgctcaatct | 2520 |
| cccacatctg | atacagctag | caacactgac | atcaatgtta | gtgctattcc | cgatatacg | 2580 |

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| | |
|---|------|
| gtatccccga ctgccacccc tacagcctcc accagatccc aaaatggaac ttctgctcct | 2640 |
| ccagcatctt ccgctcctgc ggatttaaca acagcaacc tcttcctcg gctgttacgg | 2700 |
| ttacgacaaa cctgtgccca tcctcgagtc ggttctggta acaagaaggc tctcgaaac | 2760 |
| ggtattcttc aaactgtcag tcacgtgctg gacgcatgt gcgaccagc gctcaccag | 2820 |
| ctgctgaacg acgagcgaag tctgttgtc gaagagctgg agaaggcacg agttcacgag | 2880 |
| ttaaacaac aaccagacat tggactcaca gtgcttcagt cacgtgttcc tgaagtgcag | 2940 |
| gttcgaactg gtgagatccg agatatggct gttgctgccc ctacgcccgt tgctatgaag | 3000 |
| aagaaggagg taatttccga gtggaagcgtt attggtgagg ttgataacaa ggcgaagttt | 3060 |
| gaggagagtg atgacgggtc tgctaattgtt aagaaagtca aggtcgaaaa agaggagaag | 3120 |
| gaggaagaag tggcaaaagga ggagggttcc gaagattttt aaatggaggg aactgagaac | 3180 |
| aactccatTT ttggagctcc aactgtttt ctgggctctg attcggagtc tgagagcact | 3240 |
| ggtaagatgt ccaaaccatt acaaaagtac ctgaacaact cggaggaact tcagacggag | 3300 |
| aaggagcga aacaggcttt tctgcaccgg tacaggagct ggtggatct tatgcacgg | 3360 |
| tactatTTT tcattgtcac ttttcatttc caagttggag aagcgtgagt atgacaaga | 3420 |
| tttgtaatga ogtgggtgtt ctactgggtt catgagaggt catgagacat actaacacag | 3480 |
| taaaaaaaatgt gctgaggaga agaaaagaaaa agaggatggg aaggacgtg aagagaagga | 3540 |
| agatgaagag aaggaagaga ttgaggtcaa gaaagaggag gatgaaggga ccaagagtga | 3600 |
| cgagttagtga tagagatatac atgagtggca gaataacttg tgccattcgc tcctttatg | 3660 |
| tatatgtgtta ctaacacagt ctggaaacgc actattacac gctggcagaa caaatccgaa | 3720 |
| cccaagctact tcaacgcctt attgagagag tagaccaaga cgtgggtcga cttgaacggg | 3780 |
| ccaaggagct ggagatgggtt cagatccctg ttgataacctt gactcgagat ctgtacagg | 3840 |
| cTTCTCTTT ctttgaggca cgttttccgg gtctactcga gatcatcaac caacatccg | 3900 |
| aatatcttga agaatggatg accagagttc gagagctgtt ggttgcacgt gacgagaagg | 3960 |
| acgtgaaaga aacagataag aagaagaata aaggagatgt cgagaaagt gaaggcgaaa | 4020 |
| acactgtatcc ttatgtttct ggatttagaca accaacaata tgcgttggac taccttgcgt | 4080 |
| ctatatcgta cctgtgtcaa ctcagagatg aagctctcaa tgccaaagact acggcctcag | 4140 |
| cagccgacaa gatccaagtt aacttgggtt accacaatga ctacgaagaa gagcttaccg | 4200 |
| atcttcagggt ggcctcaag gaagctctgg acgcttgcata tgtgagtccc actcttgg | 4260 |
| ccctcaaacc tatacggtgt gctctgaaga cggactctgg agctgtttca ttgtcaattt | 4320 |
| acaacccgaa atggcctccc aagttgtgtt ccaagctcaa tccgatcgaa aagacagtt | 4380 |
| cctcgacaaac caaggcttgc agagacctgt tgcgtcgatc tagaagctgtt ttcaactcga | 4440 |
| aggttgtcta ttacaagcag ctgcagcaac tgtctgacaa tgtgagcgt ctggaggaa | 4500 |
| tcatcgagcc tgggttatgtc acactggaa gcctgaacgc caaaataaac catctcgatc | 4560 |
| ctttaatcaa gcgtacaaag ggccgaatca catacttaca gagtctcaa ggtgtatgt | 4620 |
| acacaactgg agttccaaac atgactggaa ttctataatgtt gttgttcatc tgcaggatg | 4680 |
| attatattat cgtggatcc atcactgtct gtggccatta cttttgcaga aactgcctgg | 4740 |
| aagagtgggt gcaagacacat aatacggtc caatgtcaca gactgtatgt tcccgcgacg | 4800 |
| atgtgttctc ttccacccaa caggacaagg aagacaagtc acgtgcaggat tcttcgtcg | 4860 |
| ctcgatcaa tcaagatgac gccattggag caatgtatgc ggcgtgtcg gaggacactc | 4920 |

