



(43) International Publication Date
30 June 2016 (30.06.2016)

(10) International Publication Number
WO 2016/101960 A2

(51) International Patent Classification:

C12N 1/18 (2006.01) C12N 15/04 (2006.01)
C12N 15/01 (2006.01) C12C 11/00 (2006.01)

(21) International Application Number:

PCT/DK2015/050413

(22) International Filing Date:

22 December 2015 (22.12.2015)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

PA 2014 70825 23 December 2014 (23.12.2014) DK
PA 2015 70351 8 June 2015 (08.06.2015) DK

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(81) Designated States (unless otherwise indicated, for every
kind of national protection available): AE, AG, AL, AM,

AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY,
BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM,
DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT,
HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KN, KP, KR,
KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG,
MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM,
PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC,
SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN,
TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every
kind of regional protection available): ARIPO (BW, GH,
GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ,
TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU,
TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE,
DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU,
LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK,
SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ,
GW, KM, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:

— of inventorship (Rule 4.17(iv))

Published:

— without international search report and to be republished
upon receipt of that report (Rule 48.2(g))

— with sequence listing part of description (Rule 5.2(a))

(54) Title: YEAST FOR PREPARING ALCOHOLIC BEVERAGES

(57) Abstract: The invention relates to yeast cells with useful characteristics, including being capable of utilizing panose as sole carbon source and/or capable of utilizing one or more dipeptides as sole nitrogen source. The invention also relates to yeast cells with useful genotypes including comprising at least 4 allelic genes encoding IMA1p and/or at least two allelic genes encoding IMA5p.



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YEAST FOR PREPARING ALCOHOLIC BEVERAGES

Background of the invention

5 Alcoholic beverages are frequently prepared by fermentation of a carbohydrate rich liquid with yeast. For example, beer is prepared by fermenting wort with yeast. Wort contains a number of compounds, which can be utilized by yeast. For example wort is rich in sugars, in particular maltose and as well as in amino acids and small peptides. Conventional yeast can utilize maltose and thus conventional yeast can ferment
10 maltose to produce ethanol. However, wort also contain other carbohydrates in addition to maltose, some of which cannot be utilized by conventional yeast, and in particular not by lager yeast.

Lager yeast in general differs from ale yeast in several ways. Lager yeast belong to the
15 species *S. pastorianus*. Frequently, lager yeast is also referred to as "bottom-Fermenting Yeast" because they settle at the bottom during fermentation. Furthermore, lager yeast strains are best used at temperatures ranging from 7 to 15°C. In addition lager yeast is capable of using melibiose as the sole carbon source and cannot grow at 37°C.

20 In contrast, ale yeast belong to the species *S. cerevisiae*. Frequently, ale yeast is also referred to as "top-Fermenting Yeast", because they often rise to the surface during fermentation. Furthermore, Ale yeast strains are best used at temperatures ranging from 10 to 25°C, though some strains will not actively ferment below 12°C. In addition
25 ale yeast is not capable of using melibiose as the sole carbon source and can grow at 37°C.

Other yeast can also be employed in beer brewing, e.g. *Saccharomyces diastaticus*. *Saccharomyces diastaticus* belongs to the *Saccharomyces cerevisiae* specie variety (var.) *diastaticus* and has the particularity of having glucoamylase enzyme activity
30 encoded by at least one of the following genes *STA1*, *STA2* or *STA3* enabling the yeast to utilize starch as sole carbon source. The *STA* genes are in general absent in *S. cerevisiae* or *S. pastorianus* or other *Saccharomyces* species strains analyzed, but are present in the subgroup of *S. cerevisiae* var. *diastaticus*.

Summary of the invention

5 There is a need for improved yeast strains, which have characteristics of both lager beer (e.g. *S. pastorianus*) as well as of ale yeast (e.g. *S. cerevisiae*). In addition, there is a need for yeast strains, which can utilize as many different energy sources as possible. In particular, there is a need for yeast, which can utilize sugars present in wort, which are not maltose and yeast which can utilize amino acids and peptides to a high degree.

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Interestingly, the invention provides a hybrid yeast, which has several important characteristics of lager yeast, but which at the same time can utilize a lot of different energy sources present in wort.

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Accordingly, it is an aspect of the invention to provide a yeast cell having at least one of the following characteristics:

- I. Capable of utilizing isomaltose as sole carbon source;
- II. Capable of utilizing panose as sole carbon source.

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In addition to abovementioned characteristics I and II the yeast cell according to the invention may have additional characteristics, for example one or more of the following characteristics:

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- III. Capable of utilizing one or more dipeptides as sole nitrogen source;
- IV. capable of utilizing one or more tri-peptides as sole nitrogen source;
- V. capable of reducing the level of one or more amino acids to no more than 10% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells;
- VI. Capable of generating at least 4.7 promille ethanol per °Plato, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec; and/or
- 30 VII. Capable of fermenting sugar with a real degree of fermentation of at least 70, when said yeast cell is added to a wort composition having a

sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

5 It is also an aspect of the invention to provide a yeast cell having at least one of the following characteristics:

- II. Capable of utilizing panose as sole carbon source;
- III. Capable of utilizing one or more dipeptides as sole nitrogen source.

10 It is also an aspect of the invention to provide a yeast cell having at the characteristic:
II. Capable of Capable of utilizing panose as sole carbon source

In addition to abovementioned characteristics II and/or III the yeast cell according to the invention may have additional characteristics, for example one or more of the following characteristics:

- 15 I. Capable of utilizing isomaltose as sole carbon source;
- IV. capable of utilizing one or more tri-peptides as sole nitrogen source;
- V. capable of reducing the level of one or more amino acids to no more than 10% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells;
- 20 VI. Capable of generating at least 4.7 promille ethanol per °Plato, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec; and/or
- VII. Capable of fermenting sugar with a real degree of fermentation of at least 70, when said yeast cell is added to a wort composition having a
- 25 sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

It is also an aspect of the invention to provide methods for producing a beverage, said methods comprising the steps of

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- I. Providing a starting liquid
- II. Providing a yeast cell according to the invention
- III. Fermenting said starting liquid with said yeast cell.

Description of drawings

Figure 1 shows growth of various yeast strains in defined medium with 2 g/L panose as sole carbon source. The data shown is representative of biological replicates. Panel A) shows growth of Ale yeast 1, Hybrid yeast 1, Hybrid yeast 4 and Lager yeast 1. Panel B) shows growth of Ale yeast 1, Hybrid yeast 7 and Lager yeast 2. Panel C) shows growth of *S. diastaticus* and Hybrid yeast 8.

Figure 2 shows growth of yeast in defined medium with 2 g/L isomaltose as sole carbon source. The data shown is representative of biological replicates. Panel A) shows growth of Ale yeast 1, Hybrid yeast 1, Hybrid yeast 4 and Lager yeast 2. Panel B) shows growth of Ale yeast 1, Hybrid yeast 7 and Lager yeast 1. Panel C) shows growth of *S. diastaticus* and Hybrid yeast 8.

Figure 3 shows growth of yeast cells in Bioscreen C MBR in defined medium with 2 g/L melibiose as sole carbon source. Panel A shows growth of Hybrid yeast 1 and Hybrid yeast 4. Panel B shows growth of Hybrid yeast 7.

Figure 4 shows an NMR analysis of single sugars in final bottled beer brew comparing beer made with Lager yeast 1 and Hybrid yeast 1.

Figure 5 shows a protein alignment of *DAL5* from Ale Yeast 1, Lager yeast 1 and Hybrid yeast 1. The sequence of *DAL5* of Hybrid yeast 1 is denoted Sc_*DAL5*_Hybrid_1 (SEQ ID NO:6).

Figure 6 shows a protein alignment of *UBR1* encoded by *Sc* alleles of *UBR1* illustrating the presence of Lager yeast 1 *Sc* allele in Hybrid yeast 1 while Ale yeast 1 is truncated. Only part of the alignment is shown; residues in black shade differ from Hybrid yeast 1 sequence.

Figure 7 shows a protein alignment of *UBR1* encoded by *nonSc* alleles of *UBR1* illustrating the presence of Lager yeast 1 *Sc* allele in Hybrid yeast 1.

Figure 8 shows a protein alignment of IMA1p encoded by *IMA1* short alleles. The IMA1p encoded by the *IMA1* short alleles found in Hybrid yeast 1 are denoted IMA1_Sc_allele_short_A_Hybrid_1 and IMA1_Sc_allele_short_B_Hybrid_1, respectively.

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Figure 9 shows a protein alignment of IMA1p encoded by *IMA1* long alleles. Figure 9A shows an alignment of IMA1p encoded by long alleles from Ale yeast 1, Lager yeast 1 and Hybrid yeast 1. The IMA1p encoded by the *IMA1* long alleles found in Hybrid yeast 1 are denoted LONG_IMA1_A_Hyb1_pl17 and LONG_IMA1_B_Hyb1_pl18, respectively. Figure 9B shows an alignment of IMA1p encoded by long alleles from Ale yeast 1, Lager yeast 2, Hybrid yeast 4 and Hybrid yeast 7.

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Figure 10 shows a protein alignment of IMA5p encoded by *IMA5-like*. The IMA5p encoded by the *IMA5-like* found in Hybrid yeast 1 are denoted ScIMA5_Hybrid1_pl1 and non-ScIMA5_Hybrid1, respectively.

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Figure 11 shows a protein alignment of AGT1 encoded by *Sc* alleles of AGT1. Figure 11A shows an alignment of AGT1 encoded by *Sc* alleles of AGT1 from Lager yeast 1, Ale yeast 1 and Hybrid yeast 1. The AGT1 encoded by the *AGT1* found in Hybrid yeast 1 are denoted Sc_AGT1_Hybrid1_pl37, Sc_AGT1_Hybrid1_pl38 and Sc_AGT1_Hybrid1_pl39, respectively. Figure 11B shows an alignment of AGT1 encoded by *Sc* alleles of AGT1 from Lager yeast 2, Ale yeast 1, Hybrid yeast 4 and Hybrid yeast 7.

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Figure 12 shows a protein alignment of AGT1 encoded by *non-Sc* alleles of AGT1. Figure 12A shows an alignment of AGT1 encoded by *non-Sc* alleles of AGT1 from Lager yeast 1 and Hybrid yeast 1. The AGT1 encoded by the *AGT1* found in Hybrid yeast 1 is denoted Non-Sc_AGT1_Hybrid1. Figure 12A shows an alignment of AGT1 encoded by *non-Sc* alleles of AGT1 from Lager yeast 1, Lager yeast 2, Hybrid yeast 1, Hybrid yeast 4 and Hybrid yeast 7.

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Figure 13 shows growth of yeast in defined medium with 2 g/L maltotriose as sole carbon source. The data shown is representative of biological replicates.

Figure 14 shows growth of yeast in defined medium with 2 g/L maltulose as sole carbon source. The data shown is representative of biological replicates.

5 Figure 15 shows growth of yeast in defined medium with 2 g/L kojibiose as sole carbon source. The data shown is representative of biological replicates.

Figure 16 shows the apparent extract as a function of time during fermentation of wort with Lager yeast 2, Hybrid yeast 4 and Hybrid yeast 7.

10 Detailed description of the invention

Definitions

15 As used herein, "a" can mean one or more, depending on the context in which it is used.

The term "AE" as used herein is an abbreviation of "Apparent Extract". The "apparent extract" is a measure of the density of beer wort in terms of the percentage of extract by weight and is expressed in the Plato scale. It is the final gravity or specific gravity
20 measured at the end of beer fermentation. Gravity in the context of alcoholic beverages refers to the relative density of the liquid compared to water. The more sugars dissolved in the wort the higher the density of the wort.

25 Amino acids may be named herein using the IUPAC one-letter and three-letter codes.

The term "beer" as used herein refers to a beverage prepared by fermentation of wort. Preferably, said fermentation is done by yeast.

30 The term "carbon source" as used herein refers to any organic molecule, which can provide energy to yeast and provide carbon for cellular biosynthesis. In particular, said carbon source may be carbohydrates, and more preferably, the carbon source may be mono- and/or disaccharides.

The term "cells in suspension" is used herein in relation to incubation of cells in a liquid medium in a container. "Cells in suspension" are cells, which have not sedimented to the bottom of the container after incubation, but which float freely in the liquid medium. Cells in suspension can be determined by taking a sample of the liquid medium from the upper part of the container, and counting the cells therein.

The term "diacetyl in spec." refers to the level of diacetyl being below a predefined threshold, which is set at a level below the threshold considered off-flavor in lager beer. Preferably, the diacetyl is considered to be in spec when the level of diacetyl is at the most 30 ppb.

By "encoding" or "encoded", in the context of a specified nucleic acid, is meant comprising the information for translation into the specified protein. A nucleic acid or polynucleotide encoding a protein may comprise non-translated sequences, e.g. introns, within translated regions of the nucleic acid, or may lack such intervening non-translated sequences, e.g. in cDNA. The information by which a protein is encoded is specified by the use of codons.

As used herein, "expression" in the context of nucleic acids is to be understood as the transcription and accumulation of sense mRNA or antisense RNA derived from a nucleic acid fragment. "Expression" used in the context of proteins refers to translation of mRNA into a polypeptide.

The term "gene" means the segment of DNA involved in producing a polypeptide chain; it includes regions preceding and following the coding region (promoter and terminator). Furthermore, some yeast genes also comprise introns although only 5% of the genes in the *S. cerevisiae* genome comprise introns. After transcription into RNA, the introns are removed by splicing to generate a mature messenger RNA (mRNA).

The term "growth" as used herein in relation to yeast, refers to the process by which a yeast cells multiply. Thus, when yeast cells are growing, the number of yeast cells increases. The number of yeast cells may be determined by any useful method, e.g. by determining the OD (620 nm). Increase in OD (620 nm) corresponds to an increase in the number of yeast cells. Conditions allowing growth of yeast are conditions allowing

yeast cells to increase in number. Such conditions in general require the presence of adequate nutrients, e.g. a carbon source and an nitrogen source as well as an adequate temperature, which typically is in the range of 5 to 40°C.

5 The term "nitrogen source" as used herein refers to any organic nitrogen containing molecule and/or to ammonium containing molecules. In particular, said nitrogen source may be an organic nitrogen source, for example peptides, amino acids, and/or other amines. The nitrogen source may also be ammonium. Thus, N₂ is not considered a "nitrogen source" herein.

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The term "malt" refers to cereal grains, which have been malted. Malting is a special form of germination of cereal kernels (e.g. barley kernels) taking place under controlled environmental conditions – including, but not limited to steep tanks and germination boxes of the malting factory. In general malting involves steeping said kernels, followed
15 by germination. The malting process may be stopped by drying of the cereal kernels (e.g. barley kernels), for example, in a kiln drying process, which is usually performed at elevated temperatures. Malt may be processed, for example, by milling and thus referred to as "milled malt" or "flour".

20 "Mashing" is the incubation of milled malt in water. Mashing is preferably performed at a specific temperature, and in a specific volume of water. The temperature and volume of water are of importance, as these affect the rate of decrease of enzyme activity derived from the malt, and hence especially the amount of starch hydrolysis that can occur; protease action may also be of importance. Mashing can occur in the presence
25 of adjuncts, which is understood to comprise any carbohydrate source other than malt, such as, but not limited to, barley, barley syrups, or maize, or rice – either as whole kernels or processed products like grits, syrups or starch. All of the aforementioned adjuncts may be used principally as an additional source of extract (syrups are typically dosed during wort heating). The requirements for processing of the adjunct in the
30 brewery depend on the state and type of adjunct used, and in particular on the starch gelatinization or liquefaction temperatures. If the gelatinization temperature is above that for normal malt saccharification, then starch is gelatinized and liquefied before addition to the mash.

The term "Plato" as used herein refers to density as measured on the Plato scale. The Plato scale is an empirically derived hydrometer scale to measure density of beer or wort in terms of percentage of extract by weight. The scale expresses the density as the percentage of sugar by weight.

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By the term "wort" is meant a liquid extract of malt, such as milled malt, or green malt, or milled green malt. In barley brewing, wort may also be prepared by incubating an extract of un-malted barley with an enzyme mixture that hydrolyzes the barley components. In addition to said malt or barley-derived extracts, the liquid extract may be prepared from malt and additional components (e.g. adjuncts), such as additional starch-containing material partly converted into fermentable sugars. The wort is in general obtained by mashing, optionally followed by "sparging", in a process of extracting residual sugars and other compounds from spent grains after mashing with hot water. Sparging is typically conducted in a lauter tun, a mash filter, or another apparatus to allow separation of the extracted water from spent grains. The wort obtained after mashing is generally referred to as "first wort", while the wort obtained after sparging is generally referred to as the "second wort". If not specified, the term wort may be first wort, second wort, or a combination of both. During conventional beer production, wort is boiled together with hops, however the present invention provides methods for reducing boiling or avoiding boiling of wort. Wort without hops, may also be referred to as "sweet wort", whereas wort boiled/heated with hops may be referred to as "boiled wort".

The term "yeast cell capable of utilizing XX" as used herein refers to a yeast cell, which can take up and degrade XX.

The term "yeast cell capable of utilizing XX as sole carbon source" as used herein refers to a yeast cell, which can grow on a medium containing XX as the only carbon source. Thus, said medium preferably does not contain any other carbohydrates apart from XX.

Yeast cell

The present invention relates to a yeast cell having at least one of the characteristics I, II, III, IV, V, VI, VII and XI described herein below.

5 In particular it is preferred that said yeast cell at least has characteristics I and II described herein below.

It is also preferred that said yeast cell has at least characteristic II described below. It is also preferred that the yeast cell has at least characteristics II and III described below.

10 Characteristic I may be any of the characteristics I described in the section "Characteristic I" herein below. In particular characteristic I may be that the yeast cell is capable of utilizing isomaltose as sole carbon source.

15 Characteristic II may be any of the characteristics V described in the section "Characteristic II" herein below. In particular characteristic II may be that the yeast cell is capable of utilizing panose as sole carbon source.

20 Characteristic III may be any of the characteristics III described in the section "Characteristic III" herein below. In particular characteristic III may be that the yeast cell is capable of utilizing dipeptides as sole nitrogen source.

25 Characteristic IV may be any of the characteristics IV described in the section "Characteristic IV" herein below. In particular characteristic IV may be that the yeast cell is capable of utilizing tripeptides as sole nitrogen source.

30 Characteristic V may be any of the characteristics V described in the section "Characteristic III" herein below. In particular characteristic III may be that the yeast cell is capable of reducing the level of one or more amino acids to no more than 10% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells.

Characteristic VI may be any of the characteristics VI described in the section "Characteristic VI" herein below. In particular characteristic VI may be that the yeast cell is capable of generating at least 4.7 promille ethanol per °Plato, when said yeast

cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

Characteristic VII may be any of the characteristics VII described in the section

- 5 “Characteristic VII” herein below. In particular characteristic VII may be that the yeast cell is capable of fermenting sugar with a real degree of fermentation of at least 70, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

- 10 Characteristic XI may be any of the characteristics XI described in the section “Characteristic XI” herein below. In particular characteristic XI may be that the yeast cell is capable of fermenting wort with a time of primary fermentation of at the most 4 days.

- 15 The yeast cell according to the invention may have one or more of the characteristics. Thus, the yeast cell may have at least two, preferably at least three, more preferably at least four, yet more preferably at least five, such as at least 6, such as all of the characteristics I, II, III, IV, V, VI and VII. The yeast cell may also have at least two, preferably at least three, more preferably at least four, yet more preferably at least five,
20 such as at least 6, such as all of the characteristics I, II, III, IV, V, VI, VII and XI.

- Thus, the yeast cell of the invention may have the characteristics I and II. The yeast cell of the invention may also have characteristics I and III. The yeast cell of the invention may also have characteristics I and IV. The yeast cell of the invention may
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yeast cell may also have characteristics I, II, III and VII. The yeast cell may also have characteristics I, II, III and XI. The yeast cell of the invention may also have characteristics I, II, III, IV and V. The yeast cell of the invention may also have characteristics I, II, III, IV and VI. The yeast cell may also have characteristics I, II, III, IV and VII. The yeast cell may also have characteristics I, II, III, IV and XI. The yeast cell of the invention may also have characteristics I, II, III, IV, V and VI. The yeast cell may also have characteristics I, II, III, IV, V and VII. The yeast cell may also have characteristics I, II, III, IV, V and XI. The yeast cell may also have characteristics I, II, III, IV, V, VI and VII. The yeast cell may also have characteristics I, II, III, IV, V, VI and XI. The yeast cell may also have characteristics I, III and IV. The yeast cell may also have characteristics I, III and V. The yeast cell may also have characteristics I, III and VI. The yeast cell may also have characteristics I, III and VII. The yeast cell may also have characteristics I, III and XI. The yeast cell may also have characteristics I, III, IV and V. The yeast cell may also have characteristics I, III, IV and VI. The yeast cell may also have characteristics I, III, IV and VII. The yeast cell may also have characteristics I, III, IV and XI. The yeast cell may also have characteristics I, III, IV, V and VI. The yeast cell may also have characteristics I, III, IV, V and VII. The yeast cell may also have characteristics I, III, IV, V and XI. The yeast cell may also have characteristics I, III, IV, V, VI and VII. The yeast cell may also have characteristics I, III, V and VI. The yeast cell may also have characteristics I, III, V and VII. The yeast cell may also have characteristics I, III, V and XI. The yeast cell may also have characteristics I, III, VI and VII. The yeast cell may also have characteristics I, III, VI and XI. The yeast cell may also have characteristics I, III, VII and XI. The yeast cell may also have characteristics I, IV and V. The yeast cell may also have characteristics I, IV and VI. The yeast cell may also have characteristics I, IV and VII. The yeast cell may also have characteristics I, IV and XI. The yeast cell may also have characteristics I, IV, V and VI. The yeast cell may also have characteristics I, IV, V and VII. The yeast cell may also have characteristics I, IV, V and XI. The yeast cell may also have characteristics I, IV, VI and VII. The yeast cell may also have characteristics I, IV, VI and XI. The yeast cell may also have characteristics I, IV, V, VI and VII. The yeast cell may also have characteristics I, IV, V, VI and XI. The yeast cell may also have characteristics I, IV, V, VI, VII and XI. The yeast cell may also have characteristics I, V and VI. The yeast cell may also have characteristics I, V and VII. The yeast cell may also have characteristics I, V and XI. The yeast cell may also have characteristics I, V, VI and VII. The yeast cell

may also have characteristics I, V, VI and XI. The yeast cell may also have characteristics I, V, VI, VII and XI. The yeast cell may also have characteristics I, VI and VII. The yeast cell may also have characteristics I, VI and XI. The yeast cell may also have characteristics I, VI, VII and XI. The yeast cell may also have characteristics I, VII and XI. The yeast cell of the invention may also have characteristics II and III. The yeast cell of the invention may also have characteristics II and IV. The yeast cell of the invention may also have characteristics II and V. The yeast cell of the invention may also have characteristics II and VI. The yeast cell may also have characteristics II and VII. The yeast cell may also have characteristics II, and XI. The yeast cell of the invention may also have characteristics II, III and IV. The yeast cell of the invention may also have characteristics II, III and V. The yeast cell of the invention may also have characteristics II, III and VI. The yeast cell may also have characteristics II, III and VII. The yeast cell may also have characteristics II, III and XI. The yeast cell of the invention may also have characteristics II, III, IV and V. The yeast cell of the invention may also have characteristics II, III, IV and VI. The yeast cell may also have characteristics II, III, IV and VII. The yeast cell may also have characteristics II, III, IV and XI. The yeast cell of the invention may also have characteristics II, III, IV, V and VI. The yeast cell may also have characteristics II, III, IV, V and VII. The yeast cell may also have characteristics II, III, IV, V and XI. The yeast cell may also have characteristics II, III, IV, V, VI and VII. The yeast cell may also have characteristics II, III, IV, V, VI, VII and XI. The yeast cell may also have characteristics II, IV and V. The yeast cell may also have characteristics II, IV and VI. The yeast cell may also have characteristics II, IV and VII. The yeast cell may also have characteristics II, IV and XI. The yeast cell may also have characteristics II, IV, V and VI. The yeast cell may also have characteristics II, IV, V and VII. The yeast cell may also have characteristics II, IV, V and XI. The yeast cell may also have characteristics II, IV, V, VI and VII. The yeast cell may also have characteristics II, IV, V, VI and XI. The yeast cell may also have characteristics II, IV, V, VI, VII and XI. The yeast cell may also have characteristics II, V and VI. The yeast cell may also have characteristics II, V and VII. The yeast cell may also have characteristics II, V and XI. The yeast cell may also have characteristics II, V, VI and VII. The yeast cell may also have characteristics II, V, VI and XI. The yeast cell may also have characteristics II, V, VI, VII and XI. The yeast cell may also have characteristics II, VI and VII. The yeast cell may also have characteristics II, VI and XI.

The yeast cell may also have characteristics II, VI, VII and XI. The yeast cell may also have characteristics II, VII and XI. The yeast cell of the invention may also have characteristics III and IV. The yeast cell of the invention may also have characteristics III and V. The yeast cell of the invention may also have characteristics III and VI. The yeast cell may also have characteristics III and VII. The yeast cell may also have characteristics III and XI. The yeast cell of the invention may also have characteristics III, IV and V. The yeast cell of the invention may also have characteristics III, IV and VI. The yeast cell may also have characteristics III, IV and VII. The yeast cell may also have characteristics III, IV and XI. The yeast cell of the invention may also have characteristics III, IV, V and VI. The yeast cell may also have characteristics III, IV, V and VII. The yeast cell may also have characteristics III, IV, V and XI. The yeast cell may also have characteristics III, IV, V, VI and VII. The yeast cell may also have characteristics III, IV, V, VI and XI. The yeast cell may also have characteristics III, IV, V; VI, VII and XI. The yeast cell may also have characteristics III, V and VI. The yeast cell may also have characteristics III, V and VII. The yeast cell may also have characteristics III, V and XI. The yeast cell may also have characteristics III, VI and VII. The yeast cell may also have characteristics III, VI and XI. The yeast cell may also have characteristics III, VI, VII and XI. The yeast cell may also have characteristics III, VII and XI. The yeast cell of the invention may also have characteristics IV and V. The yeast cell of the invention may also have characteristics IV and VI. The yeast cell may also have characteristics IV and VII. The yeast cell of the invention may also have characteristics IV, V and VI. The yeast cell may also have characteristics IV, V and VII. The yeast cell may also have characteristics IV, V, VI and VII. The yeast cell may also have characteristics IV, VI and VII. The yeast cell may also have characteristics IV, VI and XI. The yeast cell may also have characteristics IV, VI, VII and XI. The yeast cell of the invention may also have characteristics V and VI. The yeast cell may also have characteristics V and VII. The yeast cell may also have characteristics V and XI. The yeast cell may also have characteristics V, VI and VII. The yeast cell may also have characteristics V, VII and XI. The yeast cell may also have characteristics VI and VII. The yeast cell may also have characteristics VI and XI. The yeast cell may also have characteristics VI, VII and XI. The yeast cell may also have characteristics VII and XI.

In a preferred embodiment of the invention the yeast cell has all of characteristics I, II, III, IV, V, VI and VII. In a preferred embodiment of the invention the yeast cell has all of characteristics I, II, III, IV, V, VI, VII and XI.

- 5 In addition to the characteristics outlined above, the yeast cells of the invention may have one or more additional characteristics.

Thus, in addition to one or more of characteristics I, II, III, IV, V, VI, VII and/or XI, then the yeast cell may also have characteristic VIII. Characteristic VIII may be any of the
10 characteristics VIII described in the section "Characteristic VIII" herein below. In particular characteristic VIII may be that the yeast cell is capable of utilizing melibiose as the sole carbon source.

In addition to one or more of characteristics I, II, III, IV, V, VI, VII, VIII and/or XI, then
15 the yeast cell may also have characteristic IX. Characteristic IX may be any of the characteristics IX described in the section "Characteristic IX" herein below. In particular characteristic IX may be that the yeast cell is capable of utilizing disaccharides and/or trisaccharides as the sole carbon source.

20 In addition to one or more of characteristics I, II, III, IV, V, VI, VII, VIII, IX and/or XI, then the yeast cell may also have characteristic X. Characteristic X may be any of the characteristics X described in the section "Characteristic X" herein below. In particular characteristic X may be that the yeast cell only has a low number of cells in suspension.

25

In a preferred embodiment of the invention, the yeast cell may have all of characteristics I, II, III, IV, V, VI, VII, VIII, IX, X and XI.

30 The yeast cell may be a yeast cell of any suitable species. In a preferred embodiment of the invention the yeast cell is a hybrid between a yeast cell of the species *S. pastorianus* and a yeast cell of the species *S. cerevisiae*.

Characteristic I

The yeast cell according to the invention may have the characteristic I, wherein characteristic I is that the yeast cell is capable of utilizing isomaltose. Thus, upon incubation in a medium containing isomaltose, then said yeast cell is capable of removing at least part of said isomaltose.

5

More preferably the characteristic I is that the yeast cell is capable of utilizing isomaltose as the sole carbon source. Thus, the yeast cell is capable of growing in a medium containing isomaltose as the sole carbon source. Such medium preferably do not contain any mono- and/or disaccharides apart from isomaltose, and more preferably such medium does not contain any carbohydrates apart from isomaltose.

10

Even if a yeast cell is capable of fermenting isomaltose, this does not necessarily mean that said yeast cell is capable of utilizing isomaltose as the sole carbon source. Thus, it is preferred that the yeast cell is capable both of utilizing isomaltose, and of utilizing isomaltose as the sole carbon source.

15

In particular the characteristic I may be that the yeast cell is capable of growing in a medium containing in the range of 1 to 5 g/L, for example in the range of 1 to 3 g/L, such as 2 g/L isomaltose as the sole carbon source. Such medium preferably do not contain any carbohydrates apart from said concentration of isomaltose.

20

One useful method for determining whether a yeast cell is capable of utilizing isomaltose as sole carbon source is described herein below in Example 5.

25

Yeast cells having characteristic I, preferably also have one or more of genotypes IV, V and VI, more preferably all of genotypes IV, V and VI described below.

Characteristic II

30

The yeast cell according to the invention may have the characteristic II, wherein characteristic II is that the yeast cell is capable of utilizing panose. Thus, upon incubation in a medium containing panose, then said yeast cell is capable of removing at least part of said panose. Preferably, said yeast cell is capable of removing (e.g. capable of fermenting) at least 45%, such as at least 50%, for example at least 60% of

the panose in said medium. Said medium may in particular be wort. Preferably, said yeast cell is capable of removing aforementioned amount of panose when incubated in said wort until diacetyl is in spec, e.g. for 4 to 6 days, e.g. for 5 days. Incubation may for example be at 16 to 18°C. Thus, said yeast cell may be capable of removing at
5 least 45%, such as at least 50%, for example at least 60% of the panose present in wort when determined by fermenting wort as described herein below in Example 5.

More preferably the characteristic II is that the yeast cell is capable of utilizing panose as the sole carbon source. Thus, the yeast cell is capable of growing in a medium
10 containing panose as the sole carbon source. Such medium preferably do not contain any mono-, di- and/or trisaccharides apart from panose, and more preferably such medium does not contain any carbohydrates apart from panose.

Even if a yeast cell is capable of fermenting panose, this does not necessarily mean
15 that said yeast cell is capable of utilizing panose as the sole carbon source. In one embodiment the yeast cell is capable both of utilizing panose, and of utilizing panose as the sole carbon source.

In particular the characteristic II may be that the yeast cell is capable of growing in a
20 medium containing in the range of 1 to 5 g/L, for example in the range of 1 to 3 g/L, such as 2 g/L panose as the sole carbon source. Such medium preferably do not contain any carbohydrates apart from said concentration of panose.

One useful method for determining whether a yeast cell is capable of utilizing panose
25 as sole carbon source is described herein below in Example 5.

Yeast cells having characteristic II, preferably also have one or more of genotypes IV, V and VI, more preferably all of genotypes IV, V and VI described below.

30 **Characteristic III**

The yeast cell according to the invention may have the characteristic III, wherein characteristic III is that the yeast cell is capable of utilizing dipeptides. Thus, upon

incubation in a medium containing dipeptides, then said yeast cell is capable of removing at least part of said dipeptides.

5 More preferably the characteristic III is that the yeast cell is capable of utilizing dipeptides as the sole nitrogen source. Thus, the yeast cell is capable of growing in a medium containing dipeptides as the sole nitrogen source. Such medium preferably do not contain any amino acids and peptides apart from dipeptides, and more preferably such medium does not contain any amino acids, peptides and ammonium apart from dipeptides.

10

The characteristic III may be that the yeast cell is capable of utilizing any dipeptide as the sole nitrogen source. However, it is also possible that said yeast is capable of utilizing only one or more specific dipeptides as the sole nitrogen source.

15 It is preferred that characteristic III is that the yeast cell is capable of utilizing at least one, such as at least two, for example at least three, such as at least 4, for example at least 5, such as all of the following dipeptides:

Met-Tyr

Leu-Tyr

20 Val-Met

Phe-Tyr

Ile-Leu

Ile-Asn.

25 In one embodiment, the characteristic III is that the yeast cell is capable of utilizing at least one, such as at least 3, for example at least 5, such as at least 7, for example at least 9, such as all of the following dipeptides:

Gly-Arg

Ile-Asn

30 Lys-Tyr

Met-Lys

Val-Ala

Val-Asn

Val-Gly

Val-Gln

Val-Met

Val-Ser

- 5 The characteristic III may also be that the yeast cell is capable of utilizing one or more dipeptides of the formula Val-Xaa, wherein Xaa denotes any amino acid. For example, the characteristic III may be that the yeast cell is capable of utilizing at least 3, such as at least 4, for example at least 6 different dipeptides of the formula Val-Xaa. In particular, Xaa may be an amino acid selected from the group consisting of Ala, Asn, Gly, Gln, Met and Ser.

- Characteristic III may also be that the yeast cell is capable of utilizing one or more dipeptides of the formula Ala-Xaa, wherein Xaa denotes any amino acid. In particular, Xaa may be an amino acid selected from the group consisting of Glu, Gly, His and Thr.
- 15 Frequently, the capability of utilizing a dipeptide of the formula Ala-Xaa is connected with the capability of utilizing allantoin that is an intermediate of allantoin catabolism. Thus, it is preferred that the yeast cell furthermore is capable of utilizing allantoin as sole nitrogen source.

- 20 The characteristic III may also be that the yeast cell is capable of utilizing one or more of the following dipeptides, for example at least 3 of the following dipeptides, such as at least 5 of the following dipeptides, such as all of the following dipeptides:

Met-Tyr

Leu-Tyr

- 25 Val-Met

Phe-Tyr

Ile-Leu

Ile-Asn

Ala-Xaa, wherein Xaa is any amino acid and preferably Xaa is Glu, Gly, His or Thr.

- 30

One useful method for determining whether a yeast cell is capable of utilizing dipeptides as sole nitrogen source is described herein below in Example 6. The skilled person will understand that the methods described in Example 6 can be used to test

whether any dipeptide can be utilized as sole nitrogen source by exchanging the tested dipeptides.

5 Yeast cells having characteristic III, preferably also have one or more of genotypes I, II and III, more preferably all of genotypes I, II and III described below.

Characteristic IV

10 The yeast cell according to the invention may have the characteristic IV, wherein characteristic IV is that the yeast cell is capable of utilizing tripeptides. Thus, upon incubation in a medium containing tripeptides, then said yeast cell is capable of removing at least part of said tripeptides.

15 More preferably the characteristic IV is that the yeast cell is capable of utilizing tripeptides as the sole nitrogen source. Thus, the yeast cell is capable of growing in a medium containing tripeptides as the sole nitrogen source. Such medium preferably do not contain any amino acids and peptides apart from tripeptides, and more preferably such medium does not contain any amino acids, peptides and ammonium apart from tripeptides.

20

The characteristic IV may be that the yeast cell is capable of utilizing any tripeptide as the sole nitrogen source. However, it is also possible that said yeast is capable of utilizing only one or more specific tripeptides as the sole nitrogen source.

25 It is preferred that characteristic IV is that the yeast cell is capable of utilizing the tripeptide Gly-Gly-Gly as the sole nitrogen source.

30 One useful method for determining whether a yeast cell is capable of utilizing tripeptides as sole nitrogen source is described herein below in Example 6. The skilled person will understand that the methods described in Example 6 can be used to test whether any tripeptide can be utilized as sole nitrogen source by exchanging the tested tripeptides.

Yeast cells having characteristic IV, preferably also have one or more of genotypes I, II and III, more preferably at least genotypes II and III described below.

5 **Characteristic V**

The yeast cell according to the invention may have the characteristic V, wherein characteristic V is a high utilization of amino acids.

10 In general it is preferred that the yeast cell of the invention is capable of utilizing amino acids to a high degree. This both ensures that energy stored in amino acids can be utilized as well as ensuring a low level of amino acids after fermentation. Thus, if said yeast is used for preparation of beer, then the final beer will have a low level of amino acids. Strecker aldehydes are important constituents of the "aged" flavor in beer that
15 partly originate from the amino acids of the bottled beer itself. Amino acids that have been shown to be involved in formation of Strecker aldehydes with a low sensory threshold include valine, isoleucine, leucine, methionine and phenylalanine (Table 2). Strecker aldehyde formation plays a crucial role because an increase in their concentration, gives an increasing sensory perception of "aged flavours".

20 Accordingly, it is an advantage of the yeast according to the present invention that the yeast cell is capable of utilizing amino acids to a higher degree than both conventional lager yeast and ale yeasts.

25 Thus, it is preferred that the yeast cells of the invention have characteristic V, wherein the characteristic V is that said yeast cells are capable of reducing the level of one or more amino acids to no more than 10% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells. In particular, characteristic V may be that the yeast cell is capable of reducing the level of at least 12, such as at
30 least 13, for example of at least 14 different amino acids to less than 10% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells. For example the yeast cell may be able to reduce in the range of 12 to 20, such as in the range of 14 to 20 amino to no more than 10% of the starting

concentration after incubation for 5 days under conditions allowing growth of said yeast cells.

5 The characteristic V may also be that the yeast cell is capable of reducing the total level of amino acids to less than 30%, such as less than 25% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells.

10 The characteristic V may also be that the yeast cell is capable of reducing the level of one or more amino acids to no more than 5% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells. In particular, characteristic V may be that the yeast cell is capable of reducing the level of at least 10, such as at least 11, for example of at least 13 different amino acids to less than 5% of the starting concentration after incubation for 5 days under conditions allowing
15 growth of said yeast cells.

The characteristic V may also be that the yeast cell is capable of reducing the level of one or more amino acids to no more than 1% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells. In particular,
20 characteristic V may be that the yeast cell is capable of reducing the level of at least 5, such as at least 6, for example of at least 7 different amino acids to less than 1% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells.

25 The characteristic V may also be that the yeast cell is capable of reducing the level of one or more of the Strecker aldehyde forming amino acids. Thus, characteristic V may be that the yeast cell is capable of reducing the level of Met to less than 10%, preferably less than 5%, even more preferably to at the most 2%, yet more preferably to less than 1% of the starting concentration after incubation for 5 days under
30 conditions allowing growth of said yeast cells. In particular, the yeast cell may be capable of removing essentially all Met after incubation for 5 days under conditions allowing growth of said yeast cells. Characteristic V may also be that the yeast cell is capable of reducing the level of Val to less than 10%, preferably less than 5%, even more preferably to at the most 2% of the starting concentration after incubation for 5

days under conditions allowing growth of said yeast cells. The characteristic V may also be that the yeast cell is capable of reducing the level of Ile to less than 10%, preferably less than 5%, even more preferably to at the most 2%, yet more preferably to less than 1% of the starting concentration after incubation for 5 days under

5 conditions allowing growth of said yeast cells. In particular, the yeast cell may be capable of removing essentially all Ile after incubation for 5 days under conditions allowing growth of said yeast cells. The characteristic V may also be that the yeast cell is capable of reducing the level of Leu to less than 10%, preferably less than 5%, even more preferably to at the most 2% of the starting concentration after incubation for 5

10 days under conditions allowing growth of said yeast cells. The characteristic V may also be that the yeast cell is capable of reducing the level of Phe to less than 10%, preferably less than 5%, even more preferably to at the most 2%, yet more preferably to less than 1% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells. In particular, the yeast cell may be

15 capable of removing essentially all Phe after incubation for 5 days under conditions allowing growth of said yeast cells.

The term "removing essentially all" is used herein to denote that the amino acid is removed to a level, which is below the detection level, when the detection is performed

20 by UPLC.

It is also comprised within the present invention that characteristic V is that the yeast cell is capable of reducing the level of at least 2, preferably of at least 3, more preferably of at least 4, yet more preferably of all of the amino acids Met, Val, Ile, Leu

25 and Phe to less than 10%, preferably less than 5%, even more preferably to at the most 2% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells.

The characteristic V may also be that the yeast cells are capable of utilizing at least

30 80% of at least one of the amino acids Met, Val, Ile, Leu and Phe, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

It is also preferred that the yeast cells of the invention have the characteristic V, wherein said characteristic V is that the yeast cells are capable of reducing the total level of the amino acids Met, Val, Ile, Leu and/or Phe to at the most 400 mg/L, such as at the most 100 mg/L, such as at the most 50 mg/L, for example to at the most 10 mg/L
5 after incubation for 6 days under conditions allowing growth of said yeast cell.

The characteristic V may also be a combination of any of the aforementioned characteristic Vs described in this section. Thus for example, the characteristic V may be that the yeast cell is capable of reducing the level of at least 12, such as at least 13,
10 for example of at least 14 different amino acids to less than 10% and is capable of reducing the total level of amino acids to less than 30%, such as less than 25% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells. The characteristic V may also be that the yeast cell is capable of reducing the level of at least 10 amino acids to less than 5% and is capable of reducing
15 the total level of amino acids to less than 30%, such as less than 25% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells. The characteristic V may also be that the yeast cell is capable of reducing the level of at least 5 amino acids to less than 1% and is capable of reducing the total level of amino acids to less than 30%, such as less than 25% of the starting concentration
20 after incubation for 5 days under conditions allowing growth of said yeast cells. Conditions allowing growth of said yeast cells are described herein below in the section "Method for producing a beverage". Said conditions may be any of the fermentation conditions described in that section. E.g. said conditions may be incubation at a temperature in the range of 10 to 20°C in wort. The level of amino acids may be
25 determined by any useful method, e.g. using HPLC or UPLC. Useful methods for determining whether a yeast cell has a high utilization of amino acids are described herein below in Examples 4 and 9.

Characteristic VI

30

The yeast cell according to the invention may have the characteristic VI, wherein characteristic VI is high production of alcohol. Since the amount of alcohol produced by a given yeast cell is highly influenced by the starting material, it is preferred that the characteristic I is that the yeast cell is capable of generating at least 4.7 promille

ethanol per °Plato. °Plato is a measure for the density of a liquid, and thus indicates the level of sugars and other fermentable nutrients.

5 In particular, it is preferred that the yeast cell is capable of generating at least 4.7 promille ethanol per °Plato, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

10 Preferably, the diacetyl is considered to be in spec when the level of diacetyl is at the most 30 ppb.

Characteristic VII

15 The yeast cell according to the invention may have the characteristic VII, wherein characteristic VII is a high real degree of fermentation (RDF).

The RDF measures the degree to which sugar in the starting liquid has been fermented into alcohol. Thus, if the starting liquid is a wort the RDF measures the degree to which sugar in the wort has been fermented into alcohol in the resulting beer.

20 It is preferred that the yeast cell according to the invention has characteristic VII, wherein characteristic VII is that the yeast cell is capable of fermenting sugar with an RDF of at least 68%, such as at least 69%, for example at least 70%, and more preferably with an RDF of at least 71%.

25 In particular it is preferred that the yeast cell is capable of fermenting sugar with an RDF, which is higher than the RDF of at least one of the parental strains. Thus, the yeast cell according to the invention may be a hybrid yeast cell, which is capable of fermenting sugar with an RDF which is at least 1% higher, for example at least 2% higher than the RDF of one of the parental strains. In particular the yeast cell according
30 to the invention may be a hybrid between a parent *S. pastorianus* strain and a parent *S. cerevisiae* strain. In such embodiments, the yeast cell may be capable of fermenting sugar with an RDF at least 1 higher than the RDF of the parent *S. pastorianus* strain. The yeast cell according to the invention may also be a hybrid between a parent *S. diastaticus* strain and a parent *S. cerevisiae* strain. In such embodiments, the yeast cell

may be capable of fermenting sugar with an RDF at least 1% higher, preferably at least 2% higher than the RDF of the parent *S. diastaticus* strain.

Characteristic VIII

5

The yeast cell according to the invention may have the characteristic VIII, wherein characteristic VIII is that the yeast cell is capable of utilizing melibiose. Thus, upon incubation in a medium containing melibiose, then said yeast cell is capable of removing at least part of said melibiose.

10

More preferably the characteristic VIII is that the yeast cell is capable of utilizing melibiose as the sole carbon source. Thus, the yeast cell is capable of growing in a medium containing melibiose as the sole carbon source. Such medium preferably do not contain any mono- and/or di saccharides apart from melibiose, and more preferably such medium does not contain any carbohydrates apart from melibiose.

15

One useful method for determining whether a yeast cell is capable of utilizing melibiose as sole carbon source is described herein below in Example 7.

20

Characteristic IX

The yeast cell according to the invention may have the characteristic IX, wherein characteristic IX is that the yeast cell is capable of utilizing disaccharides and/or trisaccharides. Thus, upon incubation in a medium containing di-saccharides and/or trisaccharides, then said yeast cell is capable of removing at least part of said disaccharides and/or trisaccharides.

25

More preferably the characteristic IX is that the yeast cell is capable of utilizing disaccharides and/or trisaccharides as the sole carbon source. Thus, the yeast cell is capable of growing in a medium containing disaccharides and/or trisaccharides as the sole carbon source. Such medium preferably do not contain any saccharides apart from the disaccharides and/or trisaccharides.

30

The characteristic IX may be that the yeast cell is capable of utilizing any disaccharide and trisaccharide as the sole carbon source. However, it is also possible that said yeast is capable of utilizing only one or more specific disaccharides and/trisaccharides as the sole carbon source. As described above it is preferred that the yeast cells are
5 capable of utilizing isomaltose (characteristic I), panose (characteristic II), and/or melibiose (characteristic VIII).

Thus, the characteristic IX is preferably that the yeast cell is capable of utilizing one or more disaccharide and/or trisaccharide, which is not isomaltose, panose, or melibiose.
10 Thus, characteristic IX may be that the yeast cell is capable of utilizing one or more disaccharides and/or trisaccharides in addition to isomaltose, panose, or melibiose. The yeast cell may thus be capable of utilizing one or more disaccharide and/or trisaccharide, which is not isomaltose, panose, or melibiose as sole carbon source, and in addition said yeast cell may have one or more of characteristics I, II or VIII.

15 It is preferred that characteristic IX is that the yeast cell is capable of utilizing at least one, such as at least two, for example at least three, such as at least 4, for example at least 5, such as all disaccharides selected from the group consisting of kojibiose, nigerose, sucrose, turanose, leucrose, and palatinose as sole carbon source.

20 It is also preferred that characteristic IX is that the yeast cell is capable of utilizing maltotriose and/or isomaltotriose as sole carbon source.

Thus, the yeast cells may be capable of utilizing maltotriose as the sole carbon source.
25 Thus, the yeast cell may be capable of growing in a medium containing maltotriose as the sole carbon source. Such medium preferably do not contain any mono- and/or disaccharides and/or trisaccharides apart from maltotriose, and more preferably such medium does not contain any carbohydrates apart from maltotriose.

30 In particular the characteristic IX may be that the yeast cell is capable of growing in a medium containing in the range of 1 to 5 g/L, for example in the range of 1 to 3 g/L, such as 2 g/L maltotriose as the sole carbon source. Such medium preferably do not contain any carbohydrates apart from said concentration of maltotriose.

Many yeast cells, e.g. many lager yeast cells are not capable of utilizing maltotriose as sole carbon source, in particular many lager yeast cells are not capable of utilizing maltotriose as sole carbon source, when maltotriose is present only at low levels.

5 The yeast cells may be capable of utilizing maltulose as the sole carbon source. Thus, the yeast cell may be capable of growing in a medium containing maltulose as the sole carbon source. Such medium preferably do not contain any mono- and/or disaccharides apart from maltulose, and more preferably such medium does not contain any carbohydrates apart from maltulose.

10

In particular the characteristic IX may be that the yeast cell is capable of growing in a medium containing in the range of 1 to 5 g/L, for example in the range of 1 to 3 g/L, such as 2 g/L maltulose as the sole carbon source. Such medium preferably do not contain any carbohydrates apart from said concentration of maltulose.

15

Many yeast cells, e.g. many lager yeast cells are not capable of utilizing maltulose as sole carbon source.

20

The yeast cells may be capable of utilizing kojibiose as the sole carbon source. Thus, the yeast cell may be capable of growing in a medium containing kojibiose as the sole carbon source. Such medium preferably do not contain any mono- and/or disaccharides apart from kojibiose, and more preferably such medium does not contain any carbohydrates apart from kojibiose.

25

In particular the characteristic IX may be that the yeast cell is capable of growing in a medium containing in the range of 1 to 5 g/L, for example in the range of 1 to 3 g/L, such as 2 g/L kojibiose as the sole carbon source. Such medium preferably do not contain any carbohydrates apart from said concentration of kojibiose.

30

Many yeast cells, e.g. many lager yeast cells are not capable of utilizing kojibiose as sole carbon source.

Thus, the yeast cells according to the invention may be capable of utilizing one or more of the disaccharides and/or trisaccharides described in Table 13.

Table 13

Substrate	Linkage
Disaccharides (Glc → Glu)	
Kojibiose	O- α -D-glucosyl-(1→2)- α -D-glucose
Nigerose	O- α -D-glucosyl-(1→3)- α -D-glucose
Isomaltose	O- α -D-glucosyl-(1→6)- α -D-glucose
Disaccharides (Glc → Fru)	
Sucrose	O- α -D-glucosyl-(1→2)- β -D-fructose
Turanose	O- α -D-glucosyl-(1→3)-D-fructose
Maltulose	O- α -D-glucosyl-(1→4)-D-fructose
Leucrose	O- α -D-glucosyl-(1→5)-D-fructose
Palatinose	O- α -D-glucosyl-(1→6)-D-fructose
Trisaccharides	
Maltotriose	O- α -D-glucosyl-(1→4)- α -D-glucosyl-(1→4)-D-glucose
Isomaltotriose	O- α -D-glucosyl-(1→6)- α -D-glucosyl-(1→6)-D-glucose
Panose	O- α -D-glucosyl-(1→6)- α -D-glucosyl-(1→4)-D-glucose

Useful methods for determining whether a yeast cell is capable of utilizing disaccharides and/or trisaccharides are described herein below in Examples 8 and 11.

- 5 A useful method for determining whether a yeast cell is capable of utilizing disaccharides and/or trisaccharides as sole carbon source is described herein below in Example 5. The skilled person will understand that the methods described in Example 5 can be used to test whether any disaccharide and/or trisaccharide can be utilized as sole carbon source by exchanging panose/isomaltose, with the disaccharide and/or
- 10 trisaccharide to be tested.

Yeast cells having characteristic IX, preferably also have one or more of genotypes IV, V and VI, more preferably all of genotypes IV, V and VI described below.

15

Characteristic X

The yeast cell according to the invention may have the characteristic X, wherein characteristic X is that the yeast cell only has a low number of cells in suspension, in particular the yeast cell has a low number of cells in suspension after incubation in a liquid medium in a container. Said incubation is preferably incubation for 1 to 14 days, such as from 2 to 10 days, for example from 4 to 8 days, for example from 4 to 6 days.

In particular it is preferred that characteristic X is that at the most 12 million, such as at the most 10 million cells/ml are in suspension after incubation for 4 days under conditions allowing growth of said yeast cell. Thus, characteristic X may be that at the most 12 million, such as at the most 10 million cells/ml are in suspension, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated for 4 days. Characteristic X may also be that at the most 12 million, such as at the most 10 million cells/ml are in suspension, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated for 5 days. Characteristic X may also be that at the most 12 million, such as at the most 10 million cells/ml are in suspension, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated for 6 days. Said incubation may for example be at a temperature in the range of 10 to 20°C, such as in the range of 10 to 18°C, for example at 16°C or 18°C. The starting concentration of yeast cells may for example be in the range of 10 to 20 mill cells/ml, e.g. in the range of 14 to 15 mill cells/ml.

It may also be preferred that characteristic X may be that the yeast cell has a number of cells in suspension per ml which is at the most 80%, such as at the most 70%, for example at the most 60%, such as at the most 50%, for example at the most 40% of the starting number of cells per ml after 4 to 6 days, such as for 5 days incubation under conditions allowing growth of said cells.

For example, characteristic X may be that the yeast cell has a number of cells in suspension per ml which is at the most 80%, such as at the most 70%, for example at the most 60%, such as at the most 50%, for example at the most 40% of the starting number of cells per ml after 4 to 6 days, such as for 5 days incubation in a wort composition having a sugar content of at least 10° Plato. Characteristic X may also be that the yeast cell has a number of cells in suspension per ml which is at the most 80%,

such as at the most 70%, for example at the most 60%, such as at the most 50%, for example at the most 40% of the starting number of cells per ml after 6 days incubation in a wort composition having a sugar content of at least 10° Plato. Said incubation may for example be at a temperature in the range of 15 to 20°C, such as in the range of 10 to 18°C, for example at 16°C or 18°C.

In one embodiment characteristic X is that at the most 25 million, preferably at the most 20 million cells/ml are in suspension after incubation for 7 days under conditions allowing growth of said yeast cell. Thus, characteristic X may be that at the most 25 million, such as at the most 20 million cells/ml are in suspension, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated for 7 days at 18°C.

One useful method for determining cells in suspension is described herein below in Example 2.

Characteristic XI

The yeast cell according to the invention may have the characteristic XI, wherein characteristic XI is that the yeast cell is capable of fermenting wort with a time of primary fermentation of at the most 4 days.

In a preferred embodiment of the invention the characteristic XI is that the yeast cell is capable of fermenting wort with a time primary fermentation of at the most 3.5 days.

In another embodiment of the invention the characteristic XI is that the yeast cell is capable of fermenting wort with a time of primary fermentation of at the most 3 days.

The characteristic XI may also be that the yeast cell is capable of fermenting wort with a time of primary fermentation, which is at least one day shorter than the time of primary fermentation by at least one of the parental strains under the same conditions. Thus, the yeast cell according to the invention may be a hybrid yeast cell, which is capable of fermenting wort with a time of primary fermentation, which is at least one day shorter than the time of primary fermentation by at least one of the parental strains

under the same conditions. In particular the yeast cell according to the invention may be a hybrid between a parent *S. pastorianus* strain and a parent *S. cerevisiae* strain. In such embodiments, the yeast cell may be capable of fermenting wort with a time of primary fermentation, which is at least one day shorter than the time primary
5 fermentation by the parent *S. pastorianus* strain under the same conditions.

Said wort may be any standard wort, but is preferably a wort with having a sugar content of at least 10° Plato. Thus, said wort may in particular be a wort having a sugar content of in the range of 10° Plato to 20° Plato. In particular, said wort may be wort
10 having a sugar content of 14 to 16° Plato.

The term "time of primary fermentation" is the time from pitching wort with yeast until the primary fermentation is completed. The primary fermentation is considered completed when the apparent extract is stable and/or when there is no longer active
15 CO₂ release. The apparent extract is considered to be stable when the apparent extract between two measurements does not alter by more than +/- 15%, preferably by not more than +/- 10%.

Yeast may be pitched at any useful level, for example at 10 to 20 mill viable cells/ml,
20 such as 13 to 16 mill viable cells/ml, for example 14-15 mill viable cells/ml.

The time of primary fermentation may be determined at a temperature at which the yeast cell is capable of growing. Thus, the time of primary fermentation may be determined at a temperature of in the range of 10 to 25°C, preferably at a temperature
25 in the range of 12 to 20°C, for example in the range of 14 to 18°C.

One method of determining the time of primary fermentation is described in Example 3 herein below.

30 **Genetic background**

The yeast cells according to the invention may have one or more of characteristics I to XI described herein above.

In addition to said characteristics, the yeast cell according to the invention may have one or more of the genotypes I to VI described herein below. Said genotypes may be linked to the characteristics outlined above.

- 5 In one embodiment, the yeast cell according to the invention at least has the genotype IV described herein below. In addition to having the genotype IV said yeast may also have one or more of the genotypes I, II, III, V, VI and one or more of the characteristics I to XI.
- 10 Thus, in one embodiment of the invention, the yeast cell has at least the genotype IV described below, and the genotype V described below. In addition to having the genotypes IV and V, said yeast may also have one or more of the genotypes I, II, III, VI and one or more of the characteristics I to XI.
- 15 Thus, the yeast cell of the invention may have the genotypes I and II. The yeast cell of the invention may also have genotypes I and III. The yeast cell of the invention may also have genotypes I and IV. The yeast cell of the invention may also have genotypes I and V. The yeast cell of the invention may also have genotypes I and VI. The yeast cell of the invention may also have genotypes I, II, and III. The yeast cell of the invention may also have genotypes I, II and IV. The yeast cell of the invention may also have genotypes I, II and V. The yeast cell of the invention may also have genotypes I, II and VI. The yeast cell of the invention may also have genotypes I, II, III and IV. The yeast cell of the invention may also have genotypes I, II, III and V. The yeast cell of the invention may also have genotypes I, II, III and VI. The yeast cell of the invention may also have genotypes I, II, III, IV and V. The yeast cell of the invention may also have genotypes I, II, III, IV and VI. The yeast cell of the invention may also have genotypes I, II, III, IV, V and VI. The yeast cell of the invention may also have genotypes II and III. The yeast cell of the invention may also have genotypes II and IV. The yeast cell of the invention may also have genotypes II and V. The yeast cell of the invention may also have genotypes II and VI. The yeast cell of the invention may also have genotypes II, III and IV. The yeast cell of the invention may also have genotypes II, III and V. The yeast cell of the invention may also have genotypes II, III and VI. The yeast cell of the invention may also have genotypes II, III, IV and V. The yeast cell of the invention may also have genotypes II, III, IV and VI. The yeast cell of the invention may also have
- 20
- 25
- 30

genotypes II, III, IV, V and VI. The yeast cell of the invention may also have genotypes III and IV. The yeast cell of the invention may also have genotypes III and V. The yeast cell of the invention may also have genotypes III and VI. The yeast cell of the invention may also have genotypes III, IV and V. The yeast cell of the invention may also have genotypes III, IV and VI. The yeast cell of the invention may also have genotypes III, IV, V and VI. The yeast cell of the invention may also have genotypes IV and V. The yeast cell of the invention may also have genotypes IV and VI. The yeast cell of the invention may also have genotypes IV, V and VI. The yeast cell of the invention may also have genotypes V and VI.

In a preferred embodiment of the invention the yeast cells have all of genotypes I, II, III, IV, V and VI.

In one embodiment of the invention, the yeast cell according to the invention may be a yeast cell comprising the genomic DNA sequence available under DDBJ/EMBL/GenBank accession number LOQJ000000000, in particular the DNA sequence available under DDBJ/EMBL/GenBank the accession number LOQJ000000000, version no. LOQJ01000000. This sequence is provided as a whole Genome Shotgun project and more details on this sequence are provided herein below in the Examples.

In another embodiment of the invention, the yeast cell according to the invention may be a yeast cell comprising the genomic DNA sequence available under DDBJ/EMBL/GenBank accession number LOQJ000000000, in particular the DNA sequence available under DDBJ/EMBL/GenBank the accession number LOQJ000000000, version no. LOQJ01000000. This sequence is provided as a Whole Genome Shotgun project, and more details on this sequence are provided herein below in the Examples.

Based on the genomic sequences provided herein, synthetic yeast chromosomes may be prepared. This may for example be performed as described by Callaway in Nature in 2014 (Nature DOI: doi:10.1038/nature.2014.14941), or by Annaluru et al., Science 4 April 2014; Vol. 344 no. 6179 pp. 55-58 (DOI: 10.1126/science.1249252). Also "Synthetic Yeast 2.0" provides information on how to prepare synthetic yeast

chromosomes (see e.g. <http://syntheticyeast.org/>). Yeast cells comprising said synthetic yeast chromosomes can be prepared using conventional recombinant technology.

5 Genotype I

The yeast cell according to the invention may have the genotype I, wherein the genotype I is the presence of a gene encoding DAL5. In particular, it is preferred that the yeast cell according to the invention comprises a gene encoding DAL5 of SEQ ID
10 NO:6 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith. Preferably, the genotype I is the presence of a gene encoding DAL5 of SEQ ID NO:6.

In one embodiment of the invention, the genotype I is the presence of at least one
15 allelic gene encoding DAL5, wherein the allelic gene encoding DAL5 encodes DAL5 selected from the group consisting of DAL5 of SEQ ID NO:6, DAL5 of SEQ ID NO:39, DAL5 of SEQ ID NO:40 and functional homologues thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity with any of the aforementioned.

20

In one embodiment, the genotype I may be the presence of the following 2 allelic genes:

- 25 1) a gene encoding DAL5 of SEQ ID NO:39 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) a gene encoding DAL5 of SEQ ID NO:40 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

30

DAL5 is a di-peptide transporter that is transporting di-peptides by non-N-end rule. The yeast cell may for example have genotype I in embodiments of the invention, where the

yeast cell has characteristics III, IV and/or VI, in particular when the yeast cell has characteristic III.

Genotype II

5

The yeast cell according to the invention may have the genotype II, wherein the genotype II is the presence of at least 3 genes encoding PTR2. In particular, it is preferred that the yeast cell according to the invention comprises at least 3 genes encoding PTR2, wherein PTR2 may be selected from the group consisting PTR2 of
10 SEQ ID NO:7, PTR2 of SEQ ID:8, PRT2 of SEQ ID NO:9 and functional homologues of each of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

Thus, the genotype II may be that the yeast cell comprises 3 genes selected from the
15 group consisting of:

- 1) a gene encoding PRT2 of SEQ ID NO:7 or a functional homologues of each of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity;
- 2) a gene encoding PRT2 of SEQ ID NO:8 or a functional homologues of each of
20 the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity; and
- 3) a gene encoding PRT2 of SEQ ID NO:9 or a functional homologues of each of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity.

25

Thus, the genotype II may be that the yeast cell comprises the following 3 genes:

- 1) a gene encoding PRT2 of SEQ ID NO:7 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith;
- 30 2) a gene encoding PRT2 of SEQ ID NO:8 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and

- 3) a gene encoding PRT2 of SEQ ID NO:9 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

5 Thus, the genotype II may be that the yeast cell comprises 3 genes selected from the group consisting of:

- 1) a gene encoding PRT2 of SEQ ID NO:7;
- 2) a gene encoding PRT2 of SEQ ID NO:8; and
- 3) a gene encoding PRT2 of SEQ ID NO:9.

10

In one embodiment genotype II may be that the yeast cell comprises at least 2 allelic genes encoding PTR2. For example, genotype II may be that the yeast cell comprises at least two allelic genes encoding PTR2 individually selected from the group consisting of genes encoding PTR2 of SEQ ID NO:7, PTR2 of SEQ ID:8, PRT2 of
15 SEQ ID NO:9, PRT2 of SEQ ID NO:37, PRT2 of SEQ ID NO:38, PRT2 of SEQ ID NO:43, PRT2 of SEQ ID NO:44 and functional homologues of each of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

20 In one embodiment the genotype II may be that the yeast cell comprises the following 2 allelic genes:

- 1) a gene encoding PRT2 of SEQ ID NO:37 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith;
- 25 2) a gene encoding PRT2 of SEQ ID NO:38 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In one embodiment the genotype II may be that the yeast cell comprises the following 2
30 allelic genes:

- 1) a gene encoding PRT2 of SEQ ID NO:43 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith;

- 2) a gene encoding PRT2 of SEQ ID NO:44 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

5 PRT2 is a transporter for di- and tripeptides, as well as other peptides into the yeast cell .

The yeast cell may for example have genotype II in embodiments of the invention, where the yeast cell has characteristics III, IV and/or V, such as in embodiment where
10 the yeast cell has characteristics III and/or IV.

Genotype III

The yeast cell according to the invention may have the genotype III, wherein the
15 genotype III is the presence of a gene encoding UBR1. In particular, it is preferred that the yeast cell according to the invention comprises a gene encoding UBR1 comprising SEQ ID NO:10, or UBR1 of SEQ ID NO:11 or a functional homologue of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith. Preferably, the genotype
20 III is the presence of at least two genes encoding UBR1 comprising SEQ ID NO:10, or UBR1 of SEQ ID NO:11 or a functional homologue of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

25 For example the genotype III may be the presence of the following 2 genes:

- 1) a gene encoding UBR1 comprising SEQ ID NO:10 or SEQ ID NO:45 or a functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 30 2) a gene encoding UBR1 of SEQ ID NO:11 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In particular, the genotype III may be the presence of the following 2 genes:

- 1) a gene encoding UBR1 of comprising SEQ ID NO:10; and
- 2) a gene encoding UBR1 of SEQ ID NO:11.

The yeast cell may for example have genotype III in embodiments of the invention, where the yeast cell has characteristics III and/or IV.

5

In one embodiment of the invention genotype III is that the yeast cell comprises at least one allelic genes encoding UBR1 selected from the group consisting of UBR1 comprising SEQ ID NO:10, UBR1 of SEQ ID NO:11, UBR1 comprising SEQ ID NO:41, UBR1 of SEQ ID NO:42, UBR1 comprising SEQ IDN NO:45 and functional

10 homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

15

In one embodiment of the invention genotype III is that the yeast cell comprises at least two allelic genes encoding UBR1 individually selected from the group consisting of UBR1 comprising SEQ ID NO:10, UBR1 of SEQ ID NO:11, UBR1 comprising SEQ ID NO:41, UBR1 of SEQ ID NO:42 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

20

For example the genotype III may be the presence of the following 2 genes:

- 1) a gene encoding UBR1 comprising SEQ ID NO:41 or a functional homologues thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 25 2) a gene encoding UBR1 of SEQ ID NO:42 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

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The yeast cell may for example have genotype III in embodiments of the invention, where the yeast cell has characteristics III, IV and/or V, such as in embodiment where the yeast cell has characteristics III and/or IV.

Genotype IV

The yeast cell according to the invention may have the genotype IV, wherein the genotype IV is the presence of at least 3 allelic genes, preferably at least 4 allelic genes encoding IMA1p. In particular, it is preferred that the yeast cell according to the invention comprises at least 4 allelic genes encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

IMA1p may be encoded by different alleles, for example by the short allele of IMA1, or by the long allele of IMA1. One yeast cell may comprise both long and short alleles of IMA1. In one embodiment, it may be preferred that the yeast cell according to the invention comprises at least 3 long alleles encoding IMA1p.

For example the genotype IV may be the presence of at least 2 short alleles of IMA1. Said two short alleles of IMA1 may be genes encoding IMA1p selected from the group consisting of IMA1p SEQ ID NO:12, IMA1p of SEQ ID NO:13 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In a preferred embodiment, the genotype IV may be the presence of at least 3 short alleles of IMA1. Said 3 short alleles of IMA1 may be allelic genes encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:1, IMA1p of SEQ ID NO:2, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO: 4, IMA1p of SEQ ID NO: 5, IMA1p of SEQ ID NO:33 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

For example the genotype IV may be the presence of at least 2 long alleles of IMA1. Said two long alleles of IMA1 may be genes encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least

90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In one embodiment the genotype IV may be the presence of at least 3 long alleles of IMA1. Said 3 long alleles of IMA1 may be genes encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In a preferred embodiment, the genotype IV may be the presence of at least 3 short alleles of IMA1 and at least 2 long alleles of IMA1, wherein

- a) said 3 short alleles of IMA1 individually are genes encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:1, IMA1p of SEQ ID NO:2, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO: 4, IMA1p of SEQ ID NO: 5, IMA1p of SEQ ID NO:33 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- b) said 2 long alleles of IMA1 individually are genes encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15, IMA1p of SEQ ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:23, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In one embodiment, the genotype IV may be that the yeast cell comprises at least 5 allelic genes encoding IMA1p, wherein said allelic genes individually are selected from the group consisting of genes encoding IMA1p of SEQ ID NO:1, IMA1p of SEQ ID NO:2, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO:4, IMA1p of SEQ ID NO:5, IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15, IMA1p of SEQ

ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:23, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and IMA1p of SEQ ID NO:33.

5 In one embodiment, the genotype IV may be that the yeast cell comprises the following 4 allelic genes:

- 1) a gene encoding IMA1p of SEQ ID NO:12 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 10 2) a gene encoding IMA1p of SEQ ID NO:13 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 3) a gene encoding IMA1p of SEQ ID NO:14 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%,
15 such as at least 98% sequence identity therewith; and
- 4) a gene encoding IMA1p of SEQ ID NO:15 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

20 In one embodiment, the genotype IV may be the presence of the following 4 allelic genes:

- 1) a gene encoding IMA1p of SEQ ID NO:12; and
- 2) a gene encoding IMA1p of SEQ ID NO:13; and
- 25 3) a gene encoding IMA1p of SEQ ID NO:14; and
- 4) a gene encoding IMA1p of SEQ ID NO:15.

In one embodiment, the genotype IV may be the presence of the following 3 allelic genes:

30

- 1) two genes both encoding IMA1p of SEQ ID NO:21 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and

- 2) a gene encoding IMA1p of SEQ ID NO:22 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

5 In one embodiment, the genotype IV may be the presence of the following 3 allelic genes:

- 1) a gene encoding IMA1p of SEQ ID NO:23 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%,
10 such as at least 98% sequence identity therewith; and
- 2) a gene encoding IMA1p of SEQ ID NO:24 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 3) a gene encoding IMA1p of SEQ ID NO:25 or a functional homologue thereof
15 sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In one embodiment, the genotype IV may be the presence of the following 5 allelic genes:

- 20 3) a gene encoding IMA1p of SEQ ID NO:12 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 4) a gene encoding IMA1p of SEQ ID NO:13 or a functional homologue thereof
25 sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 5) a gene encoding IMA1p of SEQ ID NO:1 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 30 6) a gene encoding IMA1p of SEQ ID NO:14 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and

- 7) a gene encoding IMA1p of SEQ ID NO:15 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

5

In one embodiment, the genotype IV may be the presence of the following 6 allelic genes:

- 10 1) a gene encoding IMA1p of SEQ ID NO:2 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) at least two genes encoding IMA1p of SEQ ID NO:3 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 15 3) at least two genes encoding IMA1p of SEQ ID NO:21 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 4) a gene encoding IMA1p of SEQ ID NO:22 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%,
20 such as at least 98% sequence identity therewith.

In one embodiment, the genotype IV may be the presence of the following 6 allelic genes:

- 25 1) a gene encoding IMA1p of SEQ ID NO:5 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) at least two genes encoding IMA1p of SEQ ID NO:33 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 30 3) at least two genes encoding IMA1p of SEQ ID NO:4 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and

- 4) a gene encoding IMA1p of SEQ ID NO:24 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 5) at least two genes encoding IMA1p of SEQ ID NO:23 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 6) a gene encoding IMA1p of SEQ ID NO:25 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

The yeast cell may for example have genotype IV in embodiments of the invention, where the yeast cell has characteristics I, II, IX and/or XI.

Genotype V

The yeast cell according to the invention may have the genotype V, wherein the genotype V is the presence of a gene encoding IMA5p. The genotype V may also be the presence of at least two allelic genes encoding IMA5p. In particular, it is preferred that the yeast cell according to the invention comprises at least one allelic gene encoding IMA5p selected from the group consisting of IMA5p of SEQ ID NO:16, IMA5p of SEQ ID NO:17 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith. Preferably, the genotype V is the presence of at least two genes encoding IMA5p of SEQ ID NO:16, or IMA5p of SEQ ID NO:17 or a functional homologue of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In one embodiment the yeast cell comprises at least two allelic genes encoding IMA5p individually selected from allelic genes encoding IMA5p of SEQ ID NO:16, IMA5p of SEQ ID NO:17, IMA5p of SEQ ID NO:34, IMA5p of SEQ ID NO:35, IMA5p of SEQ ID NO:36 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In particular, the genotype V may be that the yeast cell comprises the following 2 allelic genes:

- 5 1) a gene encoding IMA5p of SEQ ID NO:16 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) a gene encoding IMA5p of SEQ ID NO:17 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%,
10 such as at least 98% sequence identity therewith.

In one embodiment, the genotype V may be that the yeast cell comprises the following 3 allelic genes:

- 15 1) a gene encoding IMA5p of SEQ ID NO:16 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) a gene encoding IMA5p of SEQ ID NO:17 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%,
20 such as at least 98% sequence identity therewith; and
- 3) a gene encoding IMA5p of SEQ ID NO:34 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

25 In one embodiment, the genotype V may be that the yeast cell comprises the following 2 allelic genes:

- 1) a gene encoding IMA5p of SEQ ID NO:35 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%,
30 such as at least 98% sequence identity therewith; and
- 2) a gene encoding IMA5p of SEQ ID NO:36 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

In particular, the genotype V may be the presence of the following 2 genes:

- 1) a gene encoding IMA5p of SEQ ID NO:16; and
- 2) a gene encoding IMA5p of SEQ ID NO:17.

5

The yeast cell may for example have genotype V in embodiments of the invention, where the yeast cell has characteristics I, II, IX and/or XI.