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| | |
|---|------|
| aacagttgat gagcaaacag agcatcaaga gtgcgtatgg cacaaagatt gaccacgtta | 4980 |
| tcaagtatat caagatgctc actcatcggt ctcctggcac tcagattgtc atcttttctc | 5040 |
| agtgggcaga gatttcaca ttgttagctt cagccctcac tgagaacaag attgcatacg | 5100 |
| cggagccgaa aacactgtatg tctttcttgc aatcggaga agtcacgtgt ttccctgtga | 5160 |
| acgcaaaagg ccagtttactt ggcctactt ttgtaaatgc cactcacgtc attctatgcg | 5220 |
| agccccattt ctacgtgtctt cttgaggctc aggccatcaag tcgaatccac cgaatgggcc | 5280 |
| agactcagac tacccacgtg actatcttca ctatggccga tactgttga gaagagggtc | 5340 |
| tgcgttgc tattaacaag cggttgaaaa gtatggacgg tgatgagacg tttgaggaga | 5400 |
| atgaatctcg acatgtgaca tcaggagtgg gtgcgtcgc caccgataaa tccggagagg | 5460 |
| tggtcaaccg tcaggatatg tgggacgctt tgttcccag tgacgggtaa | 5510 |

What is claimed is:

1. A method of producing a lipid, lipid precursor, or oleochemical comprising:
 a) culturing a genetically modified yeast cell in a growth medium; and
 b) isolating said lipid, lipid precursor, or oleochemical, wherein the dry weight of said genetically modified yeast cell comprises greater than 60% wt/wt lipids, lipid precursors, and oleochemicals; and
 wherein said genetically modified yeast cell comprises (i) a recombinant acyl-CoA:diacylglycerol acyltransferase 1 (DGA1) gene and a UGA2 succinate semialdehyde dehydrogenase (UGA2) gene comprising a mutation, wherein said mutation results in a loss of function of succinate semialdehyde dehydrogenase of 20% or more compared to succinate semialdehyde dehydrogenase encoded by UGA2 without the mutation; or (ii) a recombinant acyl-CoA:diacylglycerol acyltransferase 2 (DGA2) gene and a UGA2 succinate semialdehyde dehydrogenase (UGA2) gene comprising a mutation, wherein said mutation results in a loss of function of succinate semialdehyde dehydrogenase of 20% or more compared to succinate semialdehyde dehydrogenase encoded by UGA2 without the mutation.
 2. The method of claim 1, wherein said growth medium comprises a majority carbon source selected from the group consisting of glucose, glycerol, xylose, fructose, mannose, ribose, sucrose, and lignocellulosic biomass.
 3. The method of claim 1, wherein said growth medium comprises lignocellulosic biomass as the majority carbon source.
 4. The method of claim 1, wherein said growth medium comprises cobalt, iron, magnesium, potassium, zinc, nickel, molybdenum, manganese, copper, or boron.
 5. The method of claim 1, wherein said growth medium comprises 5.77×10^{-5} M to 1.73×10^{-4} M cobalt, 0.001 M to

20 0.003 M magnesium, 4.52×10^{-5} M to 1.35×10^{-4} M potassium, 4.05×10^{-5} M to 1.22×10^{-4} zinc, 3.55×10^{-5} M to 1.06×10^{-4} manganese, 9.07×10^{-5} M to 2.91×10^{-4} boron, 3.76×10^{-5} M to 1.10×10^{-5} molybdenum, 2.28×10^{-5} M to 6.84×10^{-5} nickel, 3.60×10^{-5} M to 1.08×10^{-4} iron, or 4.70×10^{-5} M to 1.41×10^{-4} copper.

25 6. The method of claim 1, wherein said genetically modified yeast cell comprises a recombinant Lipid synthesis regulator (MGA2) gene, a genetically modified Lipid synthesis regulator (MGA2) gene, a recombinant Leucine Biosynthesis gene (LEU2), a genetically modified multifunctional enzyme (MFE1) gene, a genetically modified PEX10 Transcription Factor (PEX10) gene or a recombinant AMP Deaminase (AMPD) gene.

30 7. The method of claim 1, wherein said genetically modified yeast cell comprises a genetically modified multifunctional enzyme (MFE1) gene and a genetically modified PEX10 Transcription Factor (PEX10) gene.

35 8. The method of claim 1, wherein said genetically modified yeast cell comprises a recombinant Leucine Biosynthesis gene (LEU2), a genetically modified multifunctional enzyme (MFE1) gene and a genetically modified PEX10 Transcription Factor (PEX10) gene.

40 9. The method of claim 1, wherein said genetically modified yeast cell comprises a genetically modified multifunctional enzyme (MFE1) gene, a genetically modified PEX10 Transcription Factor (PEX10) gene and a recombinant AMP Deaminase (AMPD) gene.

45 10. The method of claim 1, wherein said genetically modified yeast cell comprises a recombinant Leucine Biosynthesis gene (LEU2), a genetically modified multifunctional enzyme (MFE1) gene, a genetically modified PEX10 Transcription Factor (PEX10) gene and a recombinant AMP Deaminase (AMPD) gene.

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