Genotype VI

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The yeast cell according to the invention may have the genotype VI, wherein the genotype VI is the presence of at least 3 allelic genes encoding AGT1. In particular, it is preferred that the yeast cell according to the invention comprises at least 3 allelic genes encoding AGT1 selected from the group consisting of AGT1 of SEQ ID NO:18, AGT1 of SEQ ID NO:19, AGT1 of SEQ ID NO:20, AGT1 of SEQ ID NO:26, AGT1 of SEQ ID NO:27, AGT1 of SEQ ID NO:28, AGT1 of SEQ ID NO:29, AGT1 of SEQ ID NO:30, AGT1 of SEQ ID NO:31, AGT1 of SEQ ID NO:32 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

20

In one embodiment the yeast cell may have the genotype VI, wherein the genotype VI is the presence of at least 2 allelic genes encoding full length AGT1. In particular, it is preferred that the yeast cell according to the invention comprises at least 2 allelic genes encoding AGT1 selected from the group consisting of AGT1 of SEQ ID NO:18, AGT1 of SEQ ID NO:19, AGT1 of SEQ ID NO:20, AGT1 of SEQ ID NO:27, AGT1 of SEQ ID NO:28, AGT1 of SEQ ID NO:30, AGT1 of SEQ ID NO:31, AGT1 of SEQ ID NO:32 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

30

In one embodiment, the genotype VI may be that the yeast cell comprises the following 3 allelic genes:

- 1) a gene encoding AGT1 of SEQ ID NO:18 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) a gene encoding AGT1 of SEQ ID NO:19 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 3) a gene encoding AGT1 of SEQ ID NO:20 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

10

In particular, the genotype VI may be the presence of the following 3 allelic genes:

- 1) a gene encoding AGT1 of SEQ ID NO:18; and
- 2) a gene encoding AGT1 of SEQ ID NO:19; and
- 3) a gene encoding AGT1 of SEQ ID NO:20.

15

In one embodiment, the genotype VI may be that the yeast cell comprises at the following two genes encoding AGT1:

- 1) a gene encoding AGT1 of SEQ ID NO:27 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) a gene encoding AGT1 of SEQ ID NO:28 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

20

25

In one embodiment, the genotype VI may be that the yeast cell comprises the following 3 allelic genes encoding AGT1:

- 1) a gene encoding AGT1 of SEQ ID NO:30 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and

30

- 2) gene encoding AGT1 of SEQ ID NO:31 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 3) a gene encoding AGT1 of SEQ ID NO:32 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

The yeast cell may for example have genotype VI in embodiments of the invention, where the yeast cell has characteristics I, II, IX and/or XI.

Functional homologue

The term "functional homologue" as used herein denotes a polypeptide sharing at least one biological function with a reference polypeptide. In general said functional homologue also shares a significant sequence identity with the reference polypeptide. Preferably a functional homologue of a reference polypeptide is a polypeptide, which has the same biological function as the reference protein and which shares a high level of sequence identity with the reference polypeptide.

A high level of sequence identity indicates likelihood that the first sequence is derived from the second sequence. Amino acid sequence identity requires identical amino acid sequences between two aligned sequences. Thus, a candidate sequence sharing 80% amino acid identity with a reference sequence, requires that, following alignment, 80% of the amino acids in the candidate sequence are identical to the corresponding amino acids in the reference sequence. Identity according to the present invention is determined by aid of computer analysis, such as, without limitations, the ClustalW computer alignment program (Higgins D., Thompson J., Gibson T., Thompson J.D., Higgins D.G., Gibson T.J., 1994. CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. Nucleic Acids Res. 22:4673-4680), and the default parameters suggested therein. The ClustalW software is available from as a ClustalW WWW Service at the European Bioinformatics Institute <http://www.ebi.ac.uk/clustalw>. Using this program with its default settings, the mature (bioactive) part of a query and a

reference polypeptide are aligned. The number of fully conserved residues are counted and divided by the length of the reference polypeptide. Thus, sequence identity is determined over the entire length of the reference polypeptide.

5 It may be preferred that conserved amino acids are retained in the functional homologue. Conserved amino acids may be identified by preparing an alignment of similar polypeptide, and using said alignment identifying amino acids residues conserved between the polypeptides. Examples of useful alignments are shown herein in figures 5 -12.

10

Method for producing a beverage

It is an aspect of the invention to provide methods for producing a beverage, said method comprising the steps of

15

- VIII. Providing a starting liquid
- IX. Providing a yeast cell according to the invention, e.g. a yeast cell having one or more of characteristics I to X described above,
- X. Fermenting said starting liquid with said yeast cell,

20

thereby producing a beverage.

25

The starting liquid may in particular be a cereal extract, such as wort. Said starting liquid may for example be prepared by preparing an extract of malt by mashing and optionally sparging as described herein in this section.

30

Malt is barley kernels that have been malted. By the term "malting" is to be understood germination of steeped barley kernels in a process taking place under controlled environmental conditions, followed by a drying step. Said drying step may preferably be kiln drying of the germinated kernels at elevated temperatures.

This aforementioned sequence of malting events is important for the synthesis of numerous enzymes that cause grain modification, processes that principally depolymerize cell walls of the dead endosperm to mobilize the grain nutrients and

activate other depolymerases. In the subsequent drying process, flavour and colour are generated due to chemical browning reactions.

Steeping may be performed by any conventional method known to the skilled person.

5 One non-limiting example involves steeping at a temperature in the range of 10 to 25°C with alternating dry and wet conditions. Germination may be performed by any conventional method known to the skilled person. One non-limiting example involves germination at a temperature in the range of 10 to 25°C, optionally with changing temperature in the range of 1 to 4 h.

10

The kiln drying may be performed at conventional temperatures, such as at least 75°C, for example in the range of 80 to 90°C, such as in the range of 80 to 85°C. Thus, the malt may, for example be produced by any of the methods described by Briggs et al. (1981) and by Hough et al. (1982). However, any other suitable method for producing malt may also be used with the present invention, such as methods for production of specialty malts, including, but not limited to, methods of roasting the malt.

15

Malt may be further processed, for example by milling. Preferably milling is performed in a dry state, i.e. the malt is milled while dry.

20

The malt, e.g. the milled malt may be mashed to prepare an aqueous extract of said malt. The starting liquid for preparing the beverage may be an aqueous extract of malt, e.g. an aqueous extract of malt prepared by mashing.

25

Thus, the method for preparing a beverage according to the invention may comprise a step of producing wort by mashing malt and optionally additional adjuncts. Said mashing step may also optionally comprise sparging, and accordingly said mashing step may be a mashing step including a sparging step or a mashing step excluding a sparging step.

30

In general, wort production is initiated by the milling of malt and/or barley. If additional adjuncts are added, these may also be milled depending on their nature. If the adjunct is a cereal, it may for example be milled, whereas syrups, sugars and the like will generally not be milled. Milling will facilitate water access to grain particles in the

mashing phase. During mashing enzymatic depolymerization of substrates initiated during malting may be continued.

5 In general, wort is prepared by combining and incubating milled malt and water, i.e. in a mashing process. During mashing, the malt/liquid composition may be supplemented with additional carbohydrate-rich adjunct compositions, for example milled barley, maize, or rice adjuncts. Unmalted cereal adjuncts usually contain little or no active enzymes, making it important to supplement with malt or exogenous enzymes to provide enzymes necessary for polysaccharide depolymerization etc.

10

During mashing, milled malt and/or milled barley – and optionally additional adjuncts are incubated with a liquid fraction, such as water. The incubation temperature is in general either kept constant (isothermal mashing), or gradually increased, for example increased in a sequential manner. In either case, soluble substances in the
15 malt/barley/adjuncts are liberated into said liquid fraction. A subsequent filtration confers separation of wort and residual solid particles, the latter also denoted "spent grain". The wort thus obtained may also be denoted "first wort". Additional liquid, such as water may be added to the spent grains during a process also denoted sparging. After sparging and filtration, a "second wort" may be obtained. Further worts may be
20 prepared by repeating the procedure. Non-limiting examples of suitable procedures for preparation of wort is described by Briggs et al. (supra) and Hough et al. (supra).

As mentioned above, the wort composition may be prepared by mashing unmalted barley kernels. Unmalted barley kernels lack or contain only a limited amount of
25 enzymes beneficial for wort production, such as enzymes capable of degrading cell walls or enzymes capable of depolymerising starch into sugars. Thus, in embodiments of the invention where unmalted barley is used for mashing, it is preferred that one or more suitable, external brewing enzymes are added to the mash. Suitable enzymes may be lipases, starch degrading enzymes (e.g. amylases), glucanases [preferably (1-
30 4)- and/or (1-3,1-4)- β -glucanase], and/or xylanases (such as arabinoxylanase), and/or proteases, or enzyme mixtures comprising one or more of the aforementioned enzymes, e.g. Cereflo, Ultraflo, or Ondea Pro (Novozymes).

The wort composition may also be prepared by using a mixture of malted and unmalted barley kernels, in which case one or more suitable enzymes may be added during preparation. More specifically, barley of the invention can be used together with malt in any combination for mashing – with or without external brewing enzymes – such as, but
5 not limited to, the proportions of barley : malt = approximately 100 : 0, or approximately 75 : 25, or approximately 50 : 50, or approximately 25 : 75.

In other embodiments of the invention, it is preferred that no external enzymes, in particular that no external protease, and/or no external cellulase and/or no external
10 α -amylase and/or no external β -amylase and/or no external maltogenic α -amylase is added before or during mashing.

The wort obtained after mashing may also be referred to as “sweet wort”. In conventional methods, the sweet wort is boiled with or without hops where after it may
15 be referred to as boiled wort.

The term “approximately” as used herein means $\pm 10\%$, preferably $\pm 5\%$, yet more preferably $\pm 2\%$.

20 The wort may be heated or boiled before it is subjected to fermentation with the yeast of the invention. First, second and further worts may be combined, and thereafter subjected to heating or boiling. The wort may be heated or boiled for any suitable amount of time, e.g. in the range of 60 min to 120 min.

25 Thus, the starting liquid may be wort e.g. prepared as described above. The beverage may in be prepared by fermentation of the starting liquid, e.g. by fermentation of wort.

The beverage may in one preferred embodiment be malt beverages, even more preferred fermented beverages, such as fermented malt beverages, preferably
30 alcoholic beverages, such as beer

The beverage may be a non-alcoholic beverage, such as non-alcoholic beer or other kinds of non-alcoholic beverages, such as non-alcoholic malt beverages, such as maltina.

In one preferred embodiment the beverage is beer, for example the beer may be a lager beer or an ale. Thus, the beer may for example be selected from the group consisting of altbier, Amber ale, Barley wine, Berliner weisse, Bière de Garde, Bitter, 5 Blonde Ale, Bock, Brown ale, California Common, Cream Ale, Dortmunder Export, Doppelbock, Dunkel, Dunkelweizen, Eisbock, Fruit lambic, Golden Ale, Gose, Gueuze, Hefeweizen, Helles, India pale ale, Kölsch, Lambic, Light ale, Maibock, Malt liquor, Mild, Märzenbier, Old ale, Oud bruin, Pale ale, Pilsener, Porter, Red ale, Roggenbier, Saison, Scotch ale, Steam beer, Stout, Schwarzbier, lager, Witbier, Weissbier and 10 Weizenbock.

Thus, the invention also relates to methods of producing a beverage comprising the steps of:

- 15 (i) providing a malt composition;
 (ii) processing said malt composition into a beverage.

In general terms, alcoholic beverages – such as beer – may be manufactured from malted and/or unmalted barley grains. Malt, in addition to hops and yeast, contributes 20 to flavour and colour of the beer. Furthermore, malt functions as a source of fermentable sugar and enzymes. Non-limited descriptions of examples of suitable methods for malting and brewing can be found, for example, in publications by Briggs et al. (1981) and Hough et al. (1982). Numerous, regularly updated methods for analyses of barley, malt and beer products are available, for example, but not limited 25 to, American Association of Cereal Chemists (1995), American Society of Brewing Chemists (1992), European Brewery Convention (1998), and Institute of Brewing (1997). It is recognized that many specific procedures are employed for a given brewery, with the most significant variations relating to local consumer preferences. Any such method of producing beer may be used with the present invention.

30

The first step of producing beer from wort preferably involves heating said wort as described herein above, followed by a subsequent phase of wort cooling and optionally whirlpool rest. After being cooled, the wort may be transferred to fermentation tanks containing yeast according to the invention, i.e. yeast having one or more of

characteristics I to X described above. The wort will be fermented for any suitable time period, in general in the range of 1 to 100 days. The fermentation is performed at any useful temperature e.g. at a temperature in the range of 10-20°C.

- 5 During the several-day-long fermentation process, sugar is converted to alcohol and CO₂ concomitantly with the development of some flavour substances.

Subsequently, the beer may be further processed, for example chilled. It may also be filtered and/or lagered – a process that develops a pleasant aroma and a less yeasty
10 flavour. Also additives may be added. Furthermore, CO₂ may be added. Finally, the beer may be pasteurized and/or filtered, before it is packaged (e.g. bottled or canned).

The beer produced by fermentation with the yeast according to the invention in general has a superior pleasant taste. Taste may be analyzed, for example, by a specialist beer
15 taste panel. Preferably, said panel is trained in tasting and describing beer flavours, with special focus on aldehydes, papery taste, old taste, esters, higher alcohols, fatty acids and sulphury components.

In general, the taste panel will consist of in the range of 3 to 30 members, for example in the range of 5 to 15 members, preferably in the range of 8 to 12 members. The taste
20 panel may evaluate the presence of various flavours, such as papery, oxidized, aged, and bready off-flavours as well as flavours of esters, higher alcohols, sulfur components and body of beer.

Sequence listing

SEQ ID NO:1	Amino acid sequence of IMA1p short encoded by a unique allele from Hybrid yeast 1.
SEQ ID NO:2	Amino acid sequence of _IMA1p short_from Hybrid yeast 4
SEQ ID NO:3	Amino acid sequence of IMA1p short_from Hybrid yeast_4
SEQ ID NO:4	Amino acid sequence of IMA1p short from Hybrid yeast 7
SEQ ID NO:5	Amino acid sequence of IMA1p short from Hybrid yeast 7
SEQ ID NO:6	Amino acid sequence of DAL5 encoded by Sc allele from Hybrid yeast 1
SEQ ID NO:7	Amino acid sequence of PTR2 encoded by Sc allele from Ale

	yeast 1
SEQ ID NO:8	Amino acid sequence of PTR2 encoded by Sc allele from Lager yeast 1
SEQ ID NO:9	Amino acid sequence of PTR2 encoded by non-Sc allele from Hybrid yeast 1.
SEQ ID NO:10	Partial amino acid sequence of UBR1 encoded by Sc allele from Hybrid yeast 1
SEQ ID NO:11	Amino acid sequence of UBR1 encoded by non-Sc allele from Hybrid yeast 1
SEQ ID NO:12	Amino acid sequence of IMA1p encoded by short Sc allele from Hybrid yeast 1
SEQ ID NO:13	Amino acid sequence of IMA1p encoded by short Sc allele from Hybrid yeast 1
SEQ ID NO:14	Amino acid sequence of IMA1p encoded by the long Sc allele from Hybrid yeast 1
SEQ ID NO:15	Amino acid sequence of IMA1p encoded by a long Sc allele from Hybrid yeast 1
SEQ ID NO:16	Amino acid sequence of IMA5p from Hybrid yeast 1 encoded by non-Sc IMA5 like
SEQ ID NO: 17	Amino acid sequence of IMA5p from Hybrid yeast 1 encoded by Sc-IMA5 like
SEQ ID NO:18	Amino acid sequence of AGT1 from Hybrid yeast 1 encoded by non-Sc allele
SEQ ID NO:19	Amino acid sequence of AGT1 from Hybrid yeast 1 encoded by an Sc allele
SEQ ID NO:20	Amino acid sequence of AGT1 from Hybrid yeast 1 encoded by an Sc allele
SEQ ID NO:21	Amino acid sequence of IMA1p from Hybrid yeast 4 encoded by long IMA1 allele
SEQ ID NO:22	Amino acid sequence of IMA1p from Hybrid yeast 4 encoded by a long IMA1 allele
SEQ ID NO:23	Amino acid sequence of IMA1p from Hybrid yeast 7 encoded by a long IMA1 allele

SEQ ID NO:24	Amino acid sequence of IMA1p from Hybrid yeast 7 encoded by a long IMA1 allele
SEQ ID NO:25	Amino acid sequence of IMA1p from Hybrid yeast 7 encoded by a long IMA1 allele
SEQ ID NO:26	Amino acid sequence of truncated AGT1 from Hybrid yeast 4 encoded by an Sc allele
SEQ ID NO:27	Amino acid sequence of AGT1 from Hybrid yeast 4 encoded by a non-Sc allele
SEQ ID NO:28	Amino acid sequence of AGT1 from Hybrid yeast 4 encoded by a non-Sc allele
SEQ ID NO:29	Amino acid sequence of truncated AGT1 from Hybrid yeast 7 encoded by an Sc allele
SEQ ID NO:30	Amino acid sequence of AGT1 from Hybrid yeast 7 encoded by an Sc allele
SEQ ID NO:31	Amino acid sequence of AGT1 from Hybrid yeast 7 encoded by a non-Sc allele
SEQ ID NO:32	Amino acid sequence of AGT1 from Hybrid yeast 7 encoded by a non-Sc allele
SEQ ID NO:33	Amino acid sequence of IMA1p short from Hybrid yeast 7
SEQ ID NO:34	Amino acid sequence of IMA5 from Hybrid yeast 1
SEQ ID NO:35	Amino acid sequence of IMA5 from Hybrid yeast 7
SEQ ID NO:36	Amino acid sequence of IMA5 from Hybrid yeast 7
SEQ ID NO:37	Partial amino acid sequence of PTR2 from Hybrid yeast 7
SEQ ID NO:38	Amino acid sequence of PTR2 of Hybrid yeast 7
SEQ ID NO:39	Amino acid sequence of DAL 5 of Hybrid yeast 7
SEQ ID NO:40	Amino acid sequence of DAL5 of Hybrid yeast 7
SEQ ID NO:41	Partial amino acid sequence of UBR1 of Hybrid yeast 7
SEQ ID NO:42	Amino acid sequence of UBR1 of Hybrid yeast 7
SEQ ID NO:43	Amino acid sequence of PTR2 of Hybrid yeast 1
SEQ ID NO:44	Amino acid sequence of PTR2 of Hybrid yeast 1
SEQ ID NO:45	Partial amino acid sequence of UBR1 of Hybrid yeast 1

SEQ ID NO:1

Amino acid sequence of SHORT_IMA1_from Hybrid yeast_1

MGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSHEWFKESRSSKTNPKRD
WFFWRPPKGYDAEGKPIPPNNWKS YFGGSAWTFDEKTQEFYLR LFCSTQPD LNWENEDCRKAIY
5 ESAVGYWLDHGV DGRIDVGS LYSKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQEMNQF
IRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSHTDVGTSPLFRYNLVPFELKD
WKIALAELFRYINGTDCWSTIYLENHDQPR SITRFGDDSPKNRVISGKLLSVLLSALTGTLYVY
QGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEGIALVSRDHARTPMPWTP
NEPNAGFSGPNTKPFYLNESFRQGINVEEEQKNSDSVLA FWKKALEFRKNHKDI AVYGFDKF
10 IDLDNKKLFSFTKRYNNKTLFAALNFSSDATDFKIPNDGSSFKLEFGNYPKNEVDASSRTLKPW
EGRIHINE.

SEQ ID NO:2

Amino acid sequence of SHORT_IMA1_from Hybrid yeast 4

15 MGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSHEWFKESRSSKTNPKRD
WFFWRPPKGYDAEGKPIPPNNWKS YFGGSAWTFDEKTQEFYLR LFCSTQPD LNWENEDCRKAIY
ESAVGYWLDHGV DGRIDVGS LYSKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQEMNQF
IRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSHTDVGTSPLFRYNLVPFELKD
WKIALAELFRYINGTDCWSTIYLENHDQPR SITRFGDDSPKNRVISGKLLSVLLSALTGTLYVY
20 QGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEGIALVSRDHARTPMPWTP
NEPNAGFSGPNTKPFYLNESFRQGINVEEEQKNSDSVLA FWKKALEFRKNHKDI AVYGFDKF
IDLDNKKLFSFTKRYNNKTLFAALNFSSDATDFKIPNDGSSFKLEFGNYPKNEVDASSRTLKPW
EGRIYINE

25 SEQ ID NO:3

Amino acid sequence of SHORT_IMA1_from Hybrid yeast_4_

MGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSHEWFKESRSSKTNPKRD
WFFWRPPKGYDAEGKPIPPNNWKS YFGGSAWTFDEKTQEFYLR LFCSTQPD LNWENEDCRKAIY
ESAVGYWLDHGV DGRIDVGS LYSKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQEMNQF
30 IRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSHTDVGTSPLFRYNLVPFELKD
WKIALAELFRFINGTDCWSTIYLENHDQPR SITRFGDDSPKNRVISGKLLSVLLSALTGTLYVY
QGQELGQINFKNWSVEKYEDVEIRNNYRLIKEECGENSEEMKKFLEGIALVSRDHARTPMPWTP
NEPNAGFSGPNTKPFYLNESFRQGINVEEEQKNSDSVLA FWKKALEFRKNHKDI AVYGFDKF
IDLDNKKLFSFTKRYNNKTLFAALNFSSDATDFKIPNDGSSFKLEFGNYPKNEVDASSRTLKPW
35 EGRIHINE.

SEQ ID NO:4

Amino acid sequence of SHORT_IMA1_from Hybrid yeast_7_

40 MGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSHEWFKESRSSKTNPKRD
WFFWRPPKGYDAEGKPIPPNNWKS YFGGSAWTFDEKTQEFYLR LFCSTQPD LNWENEDCRKAIY
ESAVGYWLDHGV DGRIDVGS LYSKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQEMNQF
IRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSHTDVGTSPLFRYNLVPFELKD
WKIALAELFRYINGTDCWSTIYLENHDQPR SITRFGDDSPKNRVISGKLLSVLLSALTGTLYVY
QGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEGIALVSRDHARTPMPWTP
45 NEPNAGFSGPNTKPFYLNESFRQGINVEEEQKNSDSVLA FWKKALEFRKNHKDI AVYGFDKF
IDLDNKKLFSFTKRYNNKTLFAALNFSSDATDFKIPNDGSSFKLEFGNYPKNEVDVSSRTLKPW
EGRIYINE.

SEQ ID NO:5

50 Amino acid sequence of SHORT_IMA1 from_Hybrid_7_

MGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSSEHEWFKESRSSKTNPKRD
WFFWRPPKGYDAEGKPIPPNNWKS YFGGSAWTFDEKTQEFYLR LFCSTQPD LNWENEDCRKAIY
ESAVGYWLDHGV DGRIDV GSLY SKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQEMNQF
IRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNF SHTDVGTSPLFRYNLVPFELKD
WKIALAELFRYINGTDCWSTIYLENHDQPR SITRFGDDSPKNRVISGKLLSVLLSALTGTLYVY
QGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEGIALVSRDHARTPMPWTP
NEPNAGFSGPNTKPFYLNESFRQGINVEEEQKNSDSVLAFWKKALEFRKNHKDIAVYGFDFKF
IDLDNKKLFSFTKRYNNKTLFAALNFSSDATDFKIPNDGSSFKLEFGNYPKNEVDASSRTLKPW
EGRIYINE.

SEQ ID NO:6

Amino acid sequence of DAL5 encoded by Sc allele from Hybrid yeast 1

MSADASTNSNASLDEKNLNITSEAEIKNEDVTAEPVLSTVLSPNGKIVYISDKVDEAMKLAEEA
KEIEVTPPEEDRKLRLWKIDYCMFPLMCILYAVQFMDKISTSSAAMGLRTDLKMHGDQYSWVTS
FYFGYLFMNLGPVQFIFQRTSHMSKMLAVFIVIWGMLLALHAAPT VKYPSFIVLRVLLGCAESV
VTPCFITIIITAQYWKTEEQFTRVSIWFGMNLG SILINAIAYGVYIHQDSYAIKGWRTLFVITGV
ITIFIGILIFLWIPDDPSKARFLSKREKLMVVQRIRSNQQGFGNHEIKKYQIIEALKDVRTWLY
FLFTVSSNIPNGGISSFMSILLNSDFGYSSKETLLMGLPTGAVELVGCPLFGILAVYAANKKIP
FWKYKLSWAIFA AVLALIASCMLGFATNSKKARLAGAYLWYISPVSFICVLSNISANSSGYSK
WTVSSINLVAYAAAANLAGPQTFIAKQAPKYHGAKVAMVVCYAVMIVLLSILLIVNLRENKRDK
IAAERGFP EETENLEFSDLTDFENPNFRYTL.

SEQ ID NO:7

Amino acid sequence of PTR2 of encoded by Sc allele from Ale yeast 1

MLNHPSQGSDDAQDEKQGDFFVIEEEKTQAVMLKDSYVSDDVANSTERYNLSPSPEDEDFEAPT
EEEMQTLRHVGGKIPMRCWLI AIVELSERFSYYGLSAPFQNYMEYGPNDSPKGVLSLNSQGATG
LSYFFQFWCYVTPVFGGYVADTFWGKYNTICCGTAIYIAGIFILFITSIPSVGNRDSAIGGFIA
AIIILGIATGMIKANLSVLIADQLPKRKPSIKVLKSGERVIVDSNITLQNVFMFFYFMINVGS
SLMATTELEYHKGFWAAYLLPFCFFWIAVVT LIFGKKQYIQRPIGDKVIAKSFKVCWILT KNKF
DFNAAKPSVHPEKNYPWNDKFVDEIKRALAACKVFI FYPIYWTQYGTMISSFITQASMMELHGI
PNDFLQAFDSIALIIFIPIFEKFVYPFIRRYTPLKPITKIFXGFMFGSFAMTWA AVLQS FVYKA
GPWYNEPLGHNTPNHVHVCWQIPAYVLISFSEIFASITGLEAYASKAPASMKS FIMSIFLLTNA
FGSAIGCALSPVTVDPKFTWLTGLAVACFISGCLFWLCFRKYNDTEEEMNAMDYEEENEFDLN
PISAPKANDIEILEPMDSLRSTAKY.

SEQ ID NO:8

Amino acid sequence of PTR2 encoded by Sc allele from Lager yeast 1

MLNHPSQGSDDAQDEKQGDFFVIEEEKTQAVTLKDSYVSDDVANSTERYNLSPSPEDEDFEAPT
EEEMQTLRHVGGKIPMRCWLI AIVELSERFSYYGLSAPFQNYMEYGPNDSPKGVLSLNSQGATG
LSYFFQFWCYVTPVFGGYVADTFWGKYNTICCGTAIYIAGIFILFITSIPSVGNRDSAIGGFIA
AIIILGIATGMIKANLSVLIADQLPKRKPSIKVLKSGERVIVDSNITLQNVFMFFYFMINVGS
SLMATTELEYHKGFWAAYLLPFCFFWIAVVT LIFGKKQYIQRVPGDKVIAKSFKVCWILT KNKF
DFNAAKPSVHPEKNYPWNDKFVDEIKRALAACKVFI FYPIYWTQYGTMISSFITQASMMELHGI
PNDFLQAFDSIALIIFIPIFEKFVYPFIRRYTPLKPITKIFFGFMFGSFAMTWA AVLQS FVYKA
GPWYNEPLGHNTPNHVHVCWQIPAYVLISFSEIFASITGLEAYASKAPASMKS FIMSIFLLTNA
FGSAIGCALSPVTVDPKFTWLTGLAVACFISGCLFWLCFRKYNDTEEEMNAMDYEEENEFDLN
PISAPKANDIEILEPMESLRSTTKY

SEQ ID NO:9

Amino acid sequence of PTR2 encoded by non-Sc allele from Hybrid yeast 1

MLNHL SQSDDIQDEKQGDFFVIEEEKNQTVTLKDSYVSDDAANSTEHYNLSPSLEEDEF EAPT
 5 DEELRSLRHVGGKIPMRCWLI AIVELSERFSYYGLSAPFQNYMEYGPKDTPKGVLSLNSQGATG
 LSYFFQFWCYVTPVFGGYVADTFWGKYNTICCGTAIYIAGIFILLITSIPSVGNRDSALGGFIA
 SIILIGIATGMIKANLSVLIADQLPKRKPSIKVLKSGERVIVDSNITLQNVFMFFYFMINVGSL
 SLMATTELEYHKGFWAAYLLPFCFFWVAVVTLVFGKKQYIQRPIGDKVIAKSFRVCWILT KKNF
 DFNAAKPSVHPEKEYPWNDKFVDEIKRALAACKVFVFYPIYWTQYGTMISSFITQAGMMELHGI
 10 PNDFLQAFDSIALIIFIPIFEKFIYPFIRRYTPFKPITKIFFGFMFGLAMTWAAVLQSFVYKA
 GPWYSAPLGHNTPNHVHVCWQIPAYVLISFSEIFASITGLEAYASKAPASMKSFIMSIFLLTNA
 FGSAIGCALSPVTVDPKFTWLFTGLAVACFISGCLFWFCFRKYNDTEEEMNAMDYEEDEFDLN
 PISQPKGNDIEILEPMGSLKSTTKY

SEQ ID NO:10

15 Incomplete amino acid sequence of UBR1 encoded by Sc allele of
 Hybrid yeast 1
 FKEFCKVEGGVLIWQRVQKSNLT KSYSISFKQGLYTVETLLSKVHDPNIPLRPKEIISLLTLCK
 LFNGAWKIKRKEGEHVLHEDQNFISYLEYTTTSIYSIIQTAEKVSEKSKDSIDSKLFLNAIRIIS
 SFLGNRSLTYKLIYDSHEVIKFSVSHERVA FMNPLQTMLSFLIEKVSLKDAYEALED CSDFLKI
 20 SDFSLRSVVLCSQIDVGFWVRNGMSVLHQASYKKNPELGSYSRDIHLNQLAILWERDDIPRII
 YNILDREWELLDWFTGEVDYQHTVYEDKISFIIQQFIAFIYQILTERQYFKTFSSLKDRRMDQIK
 NSIIYNLYMKPLSYSKLLRSVPDYLTDTEFDEALEEVS VFVEPKGLADNGVFKLKASLYAKV
 DPLKLLNLENEFESS

SEQ ID NO:11

25 Amino acid sequence of UBR1 encoded by non-Sc allele of Hybrid
 yeast 1
 MSFTDNLGLSLKAHIRRTLRSIHNLPYFRFTRGPTERADMSRALKEFIYRYLYFIISNDGENLS
 TLFTAHPKQKSSNQELAVFPESLEDALDVDKITSQGTFFPYKIDESKIGDVHKHTGRNCGRKFK
 30 IGEPLYRCHECGDDTCVLCIHC FNPKDHNHVVCTDICSEFTSGICDCGDEEAWNSSLHCKAE
 EQGNDTSEDPSNFDSTKQKDVWNP ECIALVELVLSEVFDYFIDVFNQNI EPLPTIQKDITIKL
 REMTQQGKMYERAQFLNDLKYENDYMF DGTTTAKTSPSNSPEASPSLAKIDPENYTVIIYNDEY
 HNYSQATTALRQGVDPNVHIDLLTSRIDGEGRAMLKCSQDLSSVLGGFFAVQTNGLSATLTSWS
 EYLHQEACKYIILWITHCLNIPNPSFQITFRNMMGKSLCSEYLNATESRDMPVVEKYFSTKFD
 35 KDDPYRYIDLSVLAEGNQIPLGHHKVLPESS THSLSTLINDVENLTSKEYSNTRLQHILYFDNR
 YWKRLRKDIQNVIIPTLASSTLYKPIFCQQVVEIFNHITRSVAYMDREPQLTAIRECVVQLFTC
 PTNTRNIFENQSFLDILWSIIDIFKEFCKVEAGVLIWQRVQKSNLT KSYSLSFKQGLYTVETLL
 SKVNDPNITIRPKVFISLLTLGKLFNGAWKIKRKEGEHVLHEDQNFISYLEYTTTSIYSIIQTAE
 KVLEKSHDSL DLNLVLNAIRIVSSFLGNRSLTYKLIYDSHEIIKFSVSHERVA FMNPIQTMLS F
 40 LIEKVSLKDAYESLENC PDLKIADFLSLRSVVLCSQIDVGFWVRNGMSVLHQASYKKNPELGS
 YSRDIHLNQLAI IWERDDLPRVIYNILDRWELLDWFMGEAEYQHTVYEDKISFMIQQFIAFIYQ
 ILTERQYFKTFSLLRDRRMDMIKNSIMYNLYMKPLSYSKLLKSVPDYLTDTEFDEALEEVS V
 FVEPKGLADNGVFKLKAALYAKIDPLKLLNLENEFESSATIIKTHLAKNKDEVSKVVLIPQVST
 KLLDKGAMNLGEFTRNTVFAKVIYKLLQVCLDMEDSTFLNELLHLVHGIFKDELINGKDSIPE
 45 AYLAKPICNLLLSIANAKSDIFSESIVRKADYLLEKMIMKKPDEIFESLIASFGNQYIDNYKDK
 KLSQGVNLQETEKERKRMAKKHQARLLAKFNNQSKFMKEHESEFDEQDNDVDMDGEKVYESE
 DFTCALCQDSSSTDFFVIPAYHDHTPIFRPGNI FNPREFMAKWDGFYNDDDKQAYIDDEVLES L
 KENGTGRSRKV FVSCNHIIHNCFKRYVQKKRFSSNAFICPLCQTFSNCTLPICPTSRANTGLS
 LDMFLKSELSDLILSRLFKPFTEDNYRTINSIFSLMVSQCQGFDKVVRKHVNFTHKDVSLVLSV
 50 HWANTISMLEVASRLEKPHNISFFRSREQYKTKLNILICIMLFTFVIGKPSMEFEPYPVESDI
 ICNQNLQFYIVRKSLFSPASLRETITEALT VFCQFLDDFVQGLSDAEQVDKLYTEAKKLGDV
 YNVDESILITLMSITVVKTEGLESRSIYDLAYTSLLKSLLP TIRRLVMVKVLHEL VKDSENET

5 MVIDGFDVEEELEFEGLPGFVDKALKLITDKESFVDLFKTKQAIVPSHPYLERIPYEYCGIVKL
IDLSKFLNTYVTQSKEIKLREERSQHMKNADNRLDFKICLTGKVKVHLRADRHMTKHLNKNCF
KSFGAFLMPNSSEVCLHLTQPPSNIFVSAPYLNHGEVGRNAMRRGDLTTLNLKRYEHLNRLWI
NNEIPGYISRVMGDEFRVTILSNGFLFAFNREPRPRVPPTDEDDMEEGEGFFTEENDMD
VDDETGQAANLFGVGAEGIGDGGVRNFFQFFENFRNTLQPPQGNDDAPQNPPILQFLGPQFD
GATIIRNTNQRLDEDDSSSENDSDEREIW

SEQ ID NO:12
Amino acid sequence of IMA1p encoded by short Sc allele from
10 Hybrid yeast 1
MGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSHEWFKESRSSKTNPKRD
WFFWRPPKGYDAEGKPIPPNNWKSYPGGSAAWTFDEKTQEFYLRLLFCSTQPDNLWENEDCRKAIY
ESAVGYWLDHGVDFRIDVGSLSYKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQEMNQF
IRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSTDVGTSPLFRYNLVPFELKD
15 WKIALAELFRFINGTDCWSTIYLENHDQPRISITRFGDDSPKNRVISGKLLSVLLSALTGTLYVY
QGQELGQINFKNWSVEKYEDVEIRNNYRLIKEECGENSEEMKKFLEGIALVSRDHARTPMPWTP
NEPNAGFSGPNTKPFYLNESFRQGINVEEEQKNSDSVLAFWKKALEFRKNHKDIAVYGFDFKF
IDLDNKKLFSFTKRYNNKTLFAALNFSSDATDFKIPNDGSSFKLEFGNYPKNEVDASSRTLKPW
EGRIHINE

20 SEQ ID NO:13
Amino acid sequence of IMA1p encoded by short Sc allele from
Hybrid yeast 1
MGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSHEWFKESRSSKTNPKRD
25 WFFWRPPKGYDAEGKPIPPNNWKSYPGGSAAWTFDEKTQEFYLRLLFCSTQPDNLWENEDCRKAIY
ESAVGYWLDHGVDFRIDVGSLSYKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQEMNQF
IRNRVKGGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSTDVGTSPLFRYNLVPFELKD
WKIALAELFRFINGTDCWSTIYLENHDQPRISITRFGDDSPKNRVISGKLLSVLLSALTGTLYVY
QGQELGQINFKNWSVEKYEDVEIRNNYRLIKEECGENSEEMKKFLEGIALVSRDHARTPMPWTP
30 NEPNAGFSGPNTKPFYLNESFRQGINVEEEQKNSDSVLAFWKKALEFRKNHKDIAVYGFDFKF
IDLDNKKLFSFTKRYNNKTLFAALNFSSDATDFKIPNDGSSFKLEFGNYPKNEVDASSRTLKPW
EGRIHINE

35 SEQ ID NO:14
Amino acid sequence of IMA1p encoded by long Sc allele from
Hybrid yeast 1
MTISSAHPETEPKWWKEATFYQIYPASFKDSNDDGWGDMKGISSKLEYIKELGVDAIWISPLYD
SPQDDMGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSHEWFKESRSSKT
NPKRDWSFWRPPKGYDAEGKPIPPNNWKSYPGGSAAWTFDEKTQEFYLRLLFCSTQPDNLWENEDC
40 RKAIYESAVGYWLDHGVDFRIDVGSLSYKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQ
EMNQFIRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSTDVGTSPLFRYNLVP
FELKDWKIALAELFRYINGTDCWSTIYLENHDQPRISITRFGDDSPKNRVISGKLLSVLLSALTG
TLYVYQGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEGIALVSRDHARTP
MPWTPNEPNAGFSGPSAKPWFYLNDSFREGINVEDEIKDPNSVLNFWKEALKFRKAHKDITVYG
45 YDFEFIDLDNKKLFSFTKKYNNKTLFAALNFSSDATDFKIPNDSSFKLEFGNYPKKEVDASSR
TLKPWEGRYIIE

SEQ ID NO:15
Amino acid sequence of IMA1p encoded by long Sc allele from
50 Hybrid yeast 1
MTISSAHPEAEPKWWKEATFYQIYPASFKDSNDDGWGDMKGISSKLEYIKELGADAIWISPFYD
SPQDDMGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSHEWLKESRSSKT

5 NPKRDWFFWRPPKGYDAEGKPIPPNNWKS YFGGSAWIFDEKTQEFYLR LFCSTQPD LNWENEDC
RKAIYESAVGYWLDHGV DGFRI DVGSLY SKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQ
EMNQFIRNRVKDGREIMTVGEMQHASDETKKLYTSASRHELSELFNFSHTDVGTSPLFRYNLVP
FELKDWKIALAELFRFINGTDCWSTIYLENHDQPR SITRFGDDSPKNRVISGKLLSVLLSALTG
TLYVYQGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEAIALISR DHARTP
MQWSREEPNAGFSGPSAKPW FYLND SFREGINVEDEIKDPNSVLNFWKEALKFRKAHKDITVYG
YDFEFIDL DNKKLFSFTKKYNNKTLFAALNFSSDATDFKI PNDDSSFKLEFGNYPKKEVDASSR
TLKPWEGR IYISE

10 SEQ ID NO:16

Amino acid sequence of IMA5p from Hybrid yeast 1 encoded by non-Sc IMA5 like allele

15 MTIIHNPKWWKEAT IYQIYPASF KDSNNDGWGDLAGITSKLDYIKELGVDAIWVCPFYDSPQED
MGYDIANYEKVWPRYGTSEDCFQMI EESHKRG IKVIVDLVINHCSEEHEWFKESRSSKTNAKRD
WFFWKPPKGYEIDGTPIPPNNWRSFFGGS AWKYDENTEEFFLHV FAPGQPDFN WENKECRQAIY
DSSVGFWLRHNVDGFRIDVGSMYSKVEGLPDASITDPTVPYQDGTDF FVNGPRIHEYHKEMRQY
MYTQIPEGKEIMTVGEVGIGNEKDFKDYTS SKEEEFNMMFNFKHTSVGESPEFKYELIPFTLKD
FKLALAESFLFIEGTDCWSTIYLENHDQPRSVSRFGSDSPEWREISSKMLATLIISLTGT VFIY
20 QGQELGMPNFKNRKIEQIKCVEGTGT YGAIKRDYGEDSEKMKKFYEALALISR DHGRTPFPWSG
EKPYAGFSKNAKPWIDINESFVEGINAEAE LNDENS VFFFWKRALQVRKEHKMMLVYGDNFQFY
DL DNEKLFMFTKDSGDKMFAVFNFCSDSTEFSPDNKASYDMFFGNYANS DGKSYTLKPWEGR
LYYSN

SEQ ID NO: 17

25 Amino acid sequence of IMA5p from Hybrid yeast 1 encoded by Sc-IMA5 like

MTIIHNPKWWKEATVYQIYPASN KDSNNDGWGDLAGITSKLDYVKELGVDAIWVCLFYDSPQED
MGYDIANYEKVWPRYGTNEDCFQMI EEAHKRG IKVIVDLVINHCSEEHEWFKESKSSKTNPKRD
WFFWRPPKGFDEKGNPIPPNNWRSFFGGS AWRYDEKTGEFFLHV FAPGQPDFN WENEECRKAIY
30 DSSVGYWLRHNVDGFRIDVGSMYSKVEGLPDAPITDPTVPYQKGTEFFINGSR IHEYHKEMRKY
MLSQIPEGKEIMTVGEVGVGNEEDFRDYTS AKEGELNMMFNFKHTSVGESPECKYELIPFTLKD
FKLALAESFLFIENTDCWSTIYLENHDQPRSVSRFGSDSPKWRAISSKMLATLIISLTGT VFIY
QGQELGMSNFKNRRIEQIKCVEGTGT YAAIKRDYGEDSEKMKKFFEALALISR DHGRTPFPWSA
DEPSAGFSKDAKPRIDMNESFRDGINAEAE LKDN SVFFFWKKALQVRKEHKDILVYGHNFQFI
35 DL DNDKLFMFTKDTDNKKMFAVFNFSDDTDFSPDNEASYTMFFGNYANSNGDSRTLQPWEGR
LYLLK

SEQ ID NO:18

40 Amino acid sequence of AGT1 from Hybrid yeast 1 encoded by non-Sc allele

MKNILSLVGRKENTPEDVTANLADTSSTTVMQAKDLVIEDFEERKKND AFELNHLELT TNATQL
SDSDEDKENVIRVAEATDDANEANNEEKSMTLRQALRKYPKAALWSILVSTTLVMEGYDTALLS
ALYALPVFQRKF GTMNAEGSYEITSQWQIGLNM CVLCGEMIGLQITTYMVEFMGNRYTMITALS
LLTAYIFILYYCKSLAMIAVGQILSAMPWGC FQSLAVTYASEVCPLALRYMTSYSNICWLFQ
45 IFASGIMKNSQENLGNSDLGYKL PFALQWIWPAPLIIGIFFAPESPWWLVRKNKIVEAKKSLNR
ILSGTVTEKEIQVDITLQIEMTIEKERLRASKSGSFFSCFKGVDGRRTRLACL TWVAQNSSGA
VLLGYSTYFFERAGMATDKAFTFSLIQYCLGLAGTLG SWVISGRVGRWTILTYGLSFQMVCLFI
IGGMGFASGSSASNAAGLLLLALSFFYNAGIGAVVYCI VAEIPSAELRTKTIVLARICYNLMAV
FNAILT PYMLNVSDWNWGAKTGLYWG GFTALT LAWVIIDLPETTGRTFSEINELFSQGV PARKF
50 ASTVVDPF GKRLQNR PQVDNIIDRFSSASQQAL

SEQ ID NO:19

Amino acid sequence of AGT1 from Hybrid yeast 1 encoded by an Sc allele

5 MKNIIISLVSKKKAASKNEDKNISESSRDIVNQQEVFNTENFEEGRKDSAFELDHLEFTINSAQL
 GDSDEDNENVINETNTTDDANEANSEEKSM TLKQALLIYPKAALWSILVSTTLVMEGYDTALLN
 10 ALYALPVFQQRKFGLNGEYSYEITSQWQIGLNM CVQCGEIIGLQITPYMVEFMGNRYTMITALG
 LLTAYVFILYYCKSLAMIAVGQVLSAMPWGC FQGLT VTYASEVCPLALRYMTSYSNICWLFQ
 IFASGIMKNSQENLGNSDLGYKL PFALQWIWPAPLMIGIFFAPESPWWLVRKDRVAEARKSLSR
 ILSGKGAEKDIQIDLT LKQIELTIEKERLLASKSGSFLDCFKGVNRRTRLACLTWVAQNTSGA
 15 CLLGYSTYFFERAGMATDKAFTFSVIQYCLGLAGTLC SWVISGRVGRWTILTYGLAFQMVCLFI
 IGGMGFGSGSGASNGAGGLLLALSFFYNAGIGAVVY CIVTEIPSAELRTKTIVLARICYNIMAV
 INAILTPYMLNVSDWNWGAKTGLYWGGFTAVTLAWVIIDL PETSGRTFSEINELFNQGV PARKF
 ASTVVD PFGKGKTQHDSLDDESISQSSSIKQRELNAADKC

SEQ ID NO:20

15 Amino acid sequence of AGT1 from Hybrid yeast 1 encoded by an Sc allele
 MKNIIISLVSKKKAASKNEDKNISESSRDIVNQQEVFNTENFEEGRKDSAFELDHLEFTINSAQL
 GDSDEDNENVINETNTTDDANEANSEEKSM TLKQALLIYPKAALWSILVSTTLVMEGYDTALLN
 20 ALYALPVFQQRKFGLNGEYSYEITSQWQIGLNM CVQCGEIIGLQITPYMVEFMGNRYTMITALG
 LLTAYVFILYYCKSLAMIAVGQVLSAMPWGC FQGLT VTYASEVCPLALRYMTSYSNICWLFQ
 IFASGIMKNSQENLGNSDLGYKL PFALQWIWPAPLMIGIFFAPESPWWLVRKDRVAEARKSLSR
 ILSGKGAEKDIQIDLT LKQIELTIEKERLLASKSGSFFDCFKGVNRRTRLACLTWVAQNTSGA
 25 CLLGYSAYFFERAGMATDKAFTFSVIQYCLGLAGTLC SWVISGRVGRWTILTYGLAFQMVCLFI
 IGGMGFGSGSGASNGAGGLLLALSFFYNAGIGAVVY CIVTEIPSAELRTKTIVLARICYNIMAV
 INAILTPYMLNVSDWNWGAKTGLYWGGFTAVTLAWVIIDL PETSGRTFSEINELFNQGV PARKF
 ASTVVD PFGKGKTQHDSLDDESISQSSSIKQRELNAADKC

SEQ ID NO:21

30 Amino acid sequence of IMA1p encoded by long IMA1 allele from Hybrid yeast 4
 MTISSAHPETEPKWWKEATFYQIYPASF KDSNDDGWGDMKGISSKLEYIKELGVDAIWISPFYD
 SPQDDMGYDIANYEKVWPTYGTNEDCFALIEKTHKLG MKFITDLVINHCSSEHEWFKESRSSKT
 NPKRDWFFWRPPKGYDAEGKPIPPNNWKS YFGGSAWIFDEKTQEFYLR LFCSTQPD LNWENEDC
 RKAIYESAVGYWLDHGV DGFRI DVGSLY SKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQ
 35 EMNQFIRNRVKDGREIMTVGEMQHASEDETKRLYTSASRHELSELFNFSHTDVGTSPLFRYNLVP
 FELKDWKIALAELFRFINGTDCWSTIYLENHDQPR SITRFGDDSPKNRVISGKLLSVLLSALTG
 TLYVYQGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEAIALISR DHARTP
 MQWSREEPNAGFSGPSAKPWFYLNDSFREGINVEDEIKDPNSVLNFWKEALKFRKAHKDITVYG
 YDFFEFIDL DNKKLFSFTKKYNNKTLFAALNFSSDATDFKIPND DSSF KLEFGNYPKKEVDASSR
 40 TLKPWEGRIYISE.

SEQ ID NO:22

45 Amino acid sequence of IMA1p encoded by long IMA1 allele from Hybrid yeast 4
 MTISSAHPETEPKWWKEATFYQIYPASF KDSNDDGWGDMKGISSKLEYIKELGVDAIWISPFYD
 SPQDDMGYDIANYEKVWPTYGTNEDCFALIEKTHKLG MKFITDLVINHCSSEHEWFKESRSSKT
 NPKRDWFFWRPPKGYDAEGKPIPPNNWKS YFGGSAWIFDEKTQEFYLR LFCSTQPD LNWENEDC
 RKAIYESAVGYWLDHGV DGFRI DVGSLY SKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQ
 50 EMNQFIRNRVKDGREIMTVGEMQHASEDETKRLYTSASRHELSELFNFSHTDVGTSPLFRYNLVP
 FELKDWKIALAELFRYINGTDCWSTIYLENHDQPR SITRFGDDSPKNRVISGKLLSVLLSALTG
 TLYVYQGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEAIALISR DHARTP
 MQWSREEPNAGFSGPSAKPWFYLNDSFREGINVEDEIKDPNSVLNFWKEALKFRKAHKDITVYG

YDFEFIDLDNKKLFSFTKKYNNKTLFAALNFSSDATDFKIPNDSSFKLEFGNYPKKEVDASSR
TLKPWEGRIYISE.

SEQ ID NO:23

5 Amino acid sequence of IMA1p encoded by long IMA1 allele from
Hybrid yeast 7

MTISSAHPETEPKWWKEATFYQIYPASFKDSNDDGWGDMKGISSKLEYIKELGVDAIWISPFYD
SPQDDMGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSEHEWFKESRSSKT
NPKRDWFFWRPPKGYDAEGKPIPPNNWKSYPFGGSAWIFDEKTQEFYLRLLFCSTQPDNLWENEDC
10 RKAIYESAVGYWLDHGVGDFRIDVGSLSYKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQ
EMNQFIRNRVKDGREIMTVGEMQHASDETKKLYTSASRHELSELFNFSTHDVGTSPFLFRYNLVP
FELKDWKIALAELFRFINGTDCWSTIYLENHDQPRISITRFGDDSPKNRVISGKLLSVLLSALTG
TLYVYQGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEAIALISRDHARTP
MQWSREEPNAGFSGPSAKPWLYLNDSFREGINVEDEIKDPNSVLNFWKEALKFRKAHKDITVYG
15 YDFEFIDLDNKKLFSFTKKYNNKTLFAALNFSSDATDFKIPNDSSFKLEFGNYPKKEVDASSR
TLKPWEGRIYISE.

SEQ ID NO:24

20 Amino acid sequence of IMA1p encoded by long IMA1 allele from
Hybrid yeast 7

MTISSAHPETEPKWWKEATFYQIYPASFKDSNDDGWGDMKGISSKLEYIKELGVDAIWISPFYD
SPQDDMGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSEHEWFKESRSSKT
NPKRDWFFWRPPKGYDAEGKPIPPNNWKSYPFGGSAWTFDEKTQEFYLRLLFCSTQPDNLWENEDC
RKAIYESAVGYWLDHGVGDFRIDVGSLSYKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQ
25 EMNQFIRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSTHDVGTSPFLFRYNLVP
FELKDWKIALAELFRYINGTDCWSTIYLENHDQPRISITRFGDDSPKNRVISGKLLSVLLSALTG
TLYVYQGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEAIALISRDHARTP
MQWSREEPNAGFSGPSAKPWLYLNDSFREGINVEDEIKDPNSVLNFWKEALKFRKAHKDITVYG
YDFEFIDLDNKKLFSFTKKYNNKTLFAALNFSSDATDFKIPNDSSFKLEFGNYPKKEVDASSR
30 TLKPWEGRIYISE.

SEQ ID NO:25

35 Amino acid sequence of IMA1p encoded by long IMA1 allele from
Hybrid yeast 7

MTISSAHPETEPKWWKEATFYQIYPASFKDSNDDGWGDMKGISSKLEYIKELGVDAIWISPFYD
SPQDDMGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSSEHEWFKESRSSKT
NPKRDWFFWRPPKGYDAEGKPIPPNNWKSYPFGGSAWIFDEKTQEFYLRLLFCSTQPDNLWENEDC
RKAIYESAVGYWLDHGVGDFRIDVGSLSYKVVGLPDAPVVDKNSTWQSSDPYTLNGPRIHEFHQ
EMNQFIRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSTHDVGTSPFLFRYNLVP
40 FELKDWKIALAELFRYINGTDCWSTIYLENHDQPRISITRFGDDSPKNRVISGKLLSVLLSALTG
TLYVYQGQELGQINFKNWPVEKYEDVEIRNNYNAIKEEHGENSEEMKKFLEAIALISRDHARTP
MQWSREEPNAGFSGPSAKPWLYLNDSFREGINVEDEIKDPNSVLNFWKEALKFRKAHKDITVYG
YDFEFIDLDNKKLFSFTKKYNNKTLFAALNFSSDATDFKIPNDSSFKLEFGNYPKKEVDASSR
TLKPWEGRIYISE.

45

SEQ ID NO:26

Amino acid sequence of truncated AGT1 encoded by Sc allele from
Hybrid yeast 4

MKNIISLVSKKKAASKNEDKNISESSRDIVNQQEVFNTEENFEEGKKDSAFELDHLEFTTNSAQL
50 GDSDEDNENMINEMNATDEANEANSEKSMTLKQALLKYPKAALWSILVSTTLVMEGYDTALLN
ALYALPVFQRKFGTLNGEGSYEITSQWQIGLNMVCVQCGEMIGLQITTYMVEFMGNRYTMITALG
LLTAYIFILYYCKSLAMIAVGQVLSAMPWGCQGLTVTYASEVCPLALRYMTSYSNICWLFQGQ

IFASGIMKNSQENLGNSDLGYKLPFALQWIWPAPLMIGIFFAPESPWWLVRKDRVAEARKSLSR
ILSGKGAEKDIQVDLTTLKQIELTIEKERLLASKSGSFFDCFKGVNGRRTRLACLAWVAQNTSGA
CLLGYSTYFF.

5 SEQ ID NO:27

Amino acid sequence of AGT1 encoded by non-Sc allele from Hybrid yeast 4

MKNILSLVGRKENTPEDVTANLADTSSTTVMQAKDLVIEDFEERKKND AFELNHLELT TNATQL
SDSDEDKENVIRVAEATDDANEANNEEKSM TLRQALRKYPKAALWSILVSTTLVMEGYDTALLS
10 ALYALPVFQRKFGTMNAEGSYEITSQWQIGLNM CVLCGEMIGLQITTYMVEFMGNRYTMITALS
LLTAYIFILYYCKSLAMIAVGQILSAMPWGC FQSLAVTYASEVCPLALRYMTSYSNICWLFQ
IFASGIMKNSQENLGNSDLGYKLPFALQWIWPAPLIIGIFFAPESPWWLVRKNKIVEAKKSLNR
ILSGTVTEKEIQVDITLQIEMTIEKERLRASKSGSFFSCFKGVDGRRTRLACLTWVAQNSSGA
VLLGYSTYFFERAGMATDKAFTFSLIQYCLGLAGTLGSWVISGRVGRWTILTYGLSFQMVCLFI
15 IGGMGFASGSSASNAAGGLLLALSFFYNAGIGAVVYCIVAEIPSAELRKTIVLARICYNLMAV
FNAILT PYMLNVSDWNWGAKTGLYWGDF TALTLAWVIIDL PETTGRTFSEINELFSQGV PARKF
ASTVVD PFGKRGLQNR PQVDNIIDRFSSASQQAL.

SEQ ID NO:28

20 Amino acid sequence of AGT1 encoded by non-Sc allele from Hybrid yeast 4

MKNILSLVGRKENTPEDVTANLADTSSTTVMQAKDLVIEDFEERKKND AFELNHLELT TNATQL
SDSDEDKENVIRVAEATDDANEANNEEKSM TLRQALRKYPKAALWSILVSTTLVMEGYDTALLS
25 ALYALPVFQRKFGTMNAEGSYEITSQWQIGLNM CVLCGEMIGLQITTYMVEFMGNRYTMITALS
LLTAYIFILYYCKSLAMIAVGQILSAMPWGC FQSLAVTYASEVCPLALRYMTSYSNICWLFQ
IFASGIMKNSQENLGNSDLGYKLPFALQWIWPAPLIIGIFFAPESPWWLVRKNKIVEAKKSLNR
ILSGTVTEKEIQVDITLQIEMTIEKERLRASKSGSFFSCFKGVDGRRTRLACLTWVAQNSSGA
VLLGYSTYFFERAGMATDKAFTFSLIQYCLGLAGTLGSWVISGRVGRWTILTYGLSFQMVCLFI
IGGMGFASGSSASNAAGGLLLALSFFYNAGIGAVVYCIVAEIPSAELRKTIVLARICYNLMAV
30 FNAILT PYMLNVSDWNWGAKTGLYWGDF TALTLAWVIIDL PETTGRTFSEINELFSQGV PARKF
ASTVVD PFGKRGLQNR PQVDNIIDRFSSASQQAL.

SEQ ID NO:29

35 Amino acid sequence of truncated AGT1 encoded by Sc-allele from Hybrid yeast 7

MKNIISLVSKKKAASKNEDKNISESSRDIVNQQEVFN TENFEEGKKDSAFELDHLEFTTNSAQL
GDSDEDNENMINEMNATDEANEANSEEEKSM TLRQALLKYPKAALWSILVSTTLVMEGYDTALLN
ALYALPVFQRKFGTLNGEGSYEITSQWQIGLNM CVQCGEMIGLQITTYMVEFMGNRYTMITALG
LLTAYIFILYYCKSLAMIAVGQVLSAMPWGC FQGLTVTYASEVCPLALRYMTSYSNICWLFQ
40 IFASGIMKNSQENLGNSDLGYKLPFALQWIWPAPLMIGIFFAPESPWWLVRKDRVAEARKSLSR
ILSGKGAEKDIQVDLTTLKQIELTIEKERLLASKSGSFFDCFKGVNGRRTRLACLAWVAQNTSGA
CLLGYSTYFF.

SEQ ID NO:30

45 Amino acid sequence of AGT1 encoded by Sc allele from Hybrid yeast 7

MKNIISLVSKKKAASKNEDKNISESSRDIVNQQEVFN TENFEEGRKDSAFELDHLEFTINSAQL
GDSDEDNENVINETNTTDDANEANSEEEKSM TLRQALLIYPKAALWSILVSTTLVMEGYDTALLN
ALYALPVFQRKFGTLNGEGSYEITSQWQIGLNM CVQCGEII GLQITPYMVEFMGNRYTMITALG
50 LLTAYVFILYYCKSLAMIAVGQVLSAMPWGC FQGLTVTYASEVCPLALRYMTSYSNICWLFQ
IFASGIMKNSQENLGNSDLGYKLPFALQWIWPAPLMIGIFFAPESPWWLVRKDRVAEARKSLSR
ILSGKGAEKDIQIDLTTLKQIELTIEKERLLASKSGSFFDCFKGVNGRRTRLACLTWVAQNTSGA

CLLGYSTYFFERAGMATDKAFTFSVIQYCLGLAGTLCSWVISGRVGRWTILTYGLAFQMVCLFI
IGGMFGSGSGASNGAGLLLLALSFFYNAGIGAVVYCIVTEIPSAELRKTIVLARICYNIMAV
INAILTPYMLNVSDWNWGAKTGLYWGGFTAVTLAWVIIDLPESTGRTFSEINELFNQGVPARKF
ASTVVDPFQKGTQHDSDLDESISQSSSIKQRELNAADKC.

5

SEQ ID NO:31

Amino acid sequence of AGT1 encoded by non-Sc allele from Hybrid yeast 7

10 MKNILSLVGRKENTPEDVTANLADTSSTTVMQAKDLVIEDFEERKKNDAFELNHLELTNATQL
SDSDEDKENVIRVAEATDDANEANNEEKSMTRLQALRKYPKAALWSILVSTTLVMEGYDTALLS
ALYALPVFQKFGTMNAEGSYEITSQWQIGLNMVLCGEMIGLQITTYMVEFMGNRYTMITALS
LLTAYIFILYYCKSLAMIAVGQILSAMPWGCQSLAVTYASEVCPLALRYMTSYSNICWLFGQ
IFASGIMKNSQENLGNSDLGYKLFPALQWIWPAPLIIGIFFAPESPWWLVRKNKIVEAKKSLNR
15 ILSGTVTEKEIQVDITLKQIEMTIEKERLRASKSGSFFSCFKGVDGRRTLACLTWVAQNSSGA
VLLGYSTYFFERAGMATDKAFTFSLIQYCLGLAGTLGWSVISGRVGRWTILTYGLSFQMVCLFI
IGGMGFASGSSASNAAGLLLLALSFFYNAGIGAVVYCIVAEIPSAELRKTIVLARICYNLMAV
FNAILTPYMLNVSDWNWGAKTGLYWGGFTALTALWVIIDLPESTGRTFSEINELFSQGVPARKF
ASTVVDPFQKRGQLQNRPQVDNIIDRFSSASQQAL.

20

SEQ ID NO:32

Amino acid sequence of AGT1 encoded by non-Sc allele from Hybrid yeast 7

25 MKNILSLVGRKENTPEDVTANLADTSSTTVMQAKDLVIEDFEERKKNDAFELNHLELTNATQL
SDSDEDKENVIRVAEATDDANEANNEEKSMTRLQALRKYPKAALWSILVSTTLVMEGYDTALLS
ALYALPVFQKFGTMNAEGSYEITSQWQIGLNMVLCGEMIGLQITTYMVEFMGNRYTMITALS
LLTAYIFILYYCKSLAMIAVGQILSAMPWGCQSLAVTYASEVCPLALRYMTSYSNICWLFGQ
IFASGIMKNSQENLGNSDLGYKLFPALQWIWPAPLIIGIFFAPESPWWLVRKNKIVEAKKSLNR
ILSGTVTEKEIQVDITLKQIEMTIEKERLRASKSGSFFSCFKGVDGRRTLACLTWVAQNSSGA
30 VLLGYSTYFFERAGMATDKAFTFSLIQYCLGLAGTLGWSVISGRVGRWTILTYGLSFQMVCLFI
IGGMGFASGSSASNAAGLLLLALSFFYNAGIGAVVYCIVAEIPSAELRKTIVLARICYNLMAV
FNAILTPYMLNVSDWNWGAKTGLYWGGFTALTALWVIIDLPESTGRTFSEINELFSQGVPARKF
ASTVVDPFQKRGQLQNRPQVDNIIDRFSSASQQAL.

35 SEQ ID NO:33

Amino acid sequence of SHORT_IMA1 from Hybrid yeast_7_

MGYDIANYEKVWPTYGTNEDCFALIEKTHKLGMKFITDLVINHCSEHEWFKESRSSKTNPKRD
WFFWRPPKGYDAEGKPIPPNNWKSIFGGSAAWTFDEKTQEFYLRFLFCSTQPDNLWENEDCRKAIY
ESAVGYWLDHGVDFRIDVGSLSKVVLDPAPVVDKNSTWQSSDPYTLNGPRIHEFHQEMNQF
IRNRVKDGREIMTVGEMQHASDETKRLYTSASRHELSELFNFSHTDVGTSPLFRYNLVPFELKD
40 WKIALAELFRFINGTDCWSTIYLENHDQPRISITRFGDDSPKNRVISGKLLSVLLSALTGTLYVY
QGQELGQINFKNWSVEKYEDVEIRNNYRLIKEECGENSEEMKKFLEGIALVSRDHARTPMPWTP
NEPNAGFSGPNTKPFYLNESFRQGINVEEEQKNSDSVLAFWKKALEFRKNHKDIAVYGFDFKF
IDLDNKKLFSFTKRYNNKTLFAALNFSSDATDFKIPNDGSSFKLEFGNYPKNEVDASSRTLKPW
EGRIHINE.

45

SEQ ID NO:34

Amino acid sequence of IMA5 from_Hybrid yeast_1

50 MTIIHNPKWWKEATVYQIYPASNKDSNNDGWGDLGITSKLDYVKELGVDAIWVCLFYDSPQED
MGYDIANYEKVWPRYGTNEDCFQMIEEAHKGRIKVIIVDLVINHCSEHEWFKESKSSKTNPKRD
WFFWRPPKGFDEKGNPIPPNNWRSFFGGSAAWRYDEKTGEFFLHVFPAGQPDFNWEENECCRKAIY
DSSVGYWLRHNVDFRIDVGSMSYKVEGLDPAPITDPTVPYQKGTEFFINGPRIHEYHKEMRKY

5 MLSQIPEGKEIMTVGEVGVGNEEDFRDYTSAKEGELNMMFNFKHTSVGESPECKYELIPFTLKD
FKLALAESFLFIENTDCWSTIYLENHDQPRSVSRFGSDSPKWRAISSKMLATLIISLTGTVFIY
QGQELGMSNFKNRRIEQIKVEGTGTYYAAIKRDYGEDSEKMKKFFEALALISRDHGRTPFPWSA
DEPSAGFSKDAKPWIDMNESFRDGINAEAEKDKNSVFFFWKKALQVRKEHKDILVYGHNFQFI
DLDNDKLFMFTKDTDNKKMFAVFNFSNDNTDFSVPDNEASYTMFFGNYANSNGDSRTLQPWEGR
LYLLK

SEQ ID NO:35

Amino acid sequence of the Hybrid Sc_IMA5_Hybrid_7 allele

10 MTIIHNPKWWKEATVYQIYPASNKDSNNDGWGDLAGITSKLDYVKELGVDIAIWVCLFYDSPQED
MGYDIANYEKVWPRYGTNEDCFQMIEEAHKGRIKVIIVDLVINHCSEEHEWFKESKSSKTNPKRD
WFFWRPPKGFDEKGNPIPPNNWRSFFGGSARWYDEKTGEFFLHVAFAPGQPDFNWENЕКCRKAIY
DSSVGYYLWRHNVDFRIDVGSMSKVEGLPDAPITDPTVPYQKGTEFFINGPRIHEYHKEMRKY
15 MLSQIPEGKEIMTVGEVGVGNEEDFRDYTSAKEGELNMMFNFKHTSVGESPECKYELIPFTLKD
FKLALAESFLFIENTDCWSTIYLENHDQPRSVSRFGSDSPKWRAISSKMLATLIISLTGTVFIY
QGQELGMSNFKNRRIEQIKVEGTGTYYAAIKRDYGEDSEKMKKFFEALALISRDHGRTPFPWSA
DEPSAGFSKDAKPWIDMNESFRDGINAEAEKDKNSVFFFWKKALQVRKEHKDILVYGHNFQFI
DLDNDKLFMFTKDTDNKKMFAVFNFSNDNTDFSVPDNEASYTMFFGNYANSNGDSRTLQPWEGR
20 LYLLK.

SEQ ID NO:36

Amino acid sequence of the nonSc_IMA5_Hybrid_7 allele; first 6
amino acids are missing from the genomic sequence;

25 PKWWKEATIIYQIYPASFKDSNNDGWGDLAGITSKLDYIKELGVDIAIWVCPFYDSPQEDMGYDIA
NYEKVWPRYGTSED CFQMIEESHKRGRIKVIIVDLVINHCSEEHEWFKESRSSKTNAKRDWFFWKP
PKGYEIDGTPIPPNNWRSFFGGSARWYDENTEEFFLHVAFAPGQPDFNWENЕКCRQAIYDSSVGF
WLRHNVDFRIDVGSMSKVEGLPDASITDPTVPYQDGTDFVNGPRIHEYHKEMRQYMYTQIP
30 EGKEIMTVGEVGIGNEKDFKDYTSKEEEFNMMFNFKHTSVGESPEFKYELIPFTLKDFKLALA
ESFLFIEGTDCWSTIYLENHDQPRSVSRFGSDSPEWREISSKMLATLIISLTGTVFIYQGQELG
MPNFKNRKIEQIKVEGTGTYYAAIKRDYGEDSEKMKKFYEALALISRDHGRTPFPWSGEKPYAG
FSKNAKPWIDINESFVEGINAEAELENDENSVFFFWKRALQVRKEHKDILVYGDNFQFYDLDNEK
LFMFTKDSGDKMFAVFNFCSDSTEFVDPDNKASYDMFFGNYANSNGKSYTLKPWEGRLYYSN.

35

SEQ ID NO:37

Amino acid sequence of PTR2 encoded by Sc allele of_Hybrid_yeast
7 (incomplete sequence)

40 NSTERYNLSPSPEDDFEAPTEEEMQTLRHVGGKIPMRCWLIAIVELSERFSYYGLSAPFQNYM
EYGPNDSPKGVLSLNSQGATGLSYFFQFWCYVTPVFGGYVADTFWGKYNTICCGTAIYIAGIFI
LFITSIPSVGNRDSAIGGFIAAIIILIGIATGMIKANLSVLIADQLPKRKPSIKVLKSGERVIVD
SNITLQNVFMFFYFMINVGSLSLMAATTELEYHKGFWAAYLLPFCFFWIAVVTLIFGKKQYIQRP
IGDKVIAKSFKVCWILTKNKFDFNAAKPSVHPEKNYPWNDKFVDEIKRALAACKVFIFYPIYWT
45 QYGTMISSFITQASMMELHGIPNDFLQAFDSIALIIFIPIFEKFVYPFIRRYTLPKPITKIFFG
FMFGSFAMTAAVLQSFVYKAGPWYNEPLGHNTPNHVHVCWQIPAYVLISFSEIFASITGLEYA
YSKAPASMKSFIMSIFLLTNAFGSAIGCALSPVTVDPKFTWLFTGLAVACFISGCLFWLCFRKY
NDTEEEMNAMDYEEDEFDLNPISAPKANDIEILEPMESLRSTTKY.

50

SEQ ID NO:38

Protein sequence of PTR2 encoded by nonSc allele of Hybrid
yeast_7

MLNHL SQSDDIQDEKQGD FVIEEEKNQTVTLKDSYVSDDAANSTEHYNLSPSLEEDEF EAPT
 DEELRSLRHVGGKIPMRCWLI AIVELSERFSYYGLSAPFQNYMEYGP KDT PKGVLSLNSQGATG
 LSYFFQFWCYVTPVFGGYVADTFW GKYNTICCGTAIYIAGIFILLITSIPSVGNRDSALGGFIA
 SIILIGIATGMIKANLSVLIADQLPKRKPSIKVLKSGERVIVDSNITLQNVFMFFYFMINVGSL
 5 SLMATTELEYHKGFWAAYLLPFCFFWVAVVTLVFGKKQYIQRPIGDKVIAKSFRVCWILT KKNKF
 DFNAAKPSVHPEKEYPWNDKFVDEIKRALAACKVFVFYPIYWTQYGTMISSFITQAGMMELHGI
 PNDFLQAFDSIALIIFIPIFEKFIYPFIRRYTPFKPITKIFFGFMFGSLAMTWA AVLQS FVYKA
 GPWYSAPLGHNTPNHVVHCWQIPAYVLISFSEIFASITGLE YAYS KAPASMKS FIMSIFLLTNA
 FGSAIGCALSPVTVDPKFTWLF TGLAVACFISGCLFWFCFRKYNDTEEEMNAMDYEE EDEFDLN
 10 PISQPKGNDIEILEPMGSLKSTTKY.

SEQ ID NO:39

Amino acid sequence of DAL5_encoded by Sc allele of Hybrid
 yeast_7

15 MSADASTNSNASLDEKNLNITSEAEIKNEDVTAEPVLSTVLSPNGKIVYISDKVDEAMKLAEEA
 KEIEVTPEEDRKL RWKIDYCMFPLMCILYAVQFMDKISTSSAAVMGLRTDLKM HGDQYSWVTSA
 FYFGYLFMNLGVPVQFIFQRTSHMSKMLAVFIVIWGMLLALHAAPT VKYPSFIVLRVLLGCAESV
 VTPCF TIITAQYWKTEEQFTRVSIWFGMNLG SILINAIAYGVYIHQDSYAIKGWRTLFVITGV
 ITIFIGILIFLWIPDDPSKARFLSKREKLMVVQRIRSNQQGFGNHEIKKYQIIEALKDVRTWLY
 20 FLFTVSSNIPNGGISSFMSILLNSDFGYSSKETLLMGLPTGAVELVGCPLFGILAVYAANKKIP
 FWKYKLSWAIFAAVLALIASCMLGFATNSKKARLAGAYLWYISPVSFICVLSNISANSSGYSKK
 WTVSSINLVAYAAANLAGPQTFIAKQAPKYHGAKVAMVVCYAVMIVLLSILLIVNLRENKR RDK
 IAAERGFPEETENLEFSDLTDFENPNFRYTL.

25 SEQ ID NO:40

Amino acid sequence of DAL5 encoded by nonSC allele of Hybrid
 yeast_7

MSGGASTNSNASIDEKNLNITSEAEIKNEDVYAEPVLSTVLSPNGKV VYISDKVDEAMKLAD E A
 KEIEVTPEEDRKL RWKIDYCMFPLMCILYAVQFMDKISTSSAAVMGLRTDLKM HGDQYSWVTSA
 30 FYFGYLFMNLGVPVQLIFQKSKHMSKMLAIFIIVWGLLLALHAVPSVKYSSFIALRVLLGCAESV
 VTPCF TIITAQYWKTEEQFTRISIWFGMNLG SILINAIAYGVYIHQESYAIKGWRALFVITGV
 ITIFVGALIFLWIPDDPSKARFLSKREKLMVVQRIRSNQQGFGNHEIKKYQIVEALKDVRTWLY
 FLFTVSSNIPNGGISSFMSILLNSDFGYLSKDTLLMGLPTGAVELVGCPLFGILAVYAANKKIP
 FWKYKLAWAIFAAVLALIASCMLGFATSSKKARLAGAYLWYISPVSFICVLSNISANSSGYSKK
 35 WTVSSINLAAYAAANLAGPQTFIAKQAPKYHGAKVAMVVCYAVMIVLLSALLLINMRENKR RDK
 IAAERGYPEETANLEFSDLTDFENPNFRYTL.

SEQ ID NO:41

Amino acid sequence of UBR1_encoded by Sc allele of Hybrid_yeast
 40 7 (incomplete sequence)

MSVADDDLGLSLQGHIRRTLRSIHNLPHYFRYTRGPTERADMSRALKEFIYRYLYFVISNSGENLP
 TLFNAHPKQKLSNPELTVFPDSLEDAVDIDKITSQQTIPFYKIDESRIGDVHKHTGRNCGRKFK
 IGEPLYRCHECGCDDTCVLCIHC FNPKDHNHVVCTDICTEFTSGICDCGDEEAWNSPLHCKAE
 EQENDISED PATNADIKEEDVWNSVNIALVELVLAEVFDYFIDVFNQNI EPLPTIQKDITIKL
 45 REMTQQGKMYERAQFLNDLKYENDYMF DGT TTA KTSPSNSPEASPSLAKIDPENYTVIIYNDEY
 HNYSQATTALRQGVDPNVHIDLLTSRIDGEGRAMLKCSQDSSSVLG GFFAVQTNGLSATLT SWS
 EYLHQETCKYIILWITHC LNIPNSSFQTTFRNMMGKTLCSEYLNATECRDMTPVVEKYFSNKFD
 KNDPYRYIDL SILADGNQIPLGHHKILPESSTHSL SPLINDVETPTSR TYSNTRLQHILYFDNR
 YWKRLRKDIQNVIIPTLASSNLYKPIFCQQVVEIFNHITRSVAYMDREPQLTAIRECVVQLFTC
 50 PTNAKNIFENQSFLDIVWSIIDIFKEFCKVEGGVLIWQRVQKSNLT KSYSISFKQGLYTVETLL
 SKVHDPNIPLRPKEIISLLTLCKLFNGAWKIKRKEGEHVLHEDQNFI SYLEYTTTSIYSIIQTAE

KVSEKSKDSIDSKLFLNAIRIISFSLGNRSPTYKLIYDSHEVIKFSVSHERVAFMNPLQTMLSF
LIEKVSLKDAYEALEDSCDFLKI SDFSLRSVVLCSQIDVGFWVRNGMSVLHQASYKNNP

SEQ ID NO:42

5 Amino acid sequence of UBR1 encoded by nonSc allele of _Hybrid yeast_7

MSFTDNGLGSLKAHIRRTLRSIHNLPHYFRFTRGPTERADMSRALKEFIYRYLYFIISNDGENLS
TLFTAHPKQKSSNQELAVFPESLEDALDVKITSQGTFFPYKIDESKIGDVHKHTGRNCGRKFK
10 IGEPLYRCHECGCDDTCVLCIHCFNPKDHINHHVCTDICSEFTSGICDCGDEEAWNSSLHCKAE
EQGNDTSEDPSNFDSTKQKDVWNDPECIALVELVLSEVFDYFIDVFNQNI EPLPTIQKDITIKL
REMTQQGKMYERAQFLNDLKYENDYMFDTTAKTSPSNSPEASPSLAKIDPENYTVIIYNDEY
HNYSQATTALRQGVDPNVHIDLLTSRIDGEGRAMLKCSQDLSSVLGGFFAVQTNGLSATLTWS
EYLHQEACKYIILWITHCLNIPNPSFQITFRNMMGKSLCSEYLNATESRDMPVVEKYFSTKFD
15 KDDPYRYIDLSVLAEGNQIPLGHHKVLPESSHSLSTLINDVENLTSKEYSNTRLQHILYFDNR
YWKRLRKDIQNVIIPTLASSTLYKPIFCQQVVEIFNHITRSVAYMDREPQLTAIRECVVQLFTC
PTNTRNIFENQSFLDILWSIIDIFKEFCKVEAGVLIWQVRVQKSNLTKSYSLSFKQGLYTVETLL
SKVNDPNITIRPKVFISLLTLGKLFNGAWKIKRKEGEHVLHEDQNFISYLEYTTSIYSIIQTAE
KVLEKSHDSL DLNLVLNAIRIVSSFLGNRSPTYKLIYDSHEIIKFSVSHERVAFMNPIQTMLSF
LIEKVSLKDAYESENCPDFLKIADFSRLSVVLCSQIDVGFWVRNGMSVLHQASYKNNPELGS
20 YSRDIHLNQLAI IWERDDLPRVIYNILDRWELLDWFMGEAEYQHTVYEDKISFMIQQFIAFIYQ
ILTERQYFKTFSLLRDRMDMIKNSIMYNLYMKPLSYSKLLKSVDPDYLTDDETFDEALEEVS
FVEPKGLADNGVFKLKAALYAKIDPLKLLNLENEFESSATIIKTHLAKNKDEVSKVVLIPQVST
KLLDKGAMNLGEFTRNTVFAKVIYKLLQVCLDMEDSTFLNELLHLVHGIFKDELINGKDSIPE
AYLAKPICNLLLSIANAKSDIFSESIVRKADYLLEKMIMKKPDEIFESLIASFGNQYIDNYKDK
25 KLSQGVNLQETEKERKRRMAKKHQARLLAKFNNQSKFMKEHESEFDEQDNDVMDGEKVYSE
DFTCALCQDSSSTDFVVPAYHDHTPIFRPGNI FNPREFMAKWDGFYNDQKQAYIDDEVLESL
KENGTRGSRKV FVSCNHHIHHNCFKRYVQKKRFSSNAFICPLCQTFSNCTLPICPTSRANTGLS
LDMFLKSELSDLILSRLFKPFTEDNYRTINSIFSLMVSQCQGFQKVVVRKHVNFTHKDVSLVLSV
HWANTISM LEVASRLEKPHNISFFRSREQYKTLKNILICIMLFTFVIGKPSMEFEPYPVESDI
30 ICNQNLQFYIVRKSLFSPASLRETITEALTVECKQFLDDFVQGLSDAEQVDKLYTEAKKLGDV
YNVDESILITLMSITVVKTEGLESRSIYDLAYTSLKSLPTIRRLVMVKVLHELKVDSENET
MVIDGFDVEEELEFEGLPGFVDKALKLITDKESFVDLFKTKQAIVPSPHYLERIPYEYCGIVKL
IDLSKFLNTYVTQSKEIKLREERSQHMKNADNRLDFKICLTCGVKVHLRADRHMTKHLNKNCF
KSFGAFLMPNSSEVCLHLTQPPSNI FVSAPYLNSHGEVGRNAMRRGDLTTLNLKRYEHLNRLWI
35 NNEIPGYISRVMGDEFRTILSNGFLFAFNREPRPRVPPTDEDDDEMEEGEEGFFTEENDMD
VDDETGQAANLFGVGAEGIGDGGVRNFFQFFENFRNTLQPGNDDDEDAPQNPPPIQLGFPQFD
GATIIIRNTNQRLDEDDSENDDSDEREIW.

SEQ ID NO:43

40 Amino acid sequence of PTR2 encoded by Sc allele of Hybrid yeast 1

MLNHPSQGSDDAQDEKQGDFFVIEEEKTQAVMLKDSYVSDDVANSTERYNLSPSPEDEDFEAPT
EEEMQTLRHVGGKIPMRCWLI AIVELSERFSYYGLSAPFQNYMEYGPNDSPKGVLSLNSQGATG
LSYFFQFWCYVTPVFGGYVADTFWGKYNTICCGTAIYIAGIFILFITSIPSVGNRDSAIGGFIA
45 AIIILGIATGMIKANLSVLIADQLPKRKPSIKVLKSGERVIVDSNITLQNVFMFFYFMINVGSL
SLMATTELEYHKGFWAAYLLPFCFFWIAVVTLIFGKKQYIQRPIGDKVIAKSFKVCWILTNNKF
DFNAAKPSVHPEKNYPWNDFVDEIKRALAACKVFI FYPIYWTQYGTMISSFITQASMELHGI
PNDFLQAFDSIALIIFIPIFEKFVYPFIRRYTLPKIPITKIFVGFMFSGFAMTAAVLQS FVYKA
GPWYNEPLGHNTPNHVVHCWQIPAYVLISFSEIFASITGLEAYASKAPASMKSFIMSIFLLTNA
50 FGSAIGCALSPVTVDPKFTWLF TGLAVACFISGCLFWLCFRKYNDTEEMNAMDYEEENEFDLN
PISAPKANDIEILEPMDSLRSTTKY.

SEQ ID NO:44

Amino acid sequence of PTR2 encoded by nonSc allele of Hybrid yeast 1

5 MLNHL SQGSDDIQDEKQGDFPVIEEEKNQTVTLKDSYVSDDAANSTEHYNLSPSLEEDEFEAPT
DEELRSLRHVGGKIPMRCWLI AIVELSERFSYYGLSAPFQNYMEYGPKDTPKGVLSLNSQGATG
LSYFFQFWCYVTPVFGGYVADTFWGKYNTICCGTAIYIAGIFILLITSIPSVGNRDSALGGFIA
SIILIGIATGMIKANLSVLIADQLPKRKPSIKVLKSGERVIVDSNITLQNVFMFFYFMINVGSL
SLMATTELEYHKGFWAAYLLPFCFFWVAVVTLVFGKKQYIQRPIGDKVIAKSFRCWILTCKNF
10 DENAAKPSVHPEKEYPWNDKFVDEIKRALAACKVFVFYPIYWTQYGTMISSFITQAGMMELHGI
PNDFLQAFDSIALIIFIPIFEKFIYPFIRRYTPFKPITKIFFGFMFGSLAMTWA AVLQS FVYKA
GPWYSAPLGHNTPNHVHVCWQIPAYVLISFSEIFASITGLEAYASKAPASMKSFIMSI FLLTNA
FGSAIGCALSPVTVPKFTWLF TGLAVACFISGCLFWFCFRKYNDTEEMNAMDYEEDEFDLN
PISQPKGNDIEILEPMGSLKSTTKY.

15 SEQ ID NO:45

Partial amino acid sequence of UBR1 encoded by Sc allele of Hybrid yeast 1

MSVADDDLGS LQGHIRRTLRSIHNLPYFRYTRGPTERADMSRALKEFIYRYLYFVISNSGENLP
20 TLFNAHPKQKLSNPELTVFPDSLEDAVDIDKITSQQTIPFYKIDESRIGDVHKHTGRNCGRKFK
IGEPLYRCHECGDDTCVLCIHC FNPKDHVNHVCTDICTEFTSGICDCGDEEAWNSPLHCKAE
EQENDISED PATNADIKEEDVWNDSVNIALVELVLAEVFDYFIDVFNQNI EPLPTIQKDITIKL
REMTQQGKMYERAQFLNDLKYENDYMFDTTAKTSPSNSPEASPSLAKIDPENYTVIIYNDEY
HNYSQATTALRQGVDPNVHIDLLTSRIDGEGRAMLKCSQDL.

25 Items

The invention may furthermore be defined by the following items:

1. A yeast cell having at least one of the following characteristics::
 - 30 II. capable of utilizing panose as sole carbon source;
 - III. capable of utilizing one or more dipeptides as sole nitrogen source.
2. The yeast cell according to item 1 having both of characteristics II. and III.
- 35 3. The yeast cell according to any one of the preceding items, wherein the yeast cell furthermore has characteristic:
 - I. Capable of utilizing isomaltose as sole carbon source.
- 40 4. The yeast cell according to any one of the preceding items, wherein said yeast cell is capable of utilizing isomaltose as sole carbon source when said isomaltose is

present at a concentration of in the range of 1 to 5 g/L, such as on the range of 1 to 3 g/L, for example 2 g/L.

5. The yeast cell according to any one of the preceding items, wherein said yeast cell is capable of utilizing panose as sole carbon source when said panose is present at a concentration of in the range of 1 to 5 g/L, such as on the range of 1 to 3 g/L, for example 2 g/L.
6. The yeast cell according to any one of the preceding items, wherein said yeast cell is capable of removing at least 45% of the panose present in wort.
7. The yeast cell according to any one of the preceding items, wherein said yeast cell is capable of removing at least 50% of the panose present in wort after incubation for 5 days at 16°C.
8. The yeast cell according to any one of the preceding items, wherein the yeast cell further has the characteristic:
 - III. capable of utilizing one or more dipeptides as sole nitrogen source.
9. The yeast cell according to any one of the preceding items, wherein one or more of said dipeptides are selected from the group consisting of Met-Tyr, Leu-Tyr, Val-Met, Phe-Tyr, Ile-Leu and Ile-Asn.
10. The yeast cell according to any one of items 1 to 8, wherein one or more of said dipeptides are dipeptides of the formula Ala-Xaa, wherein Xaa denotes any amino acid.
11. The yeast cell according to item 10, wherein Xaa is an amino acid selected from the group consisting of Glu, Gly, His and Thr.
12. The yeast cell according to any one of items 1 to 8, wherein one or more of said dipeptides are selected from the group consisting of Gly-Arg, Ile-Asn, Lys-Tyr, Met-Lys, Val-Ala, Val-Asn, Val-Gly, Val-Gln, Val-Met and Val-Ser.

13. The yeast cell according to any one of items 1 to 8, wherein one or more of said dipeptides are dipeptides of the formula Val-Xaa, wherein Xaa denotes any amino acid.
- 5 14. The yeast cell according to any one of the preceding items, wherein the yeast cell furthermore is capable of utilizing allantoin as sole nitrogen source.
15. The yeast cell according to any one of the preceding items, wherein the yeast cell further has the characteristic:
- 10 IV. capable of utilizing one or more tri-peptides as sole nitrogen source.
16. The yeast cell according to item 15, wherein one of said tri-peptides is Gly-Gly-Gly.
17. The yeast cell according to any one of the preceding items, wherein the yeast cell further has the characteristic:
- 15 V. capable of reducing the level of one or more amino acids to no more than 10% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells.
- 20 18. The yeast cell according to any one of the preceding items, wherein the yeast cell is capable of reducing the level of at least 12, such as at least 13, for example of at least 14 different amino acids to less than 10% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells.
- 25 19. The yeast cell according to any one of the preceding items, wherein the yeast cell is capable of reducing the total level of amino acids to less than 30%, such as less than 25% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells.
- 30 20. The yeast cell according to any one of the preceding items, wherein the yeast cell is capable of reducing the level of all of the amino acids Met, Val, Ile, Leu and Phe to less than 10%, preferably less than 5%, even more preferably to at the most 2% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells.

21. The yeast cell according to any one of the preceding items, wherein the yeast cell is capable of reducing the total level of the amino acids Met, Val, Ile, Leu and/or Phe to at the most 400 mg/L, such as at the most 100 mg/L, such as at the most 50 mg/L, for example to at the most 10 mg/L after incubation for 6 days under conditions allowing growth of said yeast cell.

22. The yeast cell according to any one of the preceding items, wherein the yeast cell further has the characteristic:

10 VI. capable of generating at least 4.7 promille ethanol per °Plato, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

23. The yeast cell according to any one of the preceding items, wherein the yeast cell further has the characteristic:

15 VII. capable of fermenting sugar with a real degree of fermentation of at least 68, such as at least 70, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

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24. The yeast cell according to any one of the preceding items, wherein the yeast cell has characteristic VII, wherein characteristic VII is that the yeast cell is capable for fermenting sugar with an RDF, which is at least 1 higher than the RDF of one of its parental strains.

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25. The yeast cell according to any one of the preceding items, wherein the yeast cell further has the characteristic:

VIII. capable of utilizing melibiose as the sole carbon source.

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26. The yeast cell according to any one of the preceding items, wherein the yeast cell further has the characteristic:

X. capable of sedimentation so that at the most 12 million, such as at the most 10 million cells/ml are in suspension when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated for 4 days.

27. The yeast cell according to item 26, wherein the number of cells in suspension per ml is at the most 80%, such as at the most 70%, for example at the most 60%, such as at the most 50%, for example at the most 40% of the starting number of cells per ml
5 after 5 days incubation under conditions allowing growth of said cells.

28. The yeast cell according to any one of the preceding items, wherein the yeast cell further has characteristic:

IX. capable of utilizing one or more disaccharides and/or trisaccharides in addition
10 to isomaltose, panose, and/or melibiose.

29. The yeast cell according to item 28, wherein the disaccharide is selected from the group consisting of kojibiose, nigerose, sucrose, turanose, leucrose, and palatinose.

15 30. The yeast cell according to item 28, wherein the disaccharide is kojibiose.

31. The yeast cell according to any one of the preceding items, wherein said yeast cell is capable of utilizing kojibiose as sole carbon source when said kojibiose is present at a concentration of in the range of 1 to 5 g/L, such as on the range of 1
20 to 3 g/L, for example 2 g/L.

32. The yeast cell according to item 28, wherein the disaccharide is maltulose.

33. The yeast cell according to any one of the preceding items, wherein said yeast cell is capable of utilizing maltulose as sole carbon source when said maltulose is present at a concentration of in the range of 1 to 5 g/L, such as on the range of 1
25 to 3 g/L, for example 2 g/L.

30 34. The yeast cell according to item 28, wherein the trisaccharide is selected from the group consisting of maltotriose and isomaltotriose.

35. The yeast cell according to any one of the preceding items, wherein the yeast cell is capable or utilizing maltotriose as the sole carbon source, when said

maltotriose is present at a concentration of in the range of 1 to 5 g/L, such as on the range of 1 to 3 g/L, for example 2 g/L.

- 5 36. The yeast cell according to any one of the preceding items, wherein the yeast cell furthermore has characteristic

XI: capable of fermenting wort with a time of primary fermentation of at the most 4 days, for example at the most 3 days.

- 10 37. The yeast cells according to item 36, wherein said time of primary fermentation is determined after pitching wort with a sugar content of in the range of 10° Plato to 20° Plato with in the range of 10 to 20 million viable cells/ml.

- 15 38. The yeast cell according to any one of the preceding items, wherein the yeast cell has genotype:
 I. comprising a gene encoding DAL5.

- 20 39. A yeast cell having genotype:
 I. comprising a gene encoding DAL5.

- 25 40. The yeast cell according to any one of items 38 to 39, wherein the genotype I is that said yeast cell comprises at least one allelic gene encoding DAL5, wherein the allelic gene encoding DAL5 encodes DAL5 selected from the group consisting of DAL5 of SEQ ID NO:6, DAL5 of SEQ ID NO:39, DAL5 of SEQ ID NO:40 and functional homologues thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity with any of the aforementioned.

- 30 41. The yeast cell according to any one of items 38 to 40, wherein the genotype I is that said yeast cell comprises at least one allelic gene encoding DAL5 of SEQ ID NO:6 or a functional homologue thereof sharing at least 80%, preferably at least

90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

- 5 42. The yeast cell according to any one of items 38 to 41, wherein the yeast cell, furthermore has characteristic III, for example characteristic III as defined in any one of items 8 to 14.
- 10 43. The yeast cell according to any one of items 38 to 42, wherein the yeast cell, furthermore has characteristics IV, for example characteristic IV as defined in any one of items 15 to 16.
- 15 44. The yeast cell according to any one of items 38 to 43, wherein the yeast cell, furthermore has characteristics V, for example characteristic V as defined in any one of items 17 to 21.
45. The yeast cell according to any one of the preceding items, wherein the yeast cell has genotype:
- II. comprising at least 2 allelic genes encoding PTR2.
- 20 46. A yeast cell having genotype:
- II. comprising at least 2 allelic genes encoding PTR2.
- 25 47. The yeast cell according to any one of items 45 to 46, wherein said PTR2 may be selected from the group consisting of PTR2 of SEQ ID NO:7, PTR2 of SEQ ID:8, PRT2 of SEQ ID NO:9 and functional homologues of each of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.
- 30 48. The yeast cell according to any one of items 45 to 47, wherein genotype II is said yeast cell comprising at least two allelic genes encoding PTR2 individually selected from the group consisting of genes encoding PTR2 of SEQ ID NO:7, PTR2 of SEQ ID:8, PRT2 of SEQ ID NO:9, PRT2 comprising SEQ ID NO:37, PRT2 of SEQ ID NO:38, PRT2 of SEQ ID NO:43, PRT2 of SEQ ID NO:44 and

functional homologues of each of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

- 5 49. The yeast cell according to any one of items 45 to 48, wherein genotype II is that the yeast cell comprises the following 3 genes:
- 10 1) a gene encoding PRT2 of SEQ ID NO:7 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith;
- 15 2) a gene encoding PRT2 of SEQ ID NO:8 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 20 3) a gene encoding PRT2 of SEQ ID NO:9 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.
- 25 50. The yeast cell according to any one of items 45 to 49, wherein the yeast cell furthermore has genotype I, such as genotype I as defined in any one of items 39 to 41.
- 30 51. The yeast cell according to any one of items 45 to 50, wherein the yeast cell furthermore has characteristic III, for example characteristic III as defined in any one of items 8 to 14.
- 35 52. The yeast cell according to any one of items 45 to 51, wherein the yeast cell furthermore has characteristic IV, for example characteristic IV as defined in any one of items 15 to 16.
- 40 53. The yeast cell according to any one of items 45 to 52, wherein the yeast cell furthermore has characteristic V, for example characteristic V as defined in any one of items 17 to 21.
- 45 54. The yeast cell according to any one of the preceding items, wherein the yeast cell has the genotype:

III. comprising a gene encoding UBR1.

55. A yeast cell having the genotype:

III. comprising a gene encoding UBR1.

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56. The yeast cell according to any one of items 54 to 55, wherein genotype III is said yeast cell comprising at least two allelic genes encoding UBR1 individually selected from the group consisting of UBR1 comprising SEQ ID NO:10, UBR1 of SEQ ID NO:11, UBR1 comprising SEQ ID NO:41, UBR1 of SEQ ID NO:42, UBR1 comprising SEQ ID NO:45 and functional homologues of any of the

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mentioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

57. The yeast cell according to any one of items 54 to 56, wherein genotype III is that said yeast cell comprises a gene encoding UBR1 selected from the group consisting of UBR1 comprising SEQ ID NO:10, UBR1 of SEQ ID NO:11 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

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58. The yeast cell according to any one of items 54 to 57, wherein genotype III is that the yeast cell comprises the following 2 genes:

1) a gene encoding UBR1 comprising SEQ ID NO:10 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith;

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2) a gene encoding UBR2 of SEQ ID NO:11 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

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59. The yeast cell according to any one of items 54 to 58, wherein the yeast cell furthermore has the genotype I, for example genotype I as defined in any one of items 39 to 41.

60. The yeast cell according to any one of items 55 to 59, wherein the yeast cell furthermore has genotype II, such as genotype II as defined in any one of items 46 and 49.
- 5 61. The yeast cell according to any one of items 55 to 60, wherein the yeast cell furthermore has characteristic III, for example characteristic III as defined in any one of items 8 to 14.
- 10 62. The yeast cell according to any one of items 55 to 61, wherein the yeast cell furthermore has characteristic IV, for example characteristic IV as defined in any one of items 15 to 16.
- 15 63. The yeast cell according to any one of items 55 to 62, wherein the yeast cell furthermore has characteristic V, for example characteristic V as defined in any one of items 17 to 21.
64. The yeast cell according to any one of the preceding items, wherein the yeast cell has the genotype:
- 20 IV. comprising at least 3 genes, such as at least 4 genes encoding IMA1p.
65. A yeast cell having the genotype:
- 25 IV. comprising at least 3 genes, such as at least 4 genes encoding IMA1p.
- 30 66. The yeast cell according to any one of items 64 and 65, wherein IMA1p is selected from the group consisting of IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15, IMA1p of SEQ ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:23, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

67. The yeast cell according to any one of items 64 to 66, wherein the genotype IV is that the yeast cell comprises at least 2 short alleles of IMA1, said two short alleles of IMA1 encoding IMA1p selected from the group consisting of IMA1p SEQ ID NO:12, IMA1p of SEQ ID NO:13 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

68. The yeast cell according to any one of items 64 to 67, wherein the genotype IV that said yeast cell comprises at least 3 short alleles of IMA1, which individually are genes encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:1, IMA1p of SEQ ID NO:2, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO: 4, IMA1p of SEQ ID NO: 5, IMA1p of SEQ ID NO:33 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

69. The yeast cell according to any one of items 64 to 68, wherein the genotype IV is that the yeast cell comprises at least 2 long alleles of IMA1, said two long alleles of IMA1 encoding IMA1p selected from the group consisting of IMA1p SEQ ID NO:14, IMA1p of SEQ ID NO:15 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

70. The yeast cell according to any one of items 64 to 69, wherein the genotype IV is that said yeast cell comprises at least 3 short alleles of IMA1 and at least 2 long alleles of IMA1, wherein

a) said 3 short alleles of IMA1 individually are genes encoding IMA1p selected from the group consisting of of IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:1, IMA1p of SEQ ID NO:2, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO: 4, IMA1p of SEQ ID NO: 5, IMA1p of SEQ ID NO:33 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and

5 b) said 2 long alleles of IMA1 individually are genes encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15, IMA1p of SEQ ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:23, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

10 71. The yeast cell according to any one of items 64 to 70, wherein the genotype IV is that the yeast cell comprises at least 5 genes encoding IMA1p, wherein said genes individually are selected from the group consisting of genes encoding IMA1p of SEQ ID NO:1, IMA1p of SEQ ID NO:2, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO:4, IMA1p of SEQ ID NO:5, IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15, IMA1p of SEQ ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:23, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and IMA1p of SEQ ID NO:33.

20 72. The yeast cell according to any one of items 64 to 71, wherein the genotype IV is that the yeast cell comprises the following 4 genes:

- 1) a gene encoding IMA1p of SEQ ID NO:12 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 25 2) a gene encoding IMA1p of SEQ ID NO:13 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 3) a gene encoding IMA1p of SEQ ID NO:14 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 30 4) a gene encoding IMA1p of SEQ ID NO:15 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

73. The yeast cell according to any one of items 64 to 72, wherein the genotype IV is the presence of at least 3 long alleles of IMA1 encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

74. The yeast cell according to any one of items 64 to 73, wherein the genotype IV is that the yeast cell comprises the following 3 genes:

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- 1) two genes both encoding IMA1p of SEQ ID NO:21 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) a gene encoding IMA1p of SEQ ID NO:22 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

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75. The yeast cell according to any one of items 64 to 74, wherein the genotype IV is that the yeast cell comprises the following 3 genes:

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- 1) a gene encoding IMA1p of SEQ ID NO:23 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) a gene encoding IMA1p of SEQ ID NO:24 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 3) a gene encoding IMA1p of SEQ ID NO:25 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

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76. The yeast cell according to any one of items 64 to 75, wherein the yeast cell furthermore has the genotype I, for example genotype I as defined in any one of items 39 to 41.

77. The yeast cell according to any one of items 64 to 76, wherein the yeast cell furthermore has genotype II, such as genotype II as defined in any one of items 46 and 49.
- 5 78. The yeast cell according to any one of items 64 to 77, wherein the yeast cell furthermore has genotype III, such as genotype III as defined in any one of items 55 and 58.
- 10 79. The yeast cell according to any one of items 64 to 78, wherein the yeast cell furthermore has characteristic I, for example characteristic I as defined in any one of items 3 to 4.
- 15 80. The yeast cell according to any one of items 64 to 79, wherein the yeast cell furthermore has characteristic II, for example characteristic II as defined in any one of items 1 and 5 to 7.
- 20 81. The yeast cell according to any one of items 64 to 80, wherein the yeast cell furthermore has characteristic IX, for example characteristic IX as defined in any one of items 28 to 35.
82. The yeast cell according to any one of the preceding items, wherein the yeast cell has the genotype
V. Comprising a gene encoding IMA5p.
- 25 83. A yeast cell having the genotype
V. Comprising a gene encoding IMA5p.
- 30 84. The yeast cell according to any one of items 82 and 83, wherein IMA5p is selected from the group consisting of IMA5p of SEQ ID NO:16, IMA5p of SEQ ID NO:17 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

85. The yeast cell according to any one of items 82 and 83, wherein IMA5p is selected from the group consisting of IMA5p of SEQ ID NO:34, IMA5p of SEQ ID NO:35, IMA5p of SEQ ID NO:36 and functional homologues of any of the
5 aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.
86. The yeast cell according to any one of items 82 to 85, wherein the genotype V is that the yeast cell comprise at least two genes encoding IMA5p of SEQ ID NO:16, or IMA5p of SEQ ID NO:17 or a functional homologue of any of the
10 aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.
87. The yeast cell according to any one of items 82 to 86, wherein the genotype V is that the yeast cell comprises at least two allelic genes encoding IMA5p
15 individually selected from allelic genes encoding IMA5p of SEQ ID NO:16, IMA5p of SEQ ID NO:17, IMA5p of SEQ ID NO:34, IMA5p of SEQ ID NO:35, IMA5p of SEQ ID NO:36 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.
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88. The yeast cell according to any one of items 82 to 87, wherein the genotype V is that the yeast cell comprises the following 2 genes:
- 25 1) a gene encoding IMA5p of SEQ ID NO:16 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 30 2) a gene encoding IMA5p of SEQ ID NO:17 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.
89. The yeast cell according to any one of items 82 to 88, wherein the yeast cell furthermore has the genotype I, for example genotype I as defined in any one of items 39 to 41.

90. The yeast cell according to any one of items 82 to 89, wherein the yeast cell furthermore has genotype II, such as genotype II as defined in any one of items 46 and 49.
- 5 91. The yeast cell according to any one of items 82 to 90, wherein the yeast cell furthermore has genotype III, such as genotype III as defined in any one of items 55 and 58.
- 10 92. The yeast cell according to any one of items 82 to 91, wherein the yeast cell furthermore has genotype IV, such as genotype IV as defined in any one of items 64 to 75.
93. The yeast cell according to any one of items 82 to 92, wherein the yeast cell furthermore has characteristic I, for example characteristic I as defined in any one of items 3 to 4.
- 15 94. The yeast cell according to any one of items 82 to 93, wherein the yeast cell furthermore has characteristic II, for example characteristic II as defined in any one of items 1 and 5 to 7.
- 20 95. The yeast cell according to any one of items 82 to 94, wherein the yeast cell furthermore has characteristic IX, for example characteristic IX as defined in any one of items 28 to 35.
- 25 96. The yeast cell according to any one of the preceding items, wherein the yeast cell has the genotype;
- 30 VI. comprising at least 3 genes encoding AGT1 selected from the group consisting of AGT1 of SEQ ID NO:18, AGT1 of SEQ ID NO:19, AGT1 of SEQ ID NO:20, AGT1 of SEQ ID NO:26, AGT1 of SEQ ID NO:27, AGT1 of SEQ ID NO:28, AGT1 of SEQ ID NO:29, AGT1 of SEQ ID NO:30, AGT1 of SEQ ID NO:31, AGT1 of SEQ ID NO:32 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

97. A yeast cell having the genotype;

VI. comprising at least 3 genes encoding AGT1 selected from the group

consisting of AGT1 of SEQ ID NO:18, AGT1 of SEQ ID NO:19, AGT1 of
SEQ ID NO:20, AGT1 of SEQ ID NO:26, AGT1 of SEQ ID NO:27, AGT1 of
SEQ ID NO:28, AGT1 of SEQ ID NO:29, AGT1 of SEQ ID NO:30, AGT1 of
SEQ ID NO:31, AGT1 of SEQ ID NO:32 and functional homologues of any
of the aforementioned sharing at least 80%, preferably at least 90%, yet
more preferably at least 95%, such as at least 98% sequence identity
therewith.

98. The yeast cell according to any one of items 96 to 97, wherein the yeast cell
comprises at least two genes encoding full length AGT1 selected from the group
consisting of AGT1 of SEQ ID NO:18, AGT1 of SEQ ID NO:19, AGT1 of SEQ ID

NO:20, AGT1 of SEQ ID NO:27, AGT1 of SEQ ID NO:28, AGT1 of SEQ ID NO:30,
AGT1 of SEQ ID NO:31, AGT1 of SEQ ID NO:32 and functional homologues of any of
the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably
at least 95%, such as at least 98% sequence identity therewith.

99. The yeast cell according to any one of items 96 to 98, wherein the genotype VI is
that the yeast cell comprises the following 3 genes:

- 1) a gene encoding AGT1 of SEQ ID NO:18 or a functional homologue thereof
sharing at least 80%, preferably at least 90%, yet more preferably at least 95%,
such as at least 98% sequence identity therewith; and
- 2) a gene encoding AGT1 of SEQ ID NO:19 or a functional homologue thereof
sharing at least 80%, preferably at least 90%, yet more preferably at least 95%,
such as at least 98% sequence identity therewith; and
- 3) a gene encoding AGT1 of SEQ ID NO:20 or a functional homologue thereof
sharing at least 80%, preferably at least 90%, yet more preferably at least 95%,
such as at least 98% sequence identity therewith.

100. The yeast cell according to any one of items 96 to 99, wherein the genotype VI is
that the yeast cell comprises the following 2 genes:

- 1) a gene encoding AGT1 of SEQ ID NO:27 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) a gene encoding AGT1 of SEQ ID NO:28 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

101. The yeast cell according to any one of items 96 to 100, wherein the genotype VI is that the yeast cell comprises the following 3 genes:

- 1) a gene encoding AGT1 of SEQ ID NO:30 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 2) gene encoding AGT1 of SEQ ID NO:31 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith; and
- 3) a gene encoding AGT1 of SEQ ID NO:32 or a functional homologue thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

102. The yeast cell according to any one of items 96 to 101, wherein the yeast cell furthermore has the genotype I, for example genotype I as defined in any one of items 39 to 41.

103. The yeast cell according to any one of items 96 to 102, wherein the yeast cell furthermore has genotype II, such as genotype II as defined in any one of items 46 and 49.

104. The yeast cell according to any one of items 96 to 103, wherein the yeast cell furthermore has genotype III, such as genotype III as defined in any one of items 55 and 58.

105. The yeast cell according to any one of items 96 to 104, wherein the yeast cell furthermore has genotype IV, such as genotype IV as defined in any one of items 64 to 75.
- 5 106. The yeast cell according to any one of items 96 to 105, wherein the yeast cell furthermore has genotype V, such as genotype V as defined in any one of items 82 to 88.
- 10 107. The yeast cell according to any one of items 96 to 106, wherein the yeast cell furthermore has characteristic I, for example characteristic I as defined in any one of items 3 to 4.
- 15 108. The yeast cell according to any one of items 96 to 107, wherein the yeast cell furthermore has characteristic II, for example characteristic II as defined in any one of items 1 and 5 to 7.
- 20 109. The yeast cell according to any one of items 96 to 108, wherein the yeast cell furthermore has characteristic IX, for example characteristic IX as defined in any one of items 28 to 35.
- 25 110. A method for producing a beverage, said method comprising the steps of
- a. Providing a starting liquid
 - b. Providing a yeast cell according to any one of items 1 to 109
 - c. Fermenting said starting liquid with said yeast cell,
- thereby producing a beverage.
- 30 111. The method according to item 110, wherein the starting liquid comprises an aqueous extract of barley.
112. The method according to any one of items 110 to 111, wherein the starting liquid comprises an aqueous extract of malt,

113. The method according any one of items 110 to 112, wherein the starting liquid is wort.

114. The method according to any one of items 110 to 113, wherein the fermentation is performed at a temperature in the range of 10 to 20°C.

Examples

The invention is further illustrated by the following examples, which should however not be construed as limiting for the invention.

In the examples the following yeast strains are used:

Name of yeast strain	Species/Description
Lager yeast 1	<i>S. pastorianus</i>
Lager yeast 2	<i>S. pastorianus</i>
Ale yeast 1	<i>S. cerevisiae</i>
<i>S. diastaticus</i> 1	<i>S. diastaticus</i>
Hybrid yeast 1	Hybrid between Ale yeast 1 and Lager yeast 1
Hybrid yeast 2	Hybrid between Lager yeast 2 and Ale yeast 1
Hybrid yeast 3	Hybrid between Lager yeast 2 and Ale yeast 1
Hybrid yeast 4	Hybrid between Lager yeast 2 and Ale yeast 1
Hybrid yeast 5	Hybrid between Lager yeast 2 and Ale yeast 1
Hybrid yeast 6	Hybrid between Lager yeast 2 and Ale yeast 1
Hybrid yeast 7	Hybrid between Lager yeast 2 and Ale yeast 1
Hybrid yeast 8	Hybrid between <i>S. diastaticus</i>

	1 and Ale yeast 1
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The genomic sequence of Hybrid yeast 1 is provided as SEQ ID NO:1 in priority founding Danish patent application PA 2014 70825. SEQ ID NO:1 of PA 2014 70825 shows the sequence of assembled scaffolds from the genomic sequence of Hybrid 1.

5 The sequences are provided in fasta format. The term "scaffold" as used in this connection refers to a portion of the genome sequence reconstructed from overlapped contigs. The term "contig" refers to a contiguous overlapping sequence originating from reassembly of short DNA fragments.

10 SEQ ID NO:1 of PA 2014 70825 provides the sequence of a total of 1629 scaffolds, numbered from number 0 to 1628. In SEQ ID NO:1 of PA 2014 70825 the sequences of each scaffold is provided separated by the term ">Scaffold_X", wherein X indicates the number of the scaffold having the following sequence.

15 Thus, the genome of Hybrid yeast 1 preferably comprises all of the Scaffolds 0 to 1628 distributed over a plurality of chromosomes.

The genomic sequence of Hybrid yeast 1 is also available under the DDBJ/EMBL/GenBank accession number LOQJ000000000. Thus, the Whole Genome
20 Shotgun project regarding Hybrid yeast 1 has been deposited at DDBJ/EMBL/GenBank under the accession LOQJ000000000. The version described in this patent is version LOQJ010000000.

The data of the submission were as follows:

25

SUBID	BioProject	BioSample	Accession
SUB1207553	PRJNA304272	SAMN04297180	LOQJ000000000

The Whole Genome Shotgun project shows the sequence of assembled scaffolds from the genomic sequence of Hybrid yeast 1. The term "scaffold" as used in this connection refers to a portion of the genome sequence reconstructed from overlapped contigs.

30 The term "contig" refers to a contiguous overlapping sequence originating from reassembly of short DNA fragments. The DDBJ/EMBL/GenBank the accession

LOQJ00000000, version LOQJ01000000 provides the sequence of a total of 8919 scaffolds. Thus, the genome of Hybrid yeast 1 preferably comprises all of the Scaffolds 0 to 8919 distributed over a plurality of chromosomes. Accordingly, a yeast cell according to the invention also may comprise all of the Scaffolds 0 to 8919 distributed over a plurality of chromosomes.

The genomic sequence of Hybrid yeast 7 is available under the DDBJ/EMBL/GenBank accession number LOQK00000000.

Thus, the Whole Genome Shotgun project regarding Hybrid yeast 7 has been deposited at DBJ/EMBL/GenBank under the accession LOQK00000000. The version described in this patent is version LOQK01000000.

The data of the submission were as follows:

SUBID	BioProject	BioSample	Accession
SUB1208131	PRJNA304273	SAMN04297181	LOQK00000000

The Whole Genome Shotgun project shows the sequence of assembled scaffolds from the genomic sequence of Hybrid yeast 7. The term "scaffold" as used in this connection refers to a portion of the genome sequence reconstructed from overlapped contigs. The term "contig" refers to a contiguous overlapping sequence originating from reassembly of short DNA fragments. The DDBJ/EMBL/GenBank the accession LOQK00000000, version LOQK01000000 provides the sequence of a total of 9492 scaffolds. Thus, the genome of Hybrid yeast 7 preferably comprises all of the Scaffolds 0 to 9492 distributed over a plurality of chromosomes. Accordingly, a yeast cell according to the invention also may comprise all of the Scaffolds 0 to 9492 distributed over a plurality of chromosomes.

Example 1

10 hl beer was prepared by inoculating 10 million of viable cells/ml of yeast to a commercial malt based wort (16° Plato) supplied by Soufflet followed by fermentation at 17°C until diacetyl was below a predefined threshold, which is set at a level below the

threshold considered off-flavor in lager beer. In the present example the diacetyl threshold was set to 30 ppb.

5 In lager beers, vicinal diketones such as diacetyl and 2,3-pentanedione give undesirable off-flavour if they are present over a threshold concentration. Both, diacetyl and 2,3-pentanedione have a butterscotch aroma, but the threshold for diacetyl is 10 times lower. Part of the fermentation management is to ensure that the finished beer contains vicinal diketones, especially diacetyl, below their thresholds.

10 The yeast cells were propagated in one tank (9 hL scale) that was used to get the cell inoculum of generation 1 of the beer that was then followed by generation 2 of the beer (10 hL scale each). The purpose of the propagation is to produce a healthy pure culture of yeast in sufficient amounts to inoculate yeast for the real beer fermentation. Beer fermentation is done in successive fermentations and yeast is typically replaced after 5
15 to 10 successive fermentations; however the frequency of introducing newly propagated yeast into the brewery is an individual decision. The successive fermentations serve as cell inoculum for the following beer fermentation and often only the propagation tank provides yeast cell inoculum for the first beer fermentation so-called generation 1 of the beer. The beer fermentation made after generation 1 is
20 called generation 2 and so on and so forth. Most beer fermentations are conducted with yeast drawn from previous beer fermentation and not from the propagation tank. The cell inoculum for the beer fermentation is typically 10^7 yeast cells/ml.

25 3 different yeast strains were used in this example. The same wort and fermentation conditions were employed.

Ale yeast 1

Lager yeast 1

Hybrid yeast 1

30 Ale yeast 1 is a yeast of the species *S. cerevisiae*. Lager yeast 1 is a yeast of the species *S. pastorianus*. Ale yeast 1 and Lager yeast 1 were hybridized and one of the hybrid strains was selected named Hybrid yeast 1.

Table 1 shows the final values of Plato, Table 2 shows % ethanol (% v/v) and Table 3 shows the final beer RDF, days to have diacetyl under the predefined threshold and days for the primary fermentation from beer of generation 1 and 2 in beer prepared using the 3 yeast strains at the end of the propagation tank (9 hL) and/or in generations 1 and 2 of beer.

Hybrid strain 1 has the ability to grow at 37°C (data not shown) and also ferment well at lower temperatures such as 16°C (see Tables 1 to 3).

10 **Table 1** Plato values

Final °PLATO	Lager yeast 1	Hybrid yeast 1	Ale yeast 1
At the end of Propagation (9 hL), 16 °C	2.54	1.97	2.03
End Generation 1, 16 °C	2.55	2.35	2.48
End Generation 2, 18°C	2.86	2.37	3.36
AVERAGE Generation 1+2	2.7	2.36	2.92

Table 2 % ethanol (% v/v)

% ETHANOL (% v/v)	Lager yeast 1	Hybrid yeast 1	Ale yeast 1
Generation 1, 16 °C	7.51	7.64	7.58
Generation 2, 18 °C	7.33	7.61	7.19
AVERAGE Generation 1+2	7.42	7.625	7.38

Table 3

10 hl Scale - Average of generation 1 and 2 (beer 02 and 03)			
name (yeast)	Lager yeast 1	Hybrid yeast 1	Ale yeast 1
Plato wort	15.85	15.85	15.85
Ferm. temp.	16 °C and 18 °C	16 °C and 18 °C	16 °C and 18 °C
Pitching rate	10	10	10
beer RDF (%)	69.3	71.0	68.9
days to DA in spec.	6.5	9	13
days to primary ferm.	5.5	6	9.5

Picthing rate is the amount of viable yeast/mL added as cell inoculum to start the fermentation. The hybrid yeast 1 had improved the RDF with 2% as compared to the two parental strains (Lager yeast 1 and Ale yeast 1)(See Table 3). Hybrid yeast 1 had also lower levels of final Plato.

The hybrid yeast 1 has improved ethanol yield by 0.2% ethanol or more as compared to the two parental strains. Hybrid yeast 1 has improved fermentation performance at both 16°C and 18°C of temperature. Hybrid 1 had improved in terms of having a shorter time to have diacetyl levels below threshold (Days to DA in spec.) as compared to Ale yeast 1

The hybrid yeast 1 is almost fermenting at the same rate as Lager yeast 1 (See days to primary ferment in Table 3) but it had a bit longer time to have diacetyl under threshold (See days to DA in spec.).

Example 2

Yeast cells from frozen stocks were streaked on YPD plates. They were used to inoculate 20 ml of pasteurized conventional malt wort in 50 ml bottles and grown at 22°C of temperature. Cell cultures from the 20 ml culture were used to repitch cells into 200 ml volume of wort into 500 ml bottles and grown at 22°C. From the 200 ml volume of wort, a propagation tank of 1.8 L was inoculated aiming at inoculating 14-15 million of viable cells/ml and growing at 16 or 18°C temperature (same as the fermentation

temperature). The malt used to prepare the wort was purchased from DMG in Denmark.

The number of total and viable cells was measured with NucleoCounter. From the propagation tank, the number of viable cells was also measured. 14-15 million of viable cells were used to inoculate 2 L wort with a sugar content of 15° Plato, which was allowed to ferment for 6 days at either 16°C (Hybrid yeast 2, 3 and 4 and its respective controls) or 18°C (Hybrid yeast 1 and its respective controls) to obtain the so-called generation 1 of beer. At the end of the generation 1, 14-15 million of viable cells were used to inoculate the generation 2 of beer. At day 4 of incubation the number of cells in suspension was determined. The resulting cell numbers from the beer from generation 2 are shown in Table 4. The number of cells in suspension does not reflect the overall growth of cells, but rather the flocculation and/or sedimentation. The number of cells in suspension is generally preferred to be as low as possible at the later stages of fermentation, which indicates increased flocculation and/or sedimentation. If the flocculation is increased too early in the process time, this can lead to premature flocculation resulting in sluggish fermentation at the end of the process.

Table 4a. Generation 2 of beer made in 2 L scale. The results shown are from biological duplicates of the same experiment.

18°C Fermentations		
Yeast	pitching rate mill/ml	cells in suspension (day 4) mill/ml
Lager yeast 1	15	17
Lager yeast 1	15	19
Ale yeast 1	15	12
Ale yeast 1	15	22.6
Hybrid yeast 1	15	3.58
Hybrid yeast 1	15	3.75

Hybrid yeast 1 produced more biomass (measured by grams of harvested yeast) than lager yeast 1 but still hybrid 1 had less cells in suspension.

Table 4b

16°C Fermentations Yeast	pitching rate mill/ml	cells in susp. (day 6) mill/ml
Lager yeast 1	14	20
Lager yeast 1	14	23
Ale yeast 1	14	14
Ale yeast 1	14	13
Hybrid yeast 2	14	2.3
Hybrid yeast 2	14	3.2
Hybrid yeast 3	14	4.4
Hybrid yeast 3	14	3.3
Hybrid yeast 4	14	5.8
Hybrid yeast 4	14	5
Lager yeast 2	14	12
Lager yeast 2	14	23

As shown above hybrid yeast 2, 3 and 4 have less cells in suspension than the lager yeasts, even though they produced a bit more biomass (in grams of harvested cells at the end of fermentation).

5

In a different trial, it was found that the hybrid yeast 2 had 7 mill/ml cells in suspension after 6 days of fermentation, whereas the ale yeast 1 had only 4 mill/ml cells in suspension and the Lager yeast 2 has 39 mill/ml cells in suspension after 6 days of fermentation. Thus, also in this trial the hybrid yeast had a much lower level of cells in suspension compared to the Lager yeast.

10

The hybrids made from Ale yeast 1 and Lager yeast 1 (Hybrid yeast 1) or Lager yeast 2 (Hybrids yeast 2, 3, 4 and 7) had less cells in suspension than the two parental strains: Ale yeast 1 and either of the lager strains (Lager yeast 1 or 2). So the hybrids had improved cell sedimentation. That is of interest in brewing to avoid downstream processing of the yeast cell paste that should be collected and used for the cell re-pitching of the next generations of the beer process.

15

Results from yet another trial is shown in Table 4c. The experimental settings were as described herein above except that the indicated yeast cells were tested.

5 **Table 4c**

18°C Generation 2 yeast	Pitching rate mill/ml	Yeast cells in suspension (Day 7) mill/ml
Lager yeast 2	15.0	26
Lager yeast 2	15.0	28
Hybrid yeast 7	15.0	4
Hybrid yeast 7	15.0	8.3
Hybrid yeast 8	15.0	7.2
Hybrid yeast 8	15.0	7.5
<i>S. diastaticus</i>	15.0	40
<i>S. diastaticus</i>	15.0	39

Example 3

10

50 L beer was prepared by inoculating conventional malt based wort (18° Plato) with 10 million of viable cells/ml of yeast followed by fermentation at 18°C. The malt used to prepare the wort was purchased from DMG in Denmark.

15

2 different yeast strains were used. The same wort and fermentation conditions were employed.

Lager yeast 1

Hybrid yeast 1

20

Table 5a shows the final values of beer AE for the 2 strains compared in the 50 L scale tank from beer made with generation 1 and 2.

AE as used herein is the “apparent extract” which is a measure of the density of beer wort in terms of the percentage of extract by weight and that is expressed in the Plato scale.

5 **Table 5a**

Lager 1 versus Hybrid 1 - test in 50L		
Generation 1 beer		
Strain name	Lager yeast 1	Hybrid 1
Generation	1	1
Plato wort (Starting value)	17.76	17.76
Fermentation temperature	18	18
Pitching rate	10	10
beer AE	3.21	2.71

Generation 2 of beer		
Strain name	Lager yeast 1	Hybrid 1
Generation	2	2
Plato wort (Starting value)	17.91	17.91
Fermentation temperature	18	18
Pitching rate	10	10
beer AE	3.46	2.86

10 The hybrid yeast 1 had improved 0.5 % AE by 0.5% as compared to the lager yeast 1.

A similar test was performed with Lager yeast 2, Hybrid yeast 4, Hybrid yeast 7, Hybrid yeast 8 and *S. diastaticus*. AE and RDF at day 7 after pitching are shown in Table 5b.

15 The experimental settings were as described herein in Experiment 2 above with the indicated yeast cells tested.

Table 5b

18°C Generation 2 Yeast	Pitching rate, mill/ml	AE after 7 days, %Plato	RDF, Day 7
Lager yeast 2	15.0	3.01	64.4
Lager yeast 2	15.0	2.93	66.3
Hybrid yeast 4	15.0	2.49	69.0
Hybrid yeast 4	15.0	2.53	68.8
Hybrid yeast 7	15.0	2.35	69.0
Hybrid yeast 7	15.0	2.39	69.6
Hybrid yeast 8	12.4	1.1	75.9
Hybrid yeast 8	12.4	1.12	75.1
<i>S. diastaticus</i>	14.7	1.67	73.1
<i>S. diastaticus</i>	15.0	1.66	73.0

	Lager yeast 2	Hybrid yeast 4	Hybrid yeast 7
Av. AE, 7 days	2.97	2.51	2.37
Extra AE increase vs Lager yeast 2		0.46	0.6
Av. RDF, 7 days	65.35	68.9	69.3
Extra RDF vs Lager yeast 2		3.55	3.95

RDF is provided in %.

5

The rate of fermentation was also determined for Lager yeast 2, Hybrid yeast 4 and 7. The experimental settings were as described herein in Experiment 2 except that the apparent extract was determined at several time points during the course of fermentation and incubation was at 18°C. Figure 16 shows the apparent extract in °

10 Plato over time after pitching wort. The wort used had a starting sugar content of 15°

Plato. As shown then the time of primary fermentation for Hybrid yeast 4 and 7 is about 3 days, whereas the time of primary fermentation for Lager yeast 2 is about 4 days.

Example 4

5

The amino acid content of starting wort and beer produced as described in Example 1 was determined by HPLC with fluorescence detector.

Table 6a shows the amino acid concentration in the final beer.

10

Table 6a

		Lager yeast 1	Ale yeast 1	Hybrid yeast 1	% amino acid reduction in final beer brew with Hybrid yeast 1 as compared to Lager yeast 1
Aspartic acid W	mg/l	32	51	3	90%
Glutamic acid W	mg/l	44	47	14	68%
Serine W	mg/l	11	23	8	27%
Histidine W	mg/l	42	46	25	40%
Glycine W	mg/l	49	57	35	28%
Threonine W	mg/l	12	16	5	58%
Arginine W	mg/l	152	136	86	43%
Alanine W	mg/l	147	145	80	45%
Tyrosine W	mg/l	129	140	118	8%
Methionine W	mg/l	26	28	10	61%
Valine W	mg/l	136	159	121	11%
Phenylalanine W	mg/l	134	166	117	12%
Isoleucine W	mg/l	59	71	33	44%
Leucine W	mg/l	113	173	78	31%
Lysine W	mg/l	29	25	1	97%

Hybrid yeast 1 has much less leftover amino acids in the final beer which is beneficial in terms of beer aging and beer stability. Beer fermented with hybrid 1 will generate lower amounts of strecker aldehydes, which are being formed from those amino acids.

- 5 Strecker aldehydes are important constituents of the “aged” flavor in beer that partly originate from the amino acids of the bottled beer itself. Amino acids that have been shown to be involved in formation of Strecker aldehydes with a low sensory threshold include valine, isoleucine, leucine, methionine and phenylalanine (Table 2). Strecker aldehyde formation plays a crucial role because an increase in their concentration,
10 gives an increasing sensory perception of “aged flavours”. Beer fermented with hybrid yeast 1 will have then less aging flavours due to the higher consumption of amino acids.

- 15 **Table 2.** Amino acids that act as precursors in the formation of off-flavour strecker aldehydes.

Amino acid	Strecker aldehyde
Methionine	Methional
Leucine	3-methylbutanal
Valine	2-methylpropanal
Isoleucine	2-methylbutanal
Phenylalanine	Phenylacetaldehyde, benzaldehyde

- Another fermentation with Lager yeast 2, Hybrid yeast 4 or Hybrid yeast 7 was prepared and the amino acid concentration in the “green beer” on the Day 7 of fermentation was determined. The result is shown in Table 6b. Experimental settings
20 were as described in Example 2 and amino acid analysis was done as described in Example 9. Hybrids, especially Hybrid yeast 7, has less amino acids left in the “green beer” than Lager yeast 2; it means that there are less precursors for the formation of the aging compounds in the beer;

- 25 **Table 6b**

Amino acid		Lager yeast 2	Hybrid yeast 4	% amino acid reduction in Hybrid yeast 4 in comparison with Lager yeast 2	Hybrid yeast 7	% amino acid reduction in Hybrid yeast 7 in comparison with Lager yeast 2
His	mg/L	40	25.5	36.25%	11.5	71.25%
Asn	- " -	8.5	5.5	35.3%	3	64.7%
Ser	- " -	3	3.5	na	0	100%
Gln	- " -	32	3	90.6%	0	100%
Arg	- " -	52	24	53.85%	6.5	87.5%
Gly	- " -	50.5	49.5	2.0%	26.5	47.5%
Asp	- " -	17.5	6.5	62.9%	2	88.6%
Glu	- " -	75.5	20	73.5%	8.5	88.7%
Thr	- " -	0	2	na	0	Na
Ala	- " -	175.5	131	25.3%	36	79.5
Pro	- " -	606.5	656.5	na	599.5	1.15%
Cys	- " -	0	0	na	0	Na
Lys	- " -	2	1	na	1	Na
Tyr	- " -	110	117	na	91	17.3%
Met	- " -	7.5	5.5	26.7%	2	73.3%
Val	- " -	113.5	117	na	66.5	43.2%
Ile	- " -	44	31.5	28.4%	6	86.4%
Leu	- " -	74.5	62	16.8%	18.5	75.2%
Phe	- " -	97	99	na	58	40.2%
Trp	- " -	51.5	51	1%	40.5	21.3%

(na) - not applicable;

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Example 5

50 L beer was prepared as specified in Example 3 by inoculating conventional malt based wort (16° Plato) prepared from two different kinds of malt.

Conventional malt based wort was inoculated with 10 million of viable cells/ml of yeast followed by fermentation at 16°C until diacetyl was under the threshold in beer specified in Example 1.

Another malt was inoculated with 15 million of viable cells/ml of yeast followed by fermentation at 16°C for Lager yeast 1 and 18°C for Hybrid yeast 1, until diacetyl was under the threshold in beer specified in Example 1.

2 different yeast strains and two different worts made of 2 different malts were used. The same wort and fermentation conditions were employed to compare the 2 strains in parallel.

Lager yeast 1

Hybrid yeast 1

The level of isomaltose and panose in the beer was determined by HPLC. The results are shown in Table 7.

Table 7

Yeast	Malt	Isomaltose (mg/L)	Panose (mg/L)
Lager yeast 1	Malt 1	390	300

Hybrid yeast 1	Malt 1	15	160
Lager yeast 1	Malt 2	240	240
Hybrid yeast 1	Malt 2	15	60

The starting concentration of panose and isomaltose is wort-dependent but it has been published that it is in the range of 0.5 to 1 g/L of isomaltose and 0.4 to 0.8 g/L of panose (Clapperton et al. 1971). Thus, it is believed that Hybrid yeast 1 utilizes in the range of 60% to 93% of the panose.

Quantitative data of panose and isomaltose utilization was obtained by measuring the growth of the different yeasts in defined medium with 2 g/L panose or 2 g/L isomaltose as sole carbon sources:

10

Yeast cells from frozen stocks were streaked on YPD plates (1% Yeast extract, 2% peptone, 2% glucose and 2% agar-agar) and growing cells were inoculated into liquid YPD (1% Yeast extract, 2% peptone, 2% glucose).

15

3 µL of the overnight liquid YPD culture was inoculated into 100 µL culture of YNB (6.7 g/L) without amino acids but with ammonium sulfate and buffered with potassium hydrogen phthalate to pH 5.5 (Hahn-Hägerdal B. et al. 2005) and with 2 g/L panose or 2 g/L isomaltose as sole carbon sources. Cell growth was followed by measuring the optical density at 600 nm with continuous agitation and incubating at 20°C of temperature using Bioscreen C MBR (Oy Growth Curves Ab Ltd, Finland).

20

The selected hybrid yeast strains of lager and ale with improved fermentation performance had acquired the ability to utilize panose (Figure 1A) and isomaltose (Figure 2). This sugar utilization improvement is exemplified with two hybrids in Figure 1A and 2A.

25

Similar experiments were performed with Ale yeast 1, Lager yeast 2 and Hybrid yeast 7, as well as with *S. diastolicus* and Hybrid yeast 8 using defined medium with 2 g/L panose as sole carbon source. The results are shown in figures 1B and 1C, respectively and are representative of biological replicates. As can be seen neither

Lager yeast 1, nor Lager yeast 2, nor *S. diastaticus* is capable of utilizing panose as sole carbon source.

5 Similar experiments were also performed with Ale yeast 1, Lager yeast 2 and Hybrid yeast 7 using defined medium with 2 g/L isomaltose as sole carbon source. The results are shown in figure 2B and are representative of biological replicates. As can be seen neither Lager yeast 1, nor Lager yeast 2, nor *S. diastaticus* is capable of utilizing isomaltose as sole carbon source.

10 References

Clapperton J F, MacWilliam I C (1971) Fermentation of minor wort carbohydrates by brewing yeasts. *Journal of the Institute of Brewing* 77, 6: 519-522.

15 Hahn-Hägerdal B, Karhumaa K, Larsson CU, Gorwa-Grauslund M, Görgens J, van Zyl WH. (2005) . Role of cultivation media in the development of yeast strains for large scale industrial use. *Microbial Cell Factories* 10: 4-31.

Example 6

20 Yeast cells from frozen stocks were streaked on YPD plates. They were used to inoculate 3 ml of liquid YPD and grown overnight under agitation at 22°C in 15 ml tubes. The 3 ml grown culture was centrifuged, the supernatant was discarded and the cells were dissolved in water. The tubes were centrifuged again, the supernatant was discarded and dissolved in 3 ml of water.

25 Optical density (OD_{620 nm}) was measured and adjusted to start at OD=0.2 for all the strains and solutions of the wells of the 96 well plates commercially provided by the Biolog Inc. technology (Hayward CA, USA). All the solutions of the Biolog plates were specified for procedures for *S. cerevisiae* and other yeasts. The 96 well plates were
30 incubated for 4,5 days at 22°C.

The Biolog system makes possible to assay quantitatively the level of thousands of cellular phenotypes in a single experiment. Each well of the assay is designed to test one individual phenotype. Biolog uses redox chemistry as a general reporter system to

analyze cell respiration. It has a tetrazolium dye that is reduced, developing colour, when the cell can respire the compound present in the well or in the presence of the compound in that specific well.

- 5 Three yeast strains were compared, the 2 parental strains (Lager yeast 1 and ale yeast 1) and the resultant hybrid (Hybrid yeast 1). Details on the yeast strains are provided in Example 1. In some of the conditions the 3 strains were showing differences in the phenotype. The hybrid strain 1 was able to gain new phenotypic characteristics for example the ability to utilize several dipeptides and some tripeptides (See Table 8a).

10

Table 8a

Peptides	Lager yeast 1	Ale yeast 1	Hybrid yeast 1
MET-TYR	-	-	+
LEU-TYR	-	-	+
VAL-MET	-	-	+
PHE-TYR	-	-	+
ILE-LEU	-	-	+
ILE-ASN	-	-	+
GLY-GLY-GLY	-	-	+

+ Indicates growth on medium containing the indicated peptides as only nitrogen source

- 15 - Indicates no growth on medium containing the indicated peptides as only nitrogen source.

In a similar experiment 6 yeast strains were compared, namely, the yeast strains Lager yeast 1, Lager yeast 2, Ale yeast 1, Hybrid yeast 1, Hybrid yeast 4 and Hybrid yeast 7. The results are shown herein below in Table 8b. As is seen Hybrids have new properties that is not observed in the parents. Lager yeast 1 showed minor growth on Ile-Asn in this experiment, even-though no growth was observed in the former experiment. However, the growth of Lager yeast 1 was still significantly less than the growth of Hybrid yeast 1.

20

Table 8b

Peptides	Lager yeast 1	Lager yeast 2	Ale yeast 1	Hybrid yeast 1	Hybrid yeast 4	Hybrid yeast 7
Gly-Arg	-	-	-	-/+	+	+
Ile-Asn	-/+	-	-	+	+	+
Lys-Tyr	-/+	-	-	-/+	+	+
Met-Lys	-/+	-	-	-/+	+	+
Val-Ala	-/+	-	-	+	+	+
Val-Asn	-	-	-	-/+	+	+
Val-Gly	-	-	-	-/+	+	+
Val-Gln	-	-	-	+	+	+
Val-Met	-	-	-	+	+	-/+
Val-Ser	-/+	-	-	+	+	+

Hybrid yeast 4 and 7 did not show any significant growth on Met-Tyr, Leu-Tyr, Phe-Tyr, Ile-Leu or GLy-Gly-Gly as sole nitrogen source.

The Hybrid yeast 1 also has acquired several phenotypes not found in Lager yeast 1, for example the ability to grow on several di-peptides having the formula Ala-Xaa, wherein Xaa may be any amino acid. This ability is often linked to the ability to utilize allantoin, which the Hybrid yeast also is capable of utilizing (see Table 9a).

Table 9a

Peptides	Lager yeast 1	Ale yeast 1	Hybrid yeast 1
Ala-Glu	-	+	+
Ala-Gly	-	+	+
Ala-His	-/+	+	+
Ala-Thr	-/+	+	+

Allantoin	-	+	+
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+ Indicates growth on medium containing the indicated peptides as only nitrogen source

- Indicates no growth on medium containing the indicated peptides as only nitrogen source.

5 -/+ meaning delayed growth or growth with a lag-phase

Also Hybrid yeast 4 and 7 are capable of utilizing Ala-Xaa dipeptides as sole nitrogen source as shown in Table 9b.

10 **Table 9b**

Peptides	Lager yeast 2	Hybrid yeast 4	Hybrid yeast 7
Ala-Glu	-	-/+	+
Ala-Gly	-	-/+	+
Ala-Thr	-/+	+	+

15 Dipeptides and tripeptides are part of FAN. FAN is free amino nitrogen, and it is a measure for nitrogen content of wort or beer. FAN is made of amino acids, ammonium ions and small peptides that are in the wort and they ensure desirable fermentation performance for the yeast (Lekkas C, et al. 2009). Many different dipeptide combinations may be found in wort.

20 Hybrid yeast 1 can utilize several different tested dipeptides/tripeptides that could be present in wort and has therefore a higher range of substrates from FAN that could be precursors for cell biomass, carbon source or precursors for flavours.

References

25 Lekkas C, Hill AE, Taidi B, Hodgson J, Stewart GG (2009). The role of small wort peptides in brewing fermentations. J. Inst. Brew. 115 (2), 134-139.

Example 7

Qualitative data of melibiose utilization was done by replica-plating YPD liquid cultures of yeasts grown in 96 well plate into YPGalactose plates (1% Yeast extract, 2% peptone, 2% galactose and 2% agar-agar) with 50 µg/ml of x-alpha-gal (Clontech, Mountain View, US) and incubating the plates for 5 days at 22°C. X-alpha gal is a chromogenic analog of melibiose and if the yeast is able to utilize melibiose the yeast colony will become blue and if the yeast is unable to utilize melibiose then the yeast colony will be white.

The results show (Table 9) that all the lager yeasts tested were positive for melibiose utilization (blue colony color), all the ale yeasts tested were negative for melibiose utilization (white colony color) and the hybrids were positive or negative for melibiose utilization (blue or white colony color) depending of what they have inherited.

Table 9

Yeast	Colour of yeast colony
Lager yeast 1, 2, 3 and 4	Blue
Ale yeast 1, 2, 3, 4 and 5	White
Hybrid yeast 1	White
Hybrid yeast 4	Blue
Hybrid yeast 5	Blue
Hybrid yeast 6	Blue

Quantitative data of melibiose was done by measuring the growth of yeast in defined medium with **2 g/L melibiose** as sole carbon source:

Yeast cells from frozen stocks were streaked on YPD plates and growing cells were inoculated into liquid YPD.

3 µL of the overnight liquid YPD culture was inoculated into 100 µL culture of YNB (6.7 g/L) without aminoacids but with ammonium sulfate buffered with potassium hydrogen phthalate to pH 5.5 (Hahn-Hägerdal B. et al. 2005) and with 2 g/L melibiose as carbon source. Cell growth was followed by measuring the optical density at 600 nm with

continuous agitation and incubating at 20°C of temperature using Bioscreen C MBR (Oy Growth Curves Ab Ltd, Finland). Hybrids of lager and ale had acquired the ability to utilize melibiose but not all the hybrids (this is exemplified with three hybrids in Figure 3)

5

References:

Hahn-Hägerdal B, Karhumaa K, Larsson CU, Gorwa-Grauslund M, Görgens J, van Zyl WH. (2005) . Role of cultivation media in the development of yeast strains for large scale industrial use. Microbial Cell Factories 10: 4-31.

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Example 8

Improved disaccharide and trisaccharide utilization

50 L beer was prepared as specified in Example 3 by inoculating malt based wort with 15 mill viable cells/ml followed by fermentation until diacetyl was under 30 ppb. The malt was prepared from conventional malting barley, or from null-LOX barley. 2 different yeast strains were used: lager yeast 1 and Hybrid yeast 1. The same wort and fermentation conditions were employed to compare the 2 strains in parallel except that fermentation with Lager yeast 1 was carried out at 18°C whereas the fermentation with Hybrid yeast 1 was carried out at 16°C. There were three independent brews prepared with the same strains and the level of different sugars was determined by NMR, Representative results are shown in Figure 4 for the levels of specific sugars that were different in the final bottled beer.

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The NMR results show that the hybrid yeast 1 had improved utilization of isomaltose, panose, nigerose, kojibiose and other unidentified carbohydrates (Figure 4). However lager yeast 1 is not able to utilize those sugars.

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The disaccharide isomaltose, maltulose and the trisaccharides panose and maltotriulose are minor sugars in the wort media used for brewing beer (Clapperton and MacWilliam 1971). Our results show that there is also other disasaccharides like nigerose, kojibiose and trehalose that were present in the beer brewed with lager yeast 1 (Figure 4). Improvement of the sugar utilization of those sugars present in low

amounts can account for better total sugar utilization and an improvement of ethanol yield production by Hybrid yeast 1.

Example 9

5 50 L of beer from generation 1 was prepared by inoculating conventional malt based wort (13.6° Plato) prepared from all malt with 15 million of viable yeast cells/ml as inoculum followed by fermentation at 18°C for 5 days and at 14°C for 2 days. The cell inoculum was obtained from a previous propagation tank. The malt used to prepare the wort was purchased from DMG in Denmark. At day 6, samples of fermented wort,
10 corresponding to the samples of "green beer", were taken and centrifuged and the supernatant was frozen down at -20°C until it was analyzed (Table 10). The concentration of free amino acids were determined by UPLC with Photo Diode Array detection using the AccQ-Tag Ultra derivatization kit from Waters, essentially as described by the supplier. Separations were performed on a Waters AccQ-Tag Ultra
15 Amino acid Analysis Column using premixed eluent A and B according to manufacturer's instructions (Waters). A sample of the original wort used to ferment was also compared with the samples that have been fermented. Amino acid concentration was compared between all the green beer samples versus a sample of the original wort (Table 10).

20

The level of residual amino acids in green beer fermented with hybrid yeasts are much lower compared to green beer fermented with Lager yeast 1, which is beneficial in terms of beer aging and beer stability. It is believed that in beer made with hybrid yeasts fewer strecker aldehydes will be formed from those amino acids during storage.
25 Strecker aldehydes are important constituents of the "aged" flavor in beer that partly originate from the amino acids of the bottled beer itself. Amino acids that have shown to form strecker aldehydes with a low sensory threshold are valine, isoleucine, leucine, methionine and phenylalanine (Baert, De Clippeleer et al. 2012). Strecker aldehyde formation plays a crucial role because an increase in their concentration, gives an
30 increasing sensory perception of "aged flavours". Beer brewed with hybrid yeast 1 and 4 will have less aging flavours due to the higher consumption in amino acids and this effect will be more pronounced in high gravity fermentations or wort malts with higher concentrations of FAN sources.

The amino acid proline was also utilized by the hybrid yeasts 1 and 4 but not by the lager yeast in the green beer. The amino acid proline is the major amino acid constituent in wort although it is the most difficult to be assimilated so hybrid yeasts with improved proline utilization will have this extra ability of utilizing this nitrogen source that lager yeast cannot utilize.

5

Table 10. Aminoacid analysis (mg/L) of green beer from day

	His	Asn	Ser	Gln	Arg	Gly	Asp
ORIGINAL WORT	44	78	65	21	93	27	56
GREEN BEER Hybrid Yeast 1 Total level (% of level in wort)	4 (9%)	2 (3%)	0 (0%)	15 (71%)	0 (0%)	6 (22%)	0 (0%)
GREEN BEER Lager Yeast 1 Total level (% of level in wort)	18 (41%)	3 (4%)	2 (3%)	20 (95%)	11 (12%)	20 (74%)	3 (5%)
GREEN BEER Hybrid Yeast 4 Total level (% of level in wort)	10 (23%)	2 (3%)	0 (0%)	17 (81%)	3 (3%)	11 (41%)	0 (0%)
	Glu	Thr	Ala	Pro	Cys	Lys	Tyr
ORIGINAL WORT	84	50	92	270	0	74	78
GREEN BEER Hybrid Yeast 1 Total level (% of level in wort)	0 (0%)	0 (0%)	4 (4%)	227 (84%)	0 (0%)	2 (3%)	0 (0%)
GREEN BEER Lager Yeast 1 Total level (% of level in wort)	21 (25%)	0 (0%)	68 (74%)	270 (100%)	0 (0%)	4 (5%)	22 (28%)
GREEN BEER Hybrid Yeast 4 Total level (% of level in wort)	2 (2%)	0 (0%)	14 (15%)	250 (93%)	13 -	2 (3%)	0 (0%)
	Met	Val	Ile	Leu	Phe	Trp	Total Sum
ORIGINAL WORT	24	88	47	112	98	48	1448
GREEN BEER Hybrid Yeast 1 Total level (% of level in wort)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	3 (6%)	263 (18%)
GREEN BEER Lager Yeast 1 Total level (% of level in wort)	0 (0%)	15 (17%)	2 (4%)	2 (2%)	10 (10%)	28 (58%)	518 (36%)
GREEN BEER Hybrid Yeast 4 Total level (% of level in wort)	0 (0%)	2 (2%)	0 (0%)	2 (2%)	0 (0%)	18 (38%)	346 (24%)

Baert, J. J., J. De Clippeleer, P. S. Hughes, L. De Cooman and G. Aerts (2012). "On the Origin of Free and Bound Staling Aldehydes in Beer." Journal of Agricultural and Food Chemistry **60**(46): 11449-11472.

5

Example 10

The genomic sequence of the Hybrid yeast 1 described in Example 1 was determined as follows.

10

Genomic DNA extraction and whole genome sequencing and assembly of the genome was done by LGC Genomics GmbH (Berlin, Germany). For extra individual sequencing of genes, genomic DNA extraction of the strains was performed by MasterPure™ Yeast DNA Purification Kit (Epicentre, Illumina Denmark ApS, Copenhagen, Denmark).

15

PCR amplification from genomic DNA was performed with High Fidelity PCR enzyme mix or Dream Taq polymerase with a low number of PCR cycles both of them from Thermo Fisher Scientific Baltics UAB (Vilnius, Lithuania). PCR products were purified by NucleoSpin PCR Clean-up (Macherey-Nagel, Düren, Germany). Cloning of the PCR products was carried out with the TOPO®TA Cloning® Kit for Sequencing and selected on LB ampicillin plates supplemented with beta-X-Galactose. Plasmids were purified with the GeneJET Plasmid Miniprep kit from Thermo Fisher Scientific Baltics UAB (Vilnius, Lithuania). Plasmid sequencing was performed at Eurofins Genomics (Ebersberg, Germany).

20

25

Hybrid yeast 1, Lager yeast 1 and Ale yeast 1 were analysed for various selected genes using either the assembled genome sequence and/or the sequence of PCR products. The protein sequence was deduced from the gene sequence or from the sequence of the PCR products using the genetic code.

30

Hybrid yeast 4, Hybrid yeast 7 and Lager yeast 2 were analysed for various selected genes using the sequence of PCR products. Sequences were obtained by PCR amplification with High Fidelity PCR enzyme, followed by cloning and sequencing of the individual PCR clones when applicable. The allelic genes of Hybrid yeast 4 and Hybrid yeast 7 were identified by PCR, cloning and sequencing. The allelic genes of

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Lager yeast 2 were identified by PCR and sequencing (IMA1) or was assembled from

the genomic sequence (AGT1). The protein sequence was deduced from the sequence of the PCR products using the genetic code.

The LONG-IMA1 alleles presented here are defined by a 3 amino acids combination: I (Isoleucine) or T (Threonine) at the position 165, R (Arginine) or K (Lysine) at the position 287, and Y (Tyrosine) or F (Phenylalanine) at the position 336. The amino acid signature motif for Ale 1 LONG_IMA1 alleles being *I-R-F* and *I-K-F*. Lager yeast 1 and Lager yeast 2 have *T-R-Y* motif. Hybrid yeast 1 contains an *I-K-F* allele and a *T-R-Y* allele. Hybrid yeast 4 contains an *I-R-F* motif. Hybrid yeast 4 also contains a new hybrid allele with signature motif of *I-R-Y*. Hybrid yeast 7 contains an *I-K-F* allele, and an *T-R-Y* allele. Hybrid yeast 7 also contains a new hybrid allele with *I-R-Y* motif that is identical to the protein encoded for by Hybrid yeast_4.

Table 11a summarizes the status of various gene involved in dipeptide utilization in Lager Yeast 1, Ale yeast 1 and Hybrid yeast 1.

Table 11a

Gene	Function	Lager yeast 1	Ale yeast 1	Hybrid yeast 1
<i>DAL5*</i>	Transport of dipeptides with Ala at N-terminus	<i>nonSc</i>	<i>Sc</i>	<i>Sc</i> allele encoding SEQ ID NO:6
<i>PTR2**</i>	Peptide transporter (di-/tripeptides)	<i>Sc</i> ; <i>nonSc</i>	<i>Sc</i>	2 <i>Sc</i> alleles encoding SEQ ID NO:7 and SEQ ID NO:8 <i>nonSc</i> allele encoding SEQ ID:9
<i>UBR1***</i>	E3 ubiquitin ligase (N-	<i>Sc</i> ; <i>nonSc</i>	<i>Sc</i> (early stop codon –	<i>Sc</i> allele encoding UBR1 comprising

	recognin): peptide degradation pathway		truncated protein)	SEQ ID NO:10; <i>nonSc</i> allele encoding SEQ ID NO:11
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*DAL5 is an example of a gene inherited by Hybrid yeast 1 from parent Ale yeast 1. The term Sc in relation to DAL5 refers to the *S. cerevisiae* DAL5 protein of SEQ ID NO: 6. An alignment of DAL5 protein sequences is shown in figure 5.

5

**PTR2 is an example, where Hybrid yeast 1 has an increase copy number. Thus, Hybrid yeast 1 has inherited a *nonSc* allele from Lager yeast 1 and at least 2 Sc alleles from both its parents. Only fragments of the PTR2 alleles of Hybrid yeast 1 were investigated.

10

***UBR1 is an example of a gene with activity complementation in Hybrid yeast 1. Ale yeast 1 encodes a truncated protein (900 aa instead of 1951 aa) that leads to the absence of the domain responsible for Cup9p degradation activation and correspondingly to repression of *PTR2* expression; Hybrid yeast 1 has inherited both Sc and *nonSc* alleles from Lager yeast 1 thus complementing the Ubr1p activity toward Cup9p degradation, and correspondingly ability of *PTR2* expression to be activated further. An alignment of the Sc alleles of PTR2 from Ale yeast 1, Hybrid yeast 1 and Lager yeast 1 is shown in figure 6, and an alignment of the non Sc alleles of Lager yeast 1 and Hybrid yeast 1 is shown in figure 7.

15

20

The genomic sequences of Hybrid yeast 1 and Hybrid yeast 7 were further studied and the results summarized in Table 11b. These analyses are based on the genomic sequences available under DDBJ/EMBL/GenBank accession number LOQJ000000000, version LOQJ01000000 and DDBJ/EMBL/GenBank the accession LOQK000000000, version LOQK01000000TPTR2, respectively.

25

PTR2 analysis of the allelic variation was done based on the genomic sequences (see accession numbers above). Hybrid yeast 1 and Hybrid yeast 7 both retained nonSc_PTR2 copy. In Table 11a a fragmented Sc_copy of Hybrid yeast 1 is presented. Table 11b shows the intact copy of Sc_PTR2 in Hybrid yeast 1 in the genomic

30

sequence. It is possible that Hybrid yeast 1 contains 3 alleles encoding PTR2 as indicated in Table 11a. Hybrid yeast 7 has Sc_PTR2 as well. In both Hybrids Sc_PTR2 protein sequence shows hybridization between Sc_PTR2 Ale yeast 1 and Sc_PTR2 Lager yeast 1 and 2 copies.

5

DAL5 analysis of the allelic variation was done based on the genomic sequences (see accession numbers above). Hybrid yeast 1 and Hybrid yeast 7 has retained Sc_DAL5 from Ale yeast 1. Hybrid yeast 7 has retained nonSc_DAL5 as well.

UBR1 analysis of the allelic variation was done based on the the genomic sequences (see accession numbers above). Both Lager parental yeast has Sc_copy that is truncated differently. Previously, Lager 1 genomic data search yielded only fragmented sequence that did not allowed to determine early stop codon . Both Hybrid yeasts retained nonSc_UBR1 from Lager parent. Sc_copy detected in both Hybrids was inherited from Ale 1 parent.

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Table 11b

	Lager yeast 2 (Lager 2)	Ale yeast 1 (Ale 1)	Hybrid yeast 1 (Hybrid 1)	Hybrid yeast 7 (Hybrid 7)
<i>PTR2</i>	Sc_copy nonSc_copy	Sc_copy with 6 amino acids difference from Lagers Sc-copy	One Sc_like <u>hybrid</u> allele encoding SEQ ID NO:43; One nonSc allele encoding SEQ ID NO:44	One Sc_like <u>hybrid</u> allele encoding a protein comprising SEQ ID NO:37; One nonSc allele encoding SEQ ID NO:38
<i>DAL5</i>	nonSc_copy	Sc_copy	One Sc_copy (see Table 11a)	One Sc allele encoding SEQ ID NO:39; One nonSc allele encoding SEQ ID NO:40
<i>UBR1</i>	Sc_copy truncated, 1544 ; nonSc_copy	Sc_copy truncated, 900 amino acids;	One Sc allele encoding protein comprising SEQ ID NO:45 One nonSc allele (see Table 11a)	One Sc_allele – same as in Ale 1 encoding a protein comprising SEQ ID NO:41; One nonSc_allele encoding SEQ ID NO:42

Table 12a summarizes the status of various genes involved in sugar utilization in Lager Yeast 1, Ale yeast 1 and Hybrid yeast 1.

	Lager 1	Ale 1	Hybrid 1
<i>IMA1</i>_Sc_allele_short	Not found	1 allele	2 alleles encoding SEQ ID NO:12 and SEQ ID NO:13, respectively.
<i>IMA1</i>_Sc_allele_long	1 allele encoding protein with 99% amino acid sequence identity with <i>Saccharomyces cerevisiae</i> AWRI1631 and Kyokai no. 7	2 alleles	1 allele similar to Lager 1 encoding SEQ ID NO:14 1 allele with similar Ale 1 allele encoding SEQ ID NO:15
<i>IMA5</i>	<u>2 alleles:</u> <u>Allele 1:</u> Sc- <i>IMA5</i> like copy , <u>Allele 2:</u> non-sc- <i>IMA5</i>	<u>1 allele :</u> Sc- <i>IMA5</i> like copy	<u>2 alleles :</u> <u>Non-Sc-IMA5</u> like copy encoding SEQ ID NO: 16 <u>Sc-IMA5-like</u> similar to Ale 1 allele encoding SEQ ID NO: 17
AGT1	<u>2 alleles but only one functional:</u> <u>Allele 1:</u> Sc copy has STOP codon (extra T in poly-T) → Truncated	<u>1 allele:</u> Sc COPY	<u>3 alleles:</u> Non-sc copy 100% identical to non-sc copy of AGT1 found in Lager 1. The non-sc copy of AGT1 encodes SEQ ID NO: 18.

	CDS. <u>Allele 2: Non-Sc copy</u> encoding protein with 87% sequence identity to S288C		2 Sc alleles very similar to the Sc copy found in Ale 1 encoding SEQ ID NO:19 and SEQ ID NO:20 , respectively.
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Table 12b summarizes the status of various genes involved in sugar utilization in Lager Yeast 2, Hybrid yeast 4 and Hybrid yeast 7.

	Lager yeast 2	Hybrid yeast 4	Hybrid yeast 7
<i>LONG_IMA1</i>	<u>1 allele:</u>	3 alleles: Two alleles encoding SEQ ID NO:21 and one allele encoding SEQ ID NO:22. All alleles have hybrid nature based on nucleotide sequence.	3 alleles: One allele is identical to Lager 2 allele and encodes SEQ ID NO:24; one allele is identical to one of the ale 1 alleles and encodes SEQ ID NO: 23; and one allele has hybrid nature and encodes SEQ ID NO:25.
<i>AGT1</i>	2 alleles: <u>Allele 1: Sc allele contains a premature STOP codon, and</u>	3 alleles: <u>Allele 1: Sc allele containing a STOP codon</u> encoding a	<u>4 alleles:</u> <u>2 non-Sc alleles:</u> one identical to Lager 2 non-Sc copy encoding SEQ ID NO:31, and

	<u>encodes a truncated protein</u> <u>Allele 2: Non-Sc allele</u>	truncated protein, SEQ ID NO: 26. <u>2 nonSc alleles:</u> one identical to Lager 2 nonSc copy encoding SEQ ID NO:28, and one encoding SEQ ID NO:27 having 1 amino acid change	one encoding SEQ ID NO:32 having 1 amino acid change <u>2 Sc alleles:</u> one identical to Ale 1 Sc allele encoding SEQ ID NO:30, and another identical to Lager 2 Sc allele encoding a truncated protein SEQ ID NO:29.
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Several genes may be involved in utilization of isomaltose. This includes Agt1p that is a sugar transporter that can transport isomaltose. Furthermore, there are 5 different isomaltase enzymes which are alpha-1,6-glucosidases but which also have other glucosidase activities.

Based on genomic sequence information no **IMA1 gene** with a full coding sequence was identified in lager yeast 1 and other *S. pastorianus* strains found in the NCBI database also did not have a copy of the *S. cerevisiae* *IMA1* gene. Interestingly, Hybrid yeast 1 contains 4 different alleles of the *IMA1* gene of two different lengths.

IMA5-like sequence is present in the genome of lager yeast 1. In Hybrid yeast 1 there were 2 alleles, one Non-*S. cerevisiae* copy identical to the lager yeast 1 and one *S. cerevisiae* copy very similar to the sequence found in Ale 1 yeast but with 3 amino acid changes.

The transport of maltotriose and isomaltose has been shown to be facilitated by the high-affinity alpha-glucoside transporter encoded by the gene *AGT1*. This transporter has a broad substrate specificity.

We found in the lager yeast 1 only one full copy of the *AGT1* transporter of Non-*S. cerevisiae* origin, the *S. cerevisiae* copy was truncated. Hybrid yeast 1 in contrast had 3 full copies of *AGT1* gene, one identical to the Non-*S. cerevisiae* copy found in lager

yeast 1, and two *S. cerevisiae* alleles very similar to the AGT1 genes found in Ale yeast 1 but with one amino acid change each.

5 In Hybrid yeast 4, 2 full length copies of Non-*S. cerevisiae* AGT1 were identified with one copy carrying 1 amino acid change. In Hybrid yeast 7, 3 full length copies were identified: one fully identical to Ale yeast 1 AGT1, and 2 alleles of non-*S. cerevisiae* AGT1 with one copy carrying 1 amino acid change.

10 In addition to study of the genomic sequence as described above, additional information was obtained on IMA1 short in Lager yeast 1 and Lager yeast 2, as well as in Hybrid yeast 1, Hybrid yeast 4 and Hybrid yeast 7 by cloning and sequencing using specific primers for IMA1_short locus of Ale yeast 1 as described above. Based on genomic sequence information, no IMA1_short gene was found in Lager 1 and Lager 2 yeasts genomic sequences. However, cloning and sequencing of the IMA1-short from 15 both Lager yeasts parents showed that one gene is present, but it encodes a protein with 6 amino acid difference from the corresponding Ale yeast 1 IMA1 short protein. The data is summarized in Table 12c below.

20 In Hybrid yeast 1 cloning and sequencing identified 3 IMA1 short alleles – the two alleles described herein above and an additional allele with hybrid nature based on nucleotide sequence and also on the protein level (protein sequence provided as SEQ ID NO:1). Hybrid yeasts 4 and 7 retained IMA1_short from both parents, but cloning also identified additional alleles with unique amino acids change in both Hybrids. Thus, all three hybrid strains contained 3 IMA1 short alleles.

25 IMA5 analysis of the allelic variation was done based on the sequences available in the genomic sequences. In addition to the alleles described above in Table 12a, Hybrid yeast 1 contained one additional allele. Thus Hybrid yeast 1 and Hybrid yeast 7 both retained Sc_IMA5 and nonSc_IMA5 copies. Hybrid yeast 7 has unique hybrid 30 Sc_IMA5 allele resulted from recombination between Ale yeast 1 and Lager yeast 2 at Sc_IMA5 locus. Hybridization is evident from nucleotide sequences, and visible on the protein sequence.

Table 12c

	Lager yeast 1 (Lager 1)	Lager yeast 2 (Lager 2)	Ale yeast 1 (Ale 1)
<i>IMA1</i> short	<u>1 allele</u> cloned by PCR with 6 amino acids difference from Ale 1	<u>1 allele</u> cloned by PCR and identical to Lager 1 (SEQ ID NO:2)	<u>1 allele</u> (confirmed by cloning)
<i>IMA5</i>	<u>2 alleles</u> : <i>Sc_IMA5</i> copy (3 amino acids difference with Ale 1) <i>nonSc_IMA5</i>	<u>2 alleles</u> : <i>Sc_IMA5</i> I copy (identical to Lager 1) <i>nonSc_IMA5</i> (has 2 SNPs with respect to lager 1 in coding region)	<u>1 allele</u> : <i>Sc-IMA5</i> copy
	Hybrid yeast 1 (Hybrid 1)	Hybrid yeast 4 (Hybrid 4)	Hybrid yeast 7 (Hybrid 7)
<i>IMA1</i> short	<u>3 alleles</u> found through cloning: One Allele (vA) is identical to allele of Ale 1 and one allele (vB) is as Ale1 with one amino acid change. These two alleles encodes SEQ ID NO:12 and SEQ ID NO:13. One allele is unique in the Hybrid 1 and encodes SEQ ID NO:1	<u>3 alleles</u> found through cloning: One allele identical to allele of Lager 2 and encodes SEQ ID NO:2; One allele is identical to allele of Ale 1 encoding SEQ ID NO:3, and another allele has a nucleotide change, but also encodes SEQ ID NO:3.	<u>3 alleles</u> found through cloning: One allele is identical to allele from Lager 2 and encodes SEQ ID NO:5 One allele is identical to allele from Ale 1 and encodes SEQ ID NO:33; One allele is similar to allele from Lager 2 with nucleotide change and unique amino acid change (SEQ ID NO:4)
<i>IMA5</i>	<u>3 alleles</u> : One <i>Sc_IMA5</i> allele similar to allele of Ale 1 encoding SEQ ID NO:17; One unique allele identified by cloning encoding <i>IMA5</i> with unique 3 amino acid changes (SEQ ID NO:34) One <i>NonSc_IMA5</i> allele like allele of lager 1 encoding SEQ ID NO;16	ND	<u>2 alleles</u> : One <i>Sc_IMA5</i> allele, which is a unique hybrid copy encoding SEQ ID NO:35 One <i>nonSc_IMA5</i> allele identical to allele of lager 2 encoding SEQ ID NO; 36

Example 11

5 Data of maltulose, maltotriose and kojibiose utilization was obtained by measuring the growth of the different yeasts in defined medium with 2 g/L maltulose or 2 g/L maltotriose or 2 g/L kojibiose as sole carbon sources:

10 Yeast cells from frozen stocks were streaked on YPD plates (1% Yeast extract, 2% peptone, 2% glucose and 2% agar-agar) and growing cells were inoculated into liquid YPD (1% Yeast extract, 2% peptone, 2% glucose).

15 3µL of the overnight liquid YPD culture was inoculated into 100 µL culture of YNB (6.7 g/L) without amino acids but with ammonium sulfate and buffered with potassium hydrogen phthalate to pH 5.5 (Hahn-Hägerdal B. et al. 2005) and with 2 g/L maltulose or 2 g/L maltotriose or 2 g/L kojibiose as sole carbon sources.

Cell growth was followed by measuring the optical density at 600 nm with continuous agitation and incubating at 20°C of temperature using Bioscreen C MBR (Oy Growth Curves Ab Ltd, Finland).

20

It was tested whether Ale yeast 1, Hybrid yeast1, Hybrid yeast 4 and Hybrid yeast 7 are capable of utilizing maltotriose as sole carbon source. The results are shown in figure 13.

25 It was also tested whether Ale yeast 1, Hybrid yeast1, Hybrid yeast 4, Hybrid yeast 7, *S. diastaticus* and Hybrid yeast 8 are capable of utilizing maltulose as sole carbon source. The results are shown in figure 14.

30 It was also tested whether Ale yeast 1, Lager yeast 1, Lager yeast 2, *S. diastaticus*, Hybrid yeast 1, Hybrid yeast 4, Hybrid yeast 7 and Hybrid yeast 8 are capable of utilizing kojibiose as sole carbon source. The results are shown in figure 15. As can be seen neither Lager yeast 1 nor Lager yeast 2 can utilize kojibiose as sole carbon source, whereas *S. diastaticus* only can utilize kojibiose sole carbon source very poorly.

Example 12

To further investigate the real degree of fermentation obtained when fermenting using the Hybrid yeast 7, large scale trials were performed. Wort prepared in large scale from different mixtures were fermented in different locations with either Lager yeast 2 or with Hybrid yeast 7 until diacetyl was in spec. The real degree of fermentation (RDF) was determined. Table 13 shows the absolute increase in %RDF obtained after fermentation with Hybrid yeast 7 compared with the RDF obtained after fermentation with Lager yeast 2.

Wort was prepared by mashing different ratios of malt, barley (i.e. unmalted barley kernels) and rice. In addition varying amounts of glucose syrup was added. The ration of malt:barley:glucose syrup:rie used for preparing the different wort are also indicated in table 13.

Table 13

Country	Poland	India	Finland	Russia
Recipe (malt:barley:glucose syrup: rice)	51:20:29:0	35:0:0:65	68:22:10:0	73.20:7.0
RDF increase	2%	1%	3%	2.3%

Abbreviations

RDF – real degree of fermentation

YPD - (1% Yeast extract, 2% peptone, 2% glucose)

YPD plates (1% Yeast extract, 2% peptone, 2% glucose and 2% agar-agar)

YNB (Yeast Nitrogen Base)

OD: Optical Density

HPLC: High Performance Liquid Chromatography

YPGalactose plates (1% Yeast extract, 2% peptone, 2% galactose and 2% agar-agar)

Claims

1. A yeast cell having at least one of the following characteristics::
 - 5 II. capable of utilizing panose as sole carbon source;
 - III. capable of utilizing one or more dipeptides as sole nitrogen source.
2. The yeast cell according to any one of the preceding claims, wherein the yeast cell furthermore has characteristic:
 - 10 I. Capable of utilizing isomaltose as sole carbon source.
3. The yeast cell according to any one of the preceding claims, wherein said yeast cell is capable of utilizing panose as sole carbon source when said panose is present at
15 a concentration of in the range of 1 to 5 g/L, such as on the range of 1 to 3 g/L, for example 2 g/L.
4. The yeast cell according to any one of the preceding claims, wherein said yeast cell is capable of removing at least 45% of the panose present in wort.
20
5. The yeast cell according to any one of the preceding claims, wherein the yeast cell further has the characteristic:
 - III. capable of utilizing one or more dipeptides as sole nitrogen source.
- 25 6. The yeast cell according to any one of the preceding claims, wherein the yeast cell furthermore is capable of utilizing allantoin as sole nitrogen source.
7. The yeast cell according to any one of the preceding claims, wherein the yeast cell further has the characteristic:
 - 30 IV. capable of utilizing one or more tri-peptides as sole nitrogen source.
8. The yeast cell according to any one of the preceding claims, wherein the yeast cell further has the characteristic:

V. capable of reducing the level of one or more amino acids to no more than 10% of the starting concentration after incubation for 5 days under conditions allowing growth of said yeast cells.

- 5 9. The yeast cell according to any one of the preceding claims, wherein the yeast cell further has the characteristic:

VI. capable of generating at least 4.7 promille ethanol per °Plato, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

10

10. The yeast cell according to any one of the preceding claims, wherein the yeast cell further has the characteristic:

VII. capable of fermenting sugar with a real degree of fermentation of at least 68, such as at least 70, when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated until level of diacetyl is in spec.

15

11. The yeast cell according to any one of the preceding claims, wherein the yeast cell has characteristic VII, wherein characteristic VII is that the yeast cell is capable for fermenting sugar with an RDF, which is at least 1, such as at least 2 higher than the RDF of one of its parental strains.

20

12. The yeast cell according to any one of the preceding claims, wherein the yeast cell further has the characteristic:

25 VIII. capable of utilizing melibiose as the sole carbon source.

13. The yeast cell according to any one of the preceding claims, wherein the yeast cell further has the characteristic:

X. capable of sedimentation so that at the most 12 million, such as at the most 10 million cells/ml are in suspension when said yeast cell is added to a wort composition having a sugar content of at least 10° Plato and incubated for 4 days.

30

14. The yeast cell according to claim 13, wherein the number of cells in suspension per ml is at the most 80%, such as at the most 70%, for example at the most 60%, such as

at the most 50%, for example at the most 40% of the starting number of cells per ml after 5 days incubation under conditions allowing growth of said cells.

5 15. The yeast cell according to any one of the preceding claims, wherein the yeast cell further has characteristic:

IX. capable of utilizing one or more disaccharides and/or trisaccharides in addition to isomaltose, panose, and/or melibiose.

10 16. The yeast cell according to claim 15, wherein the disaccharide is selected from the group consisting of kojibiose, nigerose, sucrose, turanose, leucrose, and palatinose.

17. The yeast cell according to claim 15, wherein the disaccharide is kojibiose.

15 18. The yeast cell according to claim 15, wherein the disaccharide is maltulose.

19. The yeast cell according to any one of the preceding claims, wherein the yeast cell furthermore has characteristic

20 XI: capable of fermenting wort with a time of primary fermentation of at the most 4 days, for example at the most 3 days.

20. The yeast cell according to any one of the preceding claims, wherein the yeast cell has genotype:

25 I. comprising a gene encoding DAL5.

30 21. The yeast cell according to claim 20, wherein the genotype I is that said yeast cell comprises at least one allelic gene encoding DAL5, wherein the allelic gene encoding DAL5 encodes DAL5 selected from the group consisting of DAL5 of SEQ ID NO:6, DAL5 of SEQ ID NO:39, DAL5 of SEQ ID NO:40 and functional homologues thereof sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity with any of the aforementioned.

22. The yeast cell according to any one of the preceding claims, wherein the yeast cell has genotype:

II. comprising at least 2 allelic genes encoding PTR2.

5

23. The yeast cell according to claim 22, wherein genotype II is said yeast cell comprising at least two allelic genes encoding PTR2 individually selected from the group consisting of genes encoding PTR2 of SEQ ID NO:7, PTR2 of SEQ ID:8, PRT2 of SEQ ID NO:9, PRT2 comprising SEQ ID NO:37, PRT2 of SEQ ID NO:38, PRT2 of SEQ ID NO:43, PRT2 of SEQ ID NO:44 and functional homologues of each of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

10

15

24. The yeast cell according to any one of the preceding claims, wherein the yeast cell has the genotype:

III. comprising a gene encoding UBR1.

20

25. The yeast cell according to claim 24, wherein genotype III is said yeast cell comprising at least two allelic genes encoding UBR1 individually selected from the group consisting of UBR1 comprising SEQ ID NO:10, UBR1 of SEQ ID NO:11, UBR1 comprising SEQ ID NO:41, UBR1 of SEQ ID NO:42, UBR1 comprising SEQ ID NO:45 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

25

26. The yeast cell according to any one of the preceding claims, wherein the yeast cell has the genotype:

IV. comprising at least 3 genes, such as at least 4 genes encoding IMA1p.

30

27. A yeast cell having the genotype:

IV. comprising at least 3 genes, such as at least 4 genes encoding IMA1p.

28. The yeast cell according to any one of claims 26 and 27, wherein IMA1p is selected from the group consisting of IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15, IMA1p of SEQ ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:23, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and functional homologues of any of the
5 aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.
29. The yeast cell according to any one of claims 26 to 28, wherein the genotype IV
10 is that the yeast cell comprises at least 5 genes encoding IMA1p, wherein said genes individually are selected from the group consisting of genes encoding IMA1p of SEQ ID NO:1, IMA1p of SEQ ID NO:2, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO:4, IMA1p of SEQ ID NO:5, IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15,
15 IMA1p of SEQ ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:23, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and IMA1p of SEQ ID NO:33 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.
- 20
30. The yeast cell according to any one of claims 26 to 29, wherein the genotype IV is that said yeast cell comprises at least 3 short alleles of IMA1 and at least 2 long alleles of IMA1, wherein
- a) said 3 short alleles of IMA1 individually are genes encoding IMA1p selected
25 from the group consisting of of IMA1p of SEQ ID NO:12, IMA1p of SEQ ID NO:13, IMA1p of SEQ ID NO:1, IMA1p of SEQ ID NO:2, IMA1p of SEQ ID NO:3, IMA1p of SEQ ID NO: 4, IMA1p of SEQ ID NO: 5, IMA1p of SEQ ID NO:33 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such
30 as at least 98% sequence identity therewith; and
- b) said 2 long alleles of IMA1 individually are genes encoding IMA1p selected from the group consisting of IMA1p of SEQ ID NO:14, IMA1p of SEQ ID NO:15, IMA1p of SEQ ID NO:21, IMA1p of SEQ ID NO:22, IMA1p of SEQ ID NO:23, IMA1p of SEQ ID NO:24, IMA1p of SEQ ID NO:25 and functional

homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

5 31. The yeast cell according to any one of the preceding claims, wherein the yeast cell has the genotype

 V. Comprising a gene encoding IMA5p, preferably at least two allelic genes encoding IMA5p.

10 32. The yeast cell according to claim 31, wherein IMA5p is selected from the group consisting of IMA5p of SEQ ID NO:16, IMA5p of SEQ ID NO:17, IMA5p of SEQ ID NO:34, IMA5p of SEQ ID NO:35, IMA5p of SEQ ID NO:36 and functional homologues of any of the aforementioned sharing at least 80%, preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence
15 identity therewith.

33. The yeast cell according to any one of the preceding claims, wherein the yeast cell has the genotype;

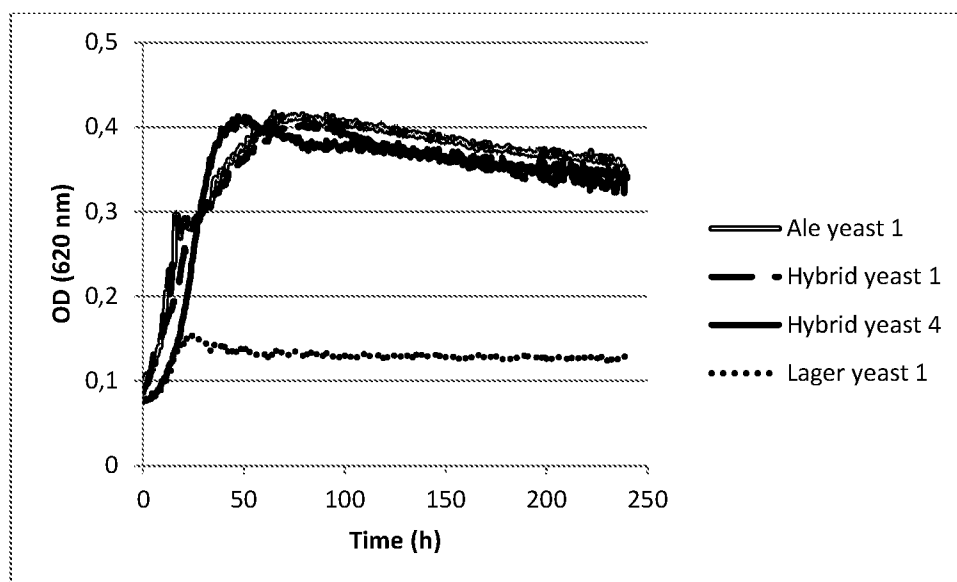
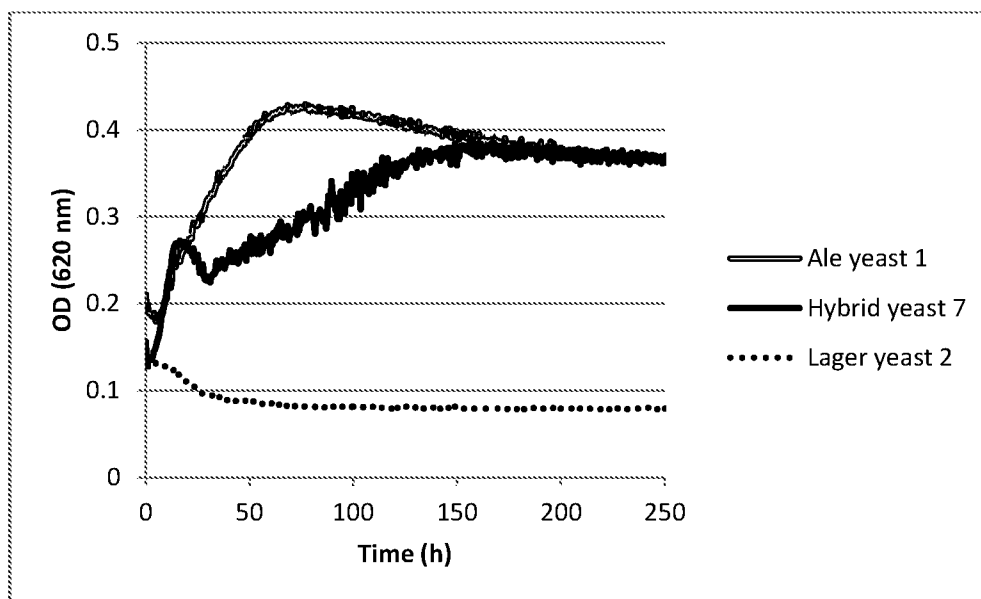
 VI. comprising at least 3 genes encoding AGT1 selected from the group
20 consisting of AGT1 of SEQ ID NO:18, AGT1 of SEQ ID NO:19, AGT1 of SEQ ID NO:20, AGT1 of SEQ ID NO:26, AGT1 of SEQ ID NO:27, AGT1 of SEQ ID NO:28, AGT1 of SEQ ID NO:29, AGT1 of SEQ ID NO:30, AGT1 of SEQ ID NO:31, AGT1 of SEQ ID NO:32 and functional homologues of any of the aforementioned sharing at least 80%,
25 preferably at least 90%, yet more preferably at least 95%, such as at least 98% sequence identity therewith.

34. A method for producing a beverage, said method comprising the steps of

30 a. Providing a starting liquid
 b. Providing a yeast cell according to any one of claims 1 to 34
 c. Fermenting said starting liquid with said yeast cell,

thereby producing a beverage.

35. The method according to claim 34, wherein the starting liquid comprises an aqueous extract of barley and/or malt.

A)**B)****Fig. 1**

C)

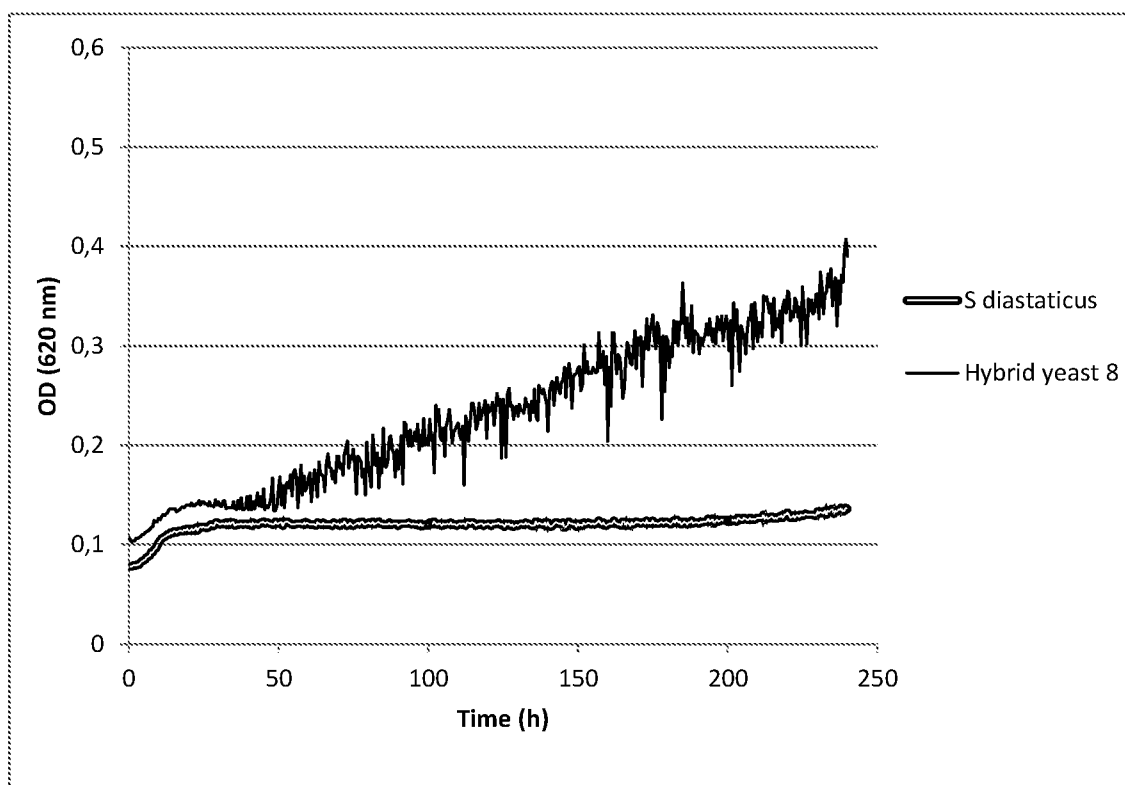
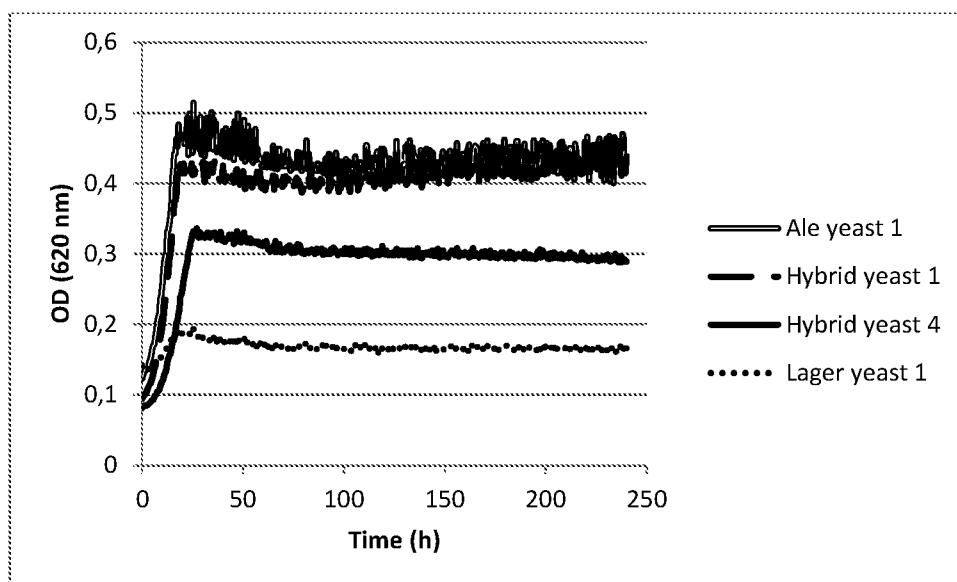


Fig. 1 - continued

A)



B)

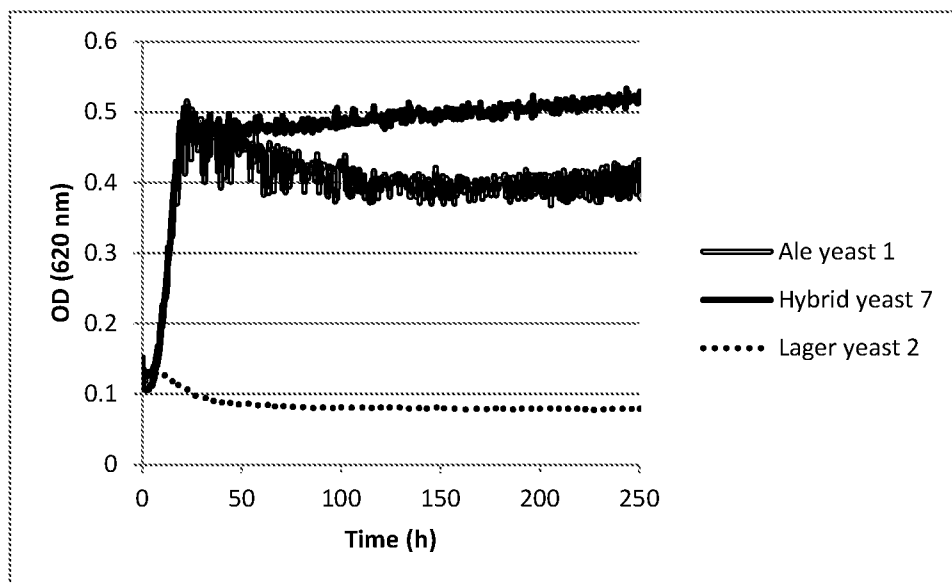


Fig. 2

C)

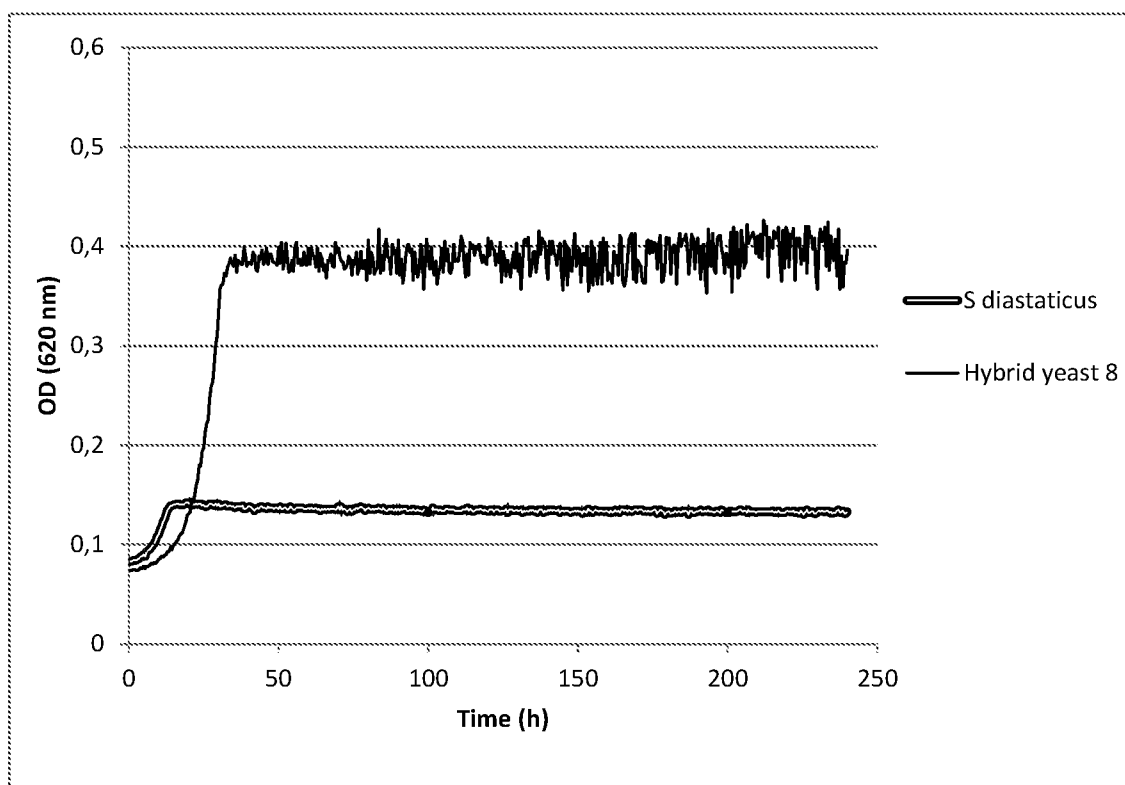
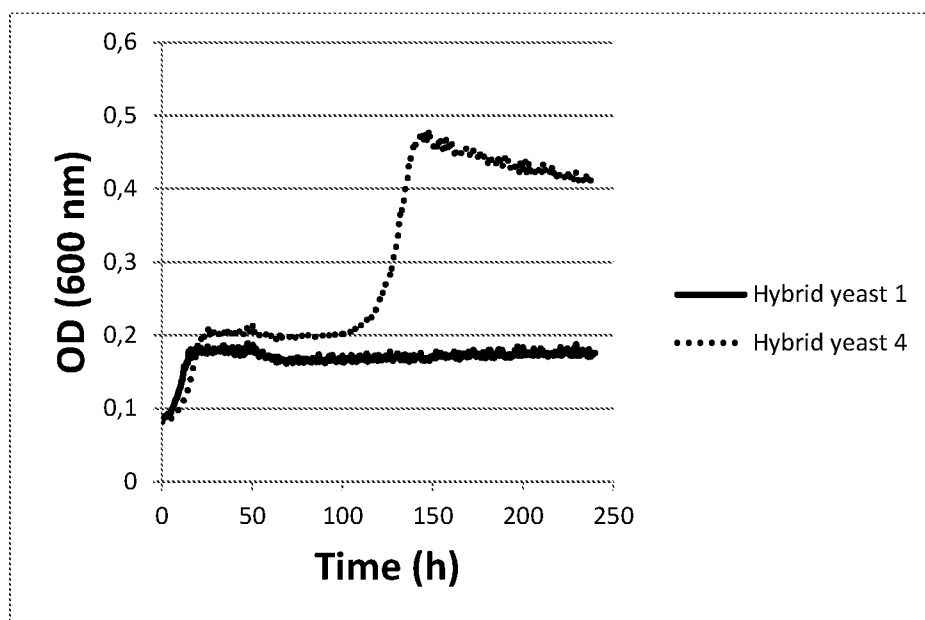


Figure 2 - continued

A)



B)

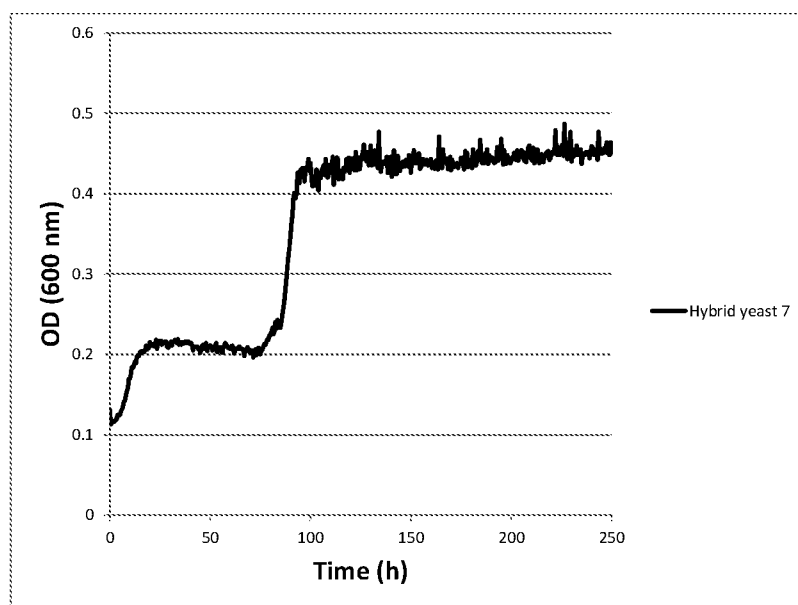


Fig. 3

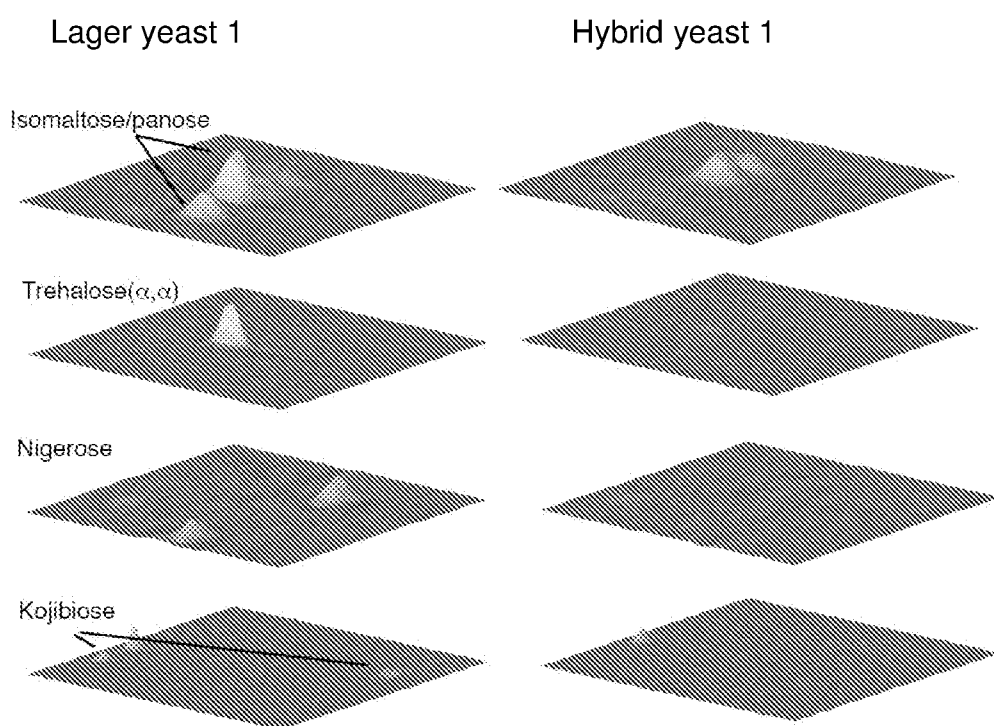


Fig. 4

ScDAL5_Ale_1	MSADASTNSNASLDEKNLNI TSEAEI KNEDVTAEPVLSTVLSPNGKI VYI SDKYDEAMKLAEAEKEI EVTPPEEDRKL RWM	80
nonSc_DAL5_Lager_1	MSGGA STNSNASIDEKNLNI TSEAEI KNEDVTAEPVLSTVLSPNGKVYI SDKYDEAMKLAEAEKEI EVTPPEEDRKL RWM	80
Sc_DAL5_Hybrid_1	MSADASTNSNASLDEKNLNI TSEAEI KNEDVTAEPVLSTVLSPNGKI VYI SDKYDEAMKLAEAEKEI EVTPPEEDRKL RWM	80
ScDAL5_Ale_1	I DYCMEPLMCI L YAVQFMDKI STSSAAVMGLRTDLKMHGDQYSWTSAFYFGYLFMNLGPVQFI FQRTSHMSKMLAVFI V	160
nonSc_DAL5_Lager_1	I DYCMEPLMCI L YAVQFMDKI STSSAAVMGLRTDLKMHGDQYSWTSAFYFGYLFMNLGPVQFI FQRTSHMSKMLAVFI V	160
Sc_DAL5_Hybrid_1	I DYCMEPLMCI L YAVQFMDKI STSSAAVMGLRTDLKMHGDQYSWTSAFYFGYLFMNLGPVQFI FQRTSHMSKMLAVFI V	160
ScDAL5_Ale_1	I WGM L LALHAAPT VKYPSFI VLRVLLGCAESVTPCFTI I TAQYWKTEEQFTRVSI WFGMNGLSI LI NAI AYGVYI HQD	240
nonSc_DAL5_Lager_1	I WGM L LALHAAPT VKYPSFI VLRVLLGCAESVTPCFTI I TAQYWKTEEQFTRVSI WFGMNGLSI LI NAI AYGVYI HQD	240
Sc_DAL5_Hybrid_1	I WGM L LALHAAPT VKYPSFI VLRVLLGCAESVTPCFTI I TAQYWKTEEQFTRVSI WFGMNGLSI LI NAI AYGVYI HQD	240
ScDAL5_Ale_1	SYAI KGMRTL FVI TGVI TI FI GI LI FLW PDDPSKARFLSKREKL MVVQRI RSNQOGF GNHEI KKYQI I EALKDVRTWL Y	320
nonSc_DAL5_Lager_1	SYAI KGMRTL FVI TGVI TI FI GI LI FLW PDDPSKARFLSKREKL MVVQRI RSNQOGF GNHEI KKYQI I EALKDVRTWL Y	320
Sc_DAL5_Hybrid_1	SYAI KGMRTL FVI TGVI TI FI GI LI FLW PDDPSKARFLSKREKL MVVQRI RSNQOGF GNHEI KKYQI I EALKDVRTWL Y	320
ScDAL5_Ale_1	FLFTVSSNI PNGGI SSFMSI LLNSDFGYSSKETLLMGLPTGAVEL VGCP LFGI LAVYAANKKI PFWKYKLSWAI FAAYLA	400
nonSc_DAL5_Lager_1	FLFTVSSNI PNGGI SSFMSI LLNSDFGYSSKETLLMGLPTGAVEL VGCP LFGI LAVYAANKKI PFWKYKLSWAI FAAYLA	400
Sc_DAL5_Hybrid_1	FLFTVSSNI PNGGI SSFMSI LLNSDFGYSSKETLLMGLPTGAVEL VGCP LFGI LAVYAANKKI PFWKYKLSWAI FAAYLA	400
ScDAL5_Ale_1	LI ASCM LGFATNSKKARLAGAYL W I SPVSFI CVLSNI SANSSGYSKKWTVSSI NLVAYAAANLAGPOTFI AKQAPKYHG	480
nonSc_DAL5_Lager_1	LI ASCM LGFATNSKKARLAGAYL W I SPVSFI CVLSNI SANSSGYSKKWTVSSI NLVAYAAANLAGPOTFI AKQAPKYHG	480
Sc_DAL5_Hybrid_1	LI ASCM LGFATNSKKARLAGAYL W I SPVSFI CVLSNI SANSSGYSKKWTVSSI NLVAYAAANLAGPOTFI AKQAPKYHG	480
ScDAL5_Ale_1	AKVAMVVCYAVMI VLLSI LLI VNLRENKR RDKI AAERGFP EETENLEFSDLTDFENPNFRYTL	544
nonSc_DAL5_Lager_1	AKVAMVVCYAVMI VLLSI LLI VNLRENKR RDKI AAERGFP EETENLEFSDLTDFENPNFRYTL	544
Sc_DAL5_Hybrid_1	AKVAMVVCYAVMI VLLSI LLI VNLRENKR RDKI AAERGFP EETENLEFSDLTDFENPNFRYTL	544

Fig. 5

Sc_UBR1_Ale_1	FKEFCCKVEGGVLIWQVQKSNLTKSYSISFKQGLYTVETL	SKVHPNIPLRPEIISLLTLCKLFNGAWKIKRKEGEHV	679		
Sc_UBR1_Lager_1	FKEFCCKVEGGVLIWQVQKSNLTKSYSISFKQGLYTVETL	SKVHPNIPLRPEIISLLTLCKLFNGAWKIKRKEGEHV	679		
Sc_UBR1_Hybrid_1	FKEFCCKVEGGVLIWQVQKSNLTKSYSISFKQGLYTVETL	SKVHPNIPLRPEIISLLTLCKLFNGAWKIKRKEGEHV	80		
Sc_UBR1_Ale_1	LHEDQNFISYLEYTTISYISIQTAEKVSEKSKDSIDSKLFLNARI	SSFLGNRSLTYKLIYDSHEVIKFSVSHERVAFM	759		
Sc_UBR1_Lager_1	LHEDQNFISYLEYTTISYISIQTAEKVSEKSKDSIDSKLFLNARI	SSFLGNRSLTYKLIYDSHEVIKFSVSHERVAFM	759		
Sc_UBR1_Hybrid_1	LHEDQNFISYLEYTTISYISIQTAEKVSEKSKDSIDSKLFLNARI	SSFLGNRSLTYKLIYDSHEVIKFSVSHERVAFM	160		
Sc_UBR1_Ale_1	NPLQTMLSFLIEKVSUKDAYEALEDCSDFLKI	SDFSLSRYVLCSQIDVGFWWRNGMSVLHQASYKKNP	ELGSYSRDIHL839		
Sc_UBR1_Lager_1	NPLQTMLSFLIEKVSUKDAYEALEDCSDFLKI	SDFSLSRYVLCSQIDVGFWWRNGMSVLHQASYKKNP	ELGSYSRDIHL839		
Sc_UBR1_Hybrid_1	NPLQTMLSFLIEKVSUKDAYEALEDCSDFLKI	SDFSLSRYVLCSQIDVGFWWRNGMSVLHQASYKKNP	ELGSYSRDIHL240		
Sc_UBR1_Ale_1	NQLALWERDDIPRIYNILDRWELLDWFTGEVDYQHTVYEDKISFI	QQFIAFIYQXISL	901		
Sc_UBR1_Lager_1	NQLALWERDDIPRIYNILDRWELLDWFTGEVDYQHTVYEDKISFI	QQFIAFIYQILTERQYFKTFSSLKDRRMDQIK	919		
Sc_UBR1_Hybrid_1	NQLALWERDDIPRIYNILDRWELLDWFTGEVDYQHTVYEDKISFI	QQFIAFIYQILTERQYFKTFSSLKDRRMDQIK	320		
Sc_UBR1_Ale_1	NSIIYNLYMKPLSYSKLLRSVPDYL	TEDTEFDEALEEVSFVEPKGADNGVFKLKASLYAKVDP	KLNL	ENEFESS	901
Sc_UBR1_Lager_1	NSIIYNLYMKPLSYSKLLRSVPDYL	TEDTEFDEALEEVSFVEPKGADNGVFKLKASLYAKVDP	KLNL	ENEFESS	998
Sc_UBR1_Hybrid_1	NSIIYNLYMKPLSYSKLLRSVPDYL	TEDTEFDEALEEVSFVEPKGADNGVFKLKASLYAKVDP	KLNL	ENEFESS	399

Fig. 6

[illegible]

Fig. 7

IWA1_Sc_albe_short_Ab_1
IWA1_Sc_albe_short_A_Hybrd_1
IWA1_Sc_albe_short_B_Hybrd_1

IWA1_Sc_albe_short_Ab_1
IWA1_Sc_albe_short_A_Hybrd_1
IWA1_Sc_albe_short_B_Hybrd_1

IWA1_Sc_albe_short_Ab_1
IWA1_Sc_albe_short_A_Hybrd_1
IWA1_Sc_albe_short_B_Hybrd_1

IWA1_Sc_albe_short_Ab_1
IWA1_Sc_albe_short_A_Hybrd_1
IWA1_Sc_albe_short_B_Hybrd_1

IWA1_Sc_albe_short_Ab_1
IWA1_Sc_albe_short_A_Hybrd_1
IWA1_Sc_albe_short_B_Hybrd_1

10 20 30 40 50 60 70 80
MGYDI ANYEKVMPTYGINEDCFAL I EKT HKL G M K F I T D L V I N H C S S E H E W F K E S R S S K T N P K R D W F F W R P P K G Y D A E G K P 80
MGYDI ANYEKVMPTYGINEDCFAL I EKT HKL G M K F I T D L V I N H C S S E H E W F K E S R S S K T N P K R D W F F W R P P K G Y D A E G K P 80
MGYDI ANYEKVMPTYGINEDCFAL I EKT HKL G M K F I T D L V I N H C S S E H E W F K E S R S S K T N P K R D W F F W R P P K G Y D A E G K P 80

90 100 110 120 130 140 150 160
I P P N N M K S Y F G G S A W F D E K T O E F Y L R L F C S T O P D L N W E N E D C R K A I Y E S A V G Y W L D H G V D G F R I D V G S L Y S K V V G L P D A 160
I P P N N M K S Y F G G S A W F D E K T O E F Y L R L F C S T O P D L N W E N E D C R K A I Y E S A V G Y W L D H G V D G F R I D V G S L Y S K V V G L P D A 160
I P P N N M K S Y F G G S A W F D E K T O E F Y L R L F C S T O P D L N W E N E D C R K A I Y E S A V G Y W L D H G V D G F R I D V G S L Y S K V V G L P D A 160

170 180 190 200 210 220 230 240
P V D K N S T W O S S D P Y T L N G P R I H E F H O E M N O F I R N B Y K D G R E I M T V G E M O H A S D E T K L Y T S A S R H E L S E L F N F S H T D V G 240
P V D K N S T W O S S D P Y T L N G P R I H E F H O E M N O F I R N B Y K D G R E I M T V G E M O H A S D E T K L Y T S A S R H E L S E L F N F S H T D V G 240
P V D K N S T W O S S D P Y T L N G P R I H E F H O E M N O F I R N B Y K D G R E I M T V G E M O H A S D E T K L Y T S A S R H E L S E L F N F S H T D V G 240

250 260 270 280 290 300 310 320
T S P L F R Y N L V P F E L K D W K I A L A E L F R E I N G T D C W S T I Y L E N H D O P R S I T R F G D S P K N R V I S G K L L S Y L L S A L T G T L Y Y 320
T S P L F R Y N L V P F E L K D W K I A L A E L F R E I N G T D C W S T I Y L E N H D O P R S I T R F G D S P K N R V I S G K L L S Y L L S A L T G T L Y Y 320
T S P L F R Y N L V P F E L K D W K I A L A E L F R E I N G T D C W S T I Y L E N H D O P R S I T R F G D S P K N R V I S G K L L S Y L L S A L T G T L Y Y 320

330 340 350 360 370 380 390 400
O G O E L G O I N F K N W S V E K Y E D V E I R N N Y R L I K E E C G E N S E E M K K L E G I A L V S R D H A R T P M P W T P N E P N A G F S G P N T K P W F 400
O G O E L G O I N F K N W S V E K Y E D V E I R N N Y R L I K E E C G E N S E E M K K L E G I A L V S R D H A R T P M P W T P N E P N A G F S G P N T K P W F 400
O G O E L G O I N F K N W S V E K Y E D V E I R N N Y R L I K E E C G E N S E E M K K L E G I A L V S R D H A R T P M P W T P N E P N A G F S G P N T K P W F 400

410 420 430 440 450 460 470 480
Y L N E S F R O G I N V E E E O K N S D S Y L A F W K K A L E F R K N H K D I A V Y G F D F K F I D L D N K K L F S F T K R Y N N K T L F A A L N F S S D A T D 480
Y L N E S F R O G I N V E E E O K N S D S Y L A F W K K A L E F R K N H K D I A V Y G F D F K F I D L D N K K L F S F T K R Y N N K T L F A A L N F S S D A T D 480
Y L N E S F R O G I N V E E E O K N S D S Y L A F W K K A L E F R K N H K D I A V Y G F D F K F I D L D N K K L F S F T K R Y N N K T L F A A L N F S S D A T D 480

490 500 510 520
F K I P N D G S S F K L E F G N Y P K N E V D A S S R T L K P W E G R I H I N E 521
F K I P N D G S S F K L E F G N Y P K N E V D A S S R T L K P W E G R I H I N E 521
F K I P N D G S S F K L E F G N Y P K N E V D A S S R T L K P W E G R I H I N E 521

Fig. 8

SUBSTITUTE SHEET (RULE 26)

[illegible]

Fig. 9B

ScIMAS_Aet1	MTI I HNP	80
ScIMAS_Lager1	MTI I HNP	80
ScIMAS_Hybrid1_p11	MTI I HNP	80
non-ScIMAS_Lager1	MTI I HNP	80
non-ScIMAS_Hybrid1	MTI I HNP	80
ScIMAS_Aet1	TNEDC	160
ScIMAS_Lager1	TNEDC	160
ScIMAS_Hybrid1_p11	TNEDC	160
non-ScIMAS_Lager1	TNEDC	160
non-ScIMAS_Hybrid1	TNEDC	160
ScIMAS_Aet1	YDEK	240
ScIMAS_Lager1	YDEK	240
ScIMAS_Hybrid1_p11	YDEK	240
non-ScIMAS_Lager1	YDEK	240
non-ScIMAS_Hybrid1	YDEK	240
ScIMAS_Aet1	I NGR	320
ScIMAS_Lager1	I NGR	320
ScIMAS_Hybrid1_p11	I NGR	320
non-ScIMAS_Lager1	I NGR	320
non-ScIMAS_Hybrid1	I NGR	320
ScIMAS_Aet1	FKL	400
ScIMAS_Lager1	FKL	400
ScIMAS_Hybrid1_p11	FKL	400
non-ScIMAS_Lager1	FKL	400
non-ScIMAS_Hybrid1	FKL	400
ScIMAS_Aet1	QI KC	480
ScIMAS_Lager1	QI KC	480
ScIMAS_Hybrid1_p11	QI KC	480
non-ScIMAS_Lager1	QI KC	480
non-ScIMAS_Hybrid1	QI KC	480
ScIMAS_Aet1	DKNS	560
ScIMAS_Lager1	DKNS	560
ScIMAS_Hybrid1_p11	DKNS	560
non-ScIMAS_Lager1	DKNS	560
non-ScIMAS_Hybrid1	DKNS	560
ScIMAS_Aet1	ANSG	582
ScIMAS_Lager1	ANSG	582
ScIMAS_Hybrid1_p11	ANSG	582
non-ScIMAS_Lager1	ANSG	582
non-ScIMAS_Hybrid1	ANSG	582

Fig. 10

Sc_AGT1_Lager1	MKNI I SLVSKKKAASKNEDKNI SESSRDI VNOQEVNTENFEEGKDSAFELDHEFTINSACL GDSDEDNENVI NEINA	80
Sc_AGT1_Aet1	MKNI I SLVSKKKAASKNEDKNI SESSRDI VNOQEVNTENFEEGKDSAFELDHEFTINSACL GDSDEDNENVI NETNT	80
Sc_AGT1_Hybrd1_p37	MKNI I SLVSKKKAASKNEDKNI SESSRDI VNOQEVNTENFEEGKDSAFELDHEFTINSACL GDSDEDNENVI NETNT	80
Sc_AGT1_Hybrd1_p38	MKNI I SLVSKKKAASKNEDKNI SESSRDI VNOQEVNTENFEEGKDSAFELDHEFTINSACL GDSDEDNENVI NETNT	80
Sc_AGT1_Hybrd1_p39	MKNI I SLVSKKKAASKNEDKNI SESSRDI VNOQEVNTENFEEGKDSAFELDHEFTINSACL GDSDEDNENVI NETNT	80
Sc_AGT1_Lager1	TDDANEANSEEEKSMTLKQALLYPKALWSI LVSTL VMEGYDTALLNAL YAL PYFQKFGTLNGEYSYEI TSQWQI GLN	160
Sc_AGT1_Aet1	TDDANEANSEEEKSMTLKQALLYPKALWSI LVSTL VMEGYDTALLNAL YAL PYFQKFGTLNGEYSYEI TSQWQI GLN	160
Sc_AGT1_Hybrd1_p37	TDDANEANSEEEKSMTLKQALLYPKALWSI LVSTL VMEGYDTALLNAL YAL PYFQKFGTLNGEYSYEI TSQWQI GLN	160
Sc_AGT1_Hybrd1_p38	TDDANEANSEEEKSMTLKQALLYPKALWSI LVSTL VMEGYDTALLNAL YAL PYFQKFGTLNGEYSYEI TSQWQI GLN	160
Sc_AGT1_Hybrd1_p39	TDDANEANSEEEKSMTLKQALLYPKALWSI LVSTL VMEGYDTALLNAL YAL PYFQKFGTLNGEYSYEI TSQWQI GLN	160
Sc_AGT1_Lager1	MCVQOGEI I GLQI TPYMVEFMGNRYTMTALGLTAYVEI LYYCKSLAMI AVGOVLSAMPWGCFOGL TVTYASEVCPAL	240
Sc_AGT1_Aet1	MCVQOGEI I GLQI TPYMVEFMGNRYTMTALGLTAYVEI LYYCKSLAMI AVGOVLSAMPWGCFOGL TVTYASEVCPAL	240
Sc_AGT1_Hybrd1_p37	MCVQOGEI I GLQI TPYMVEFMGNRYTMTALGLTAYVEI LYYCKSLAMI AVGOVLSAMPWGCFOGL TVTYASEVCPAL	240
Sc_AGT1_Hybrd1_p38	MCVQOGEI I GLQI TPYMVEFMGNRYTMTALGLTAYVEI LYYCKSLAMI AVGOVLSAMPWGCFOGL TVTYASEVCPAL	240
Sc_AGT1_Hybrd1_p39	MCVQOGEI I GLQI TPYMVEFMGNRYTMTALGLTAYVEI LYYCKSLAMI AVGOVLSAMPWGCFOGL TVTYASEVCPAL	240
Sc_AGT1_Lager1	RYVMTSYSNI CWMFGQI FASGI MKNSQENL GNSDLGYKLPFALQMWAPAPLM GIFFAPESPWML VRKDRVAEARKSLSR	320
Sc_AGT1_Aet1	RYVMTSYSNI CWMFGQI FASGI MKNSQENL GNSDLGYKLPFALQMWAPAPLM GIFFAPESPWML VRKDRVAEARKSLSR	320
Sc_AGT1_Hybrd1_p37	RYVMTSYSNI CWMFGQI FASGI MKNSQENL GNSDLGYKLPFALQMWAPAPLM GIFFAPESPWML VRKDRVAEARKSLSR	320
Sc_AGT1_Hybrd1_p38	RYVMTSYSNI CWMFGQI FASGI MKNSQENL GNSDLGYKLPFALQMWAPAPLM GIFFAPESPWML VRKDRVAEARKSLSR	320
Sc_AGT1_Hybrd1_p39	RYVMTSYSNI CWMFGQI FASGI MKNSQENL GNSDLGYKLPFALQMWAPAPLM GIFFAPESPWML VRKDRVAEARKSLSR	320
Sc_AGT1_Lager1	ILSGKGAEKDI QIDLTCLKQI ELTI EKERLLASKSGSFEDCFKGVNGRRTLACL TWAQNTSGACLLGYSTYFF	394
Sc_AGT1_Aet1	ILSGKGAEKDI QIDLTCLKQI ELTI EKERLLASKSGSFEDCFKGVNGRRTLACL TWAQNTSGACLLGYSTYFFERAGMA	400
Sc_AGT1_Hybrd1_p37	ILSGKGAEKDI QIDLTCLKQI ELTI EKERLLASKSGSFEDCFKGVNGRRTLACL TWAQNTSGACLLGYSTYFFERAGMA	400
Sc_AGT1_Hybrd1_p38	ILSGKGAEKDI QIDLTCLKQI ELTI EKERLLASKSGSFEDCFKGVNGRRTLACL TWAQNTSGACLLGYSTYFFERAGMA	400
Sc_AGT1_Hybrd1_p39	ILSGKGAEKDI QIDLTCLKQI ELTI EKERLLASKSGSFEDCFKGVNGRRTLACL TWAQNTSGACLLGYSTYFFERAGMA	400
Sc_AGT1_Lager1	TDKAFTFSVI QYCLGLAGTLCSWI SGRVGRWTI LTYGLAFQMWCLFI I GGMGFGSGSGASNGAGGLLLAL SFFYNAGI G	394
Sc_AGT1_Aet1	TDKAFTFSVI QYCLGLAGTLCSWI SGRVGRWTI LTYGLAFQMWCLFI I GGMGFGSGSGASNGAGGLLLAL SFFYNAGI G	480
Sc_AGT1_Hybrd1_p37	TDKAFTFSVI QYCLGLAGTLCSWI SGRVGRWTI LTYGLAFQMWCLFI I GGMGFGSGSGASNGAGGLLLAL SFFYNAGI G	480
Sc_AGT1_Hybrd1_p38	TDKAFTFSVI QYCLGLAGTLCSWI SGRVGRWTI LTYGLAFQMWCLFI I GGMGFGSGSGASNGAGGLLLAL SFFYNAGI G	480
Sc_AGT1_Hybrd1_p39	TDKAFTFSVI QYCLGLAGTLCSWI SGRVGRWTI LTYGLAFQMWCLFI I GGMGFGSGSGASNGAGGLLLAL SFFYNAGI G	409
Sc_AGT1_Lager1	AVVYCI VTEI PSAELRTKTI VLARI CYNIMAVI NAILTPYML NVSDMWGAKTGLYWGGTAVTLAWMI I DLPETSGRTF	394
Sc_AGT1_Aet1	AVVYCI VTEI PSAELRTKTI VLARI CYNIMAVI NAILTPYML NVSDMWGAKTGLYWGGTAVTLAWMI I DLPETSGRTF	560
Sc_AGT1_Hybrd1_p37	AVVYCI VTEI PSAELRTKTI VLARI CYNIMAVI NAILTPYML NVSDMWGAKTGLYWGGTAVTLAWMI I DLPETSGRTF	560
Sc_AGT1_Hybrd1_p38	AVVYCI VTEI PSAELRTKTI VLARI CYNIMAVI NAILTPYML NVSDMWGAKTGLYWGGTAVTLAWMI I DLPETSGRTF	560
Sc_AGT1_Hybrd1_p39	AVVYCI VTEI PSAELRTKTI VLARI CYNIMAVI NAILTPYML NVSDMWGAKTGLYWGGTAVTLAWMI I DLPETSGRTF	409
Sc_AGT1_Lager1	SEI NELFNOGVPAARKFASTVVDPF GKGTQHDSDL DDESI SQSSSI KQREL NAADKC	394
Sc_AGT1_Aet1	SEI NELFNOGVPAARKFASTVVDPF GKGTQHDSDL DDESI SQSSSI KQREL NAADKC	616
Sc_AGT1_Hybrd1_p37	SEI NELFNOGVPAARKFASTVVDPF GKGTQHDSDL DDESI SQSSSI KQREL NAADKC	617
Sc_AGT1_Hybrd1_p38	SEI NELFNOGVPAARKFASTVVDPF GKGTQHDSDL DDESI SQSSSI KQREL NAADKC	617
Sc_AGT1_Hybrd1_p39	SEI NELFNOGVPAARKFASTVVDPF GKGTQHDSDL DDESI SQSSSI KQREL NAADKC	409

Decoration "": Shade (with solid black) residues that differ from Sc_AGT1_Aet1.

Fig. 11A

Sc_AGT1_Laeger_2	MKNITSLVSKKKKAAASKNEDKNISSESRDIVNQOEVTENTENFEEGKKDSAFELDLHLEFTNSAQLOGSDDEDNENINEUNATDANEANSE	90
Sc_AGT1_Ale_1	MKNITSLVSKKKKAAASKNEDKNISSESRDIVNQOEVTENTENFEEGKKDSAFELDLHLEFTNSAQLOGSDDEDNENINETNTDDANEANSE	90
Sc_AGT1_Hybrid_4_p1#2	MKNITSLVSKKKKAAASKNEDKNISSESRDIVNQOEVTENTENFEEGKKDSAFELDLHLEFTNSAQLOGSDDEDNENINEUNATDANEANSE	90
Sc_AGT1_Hybrid_7_p1#11	MKNITSLVSKKKKAAASKNEDKNISSESRDIVNQOEVTENTENFEEGKKDSAFELDLHLEFTNSAQLOGSDDEDNENINEUNATDANEANSE	90
Sc_AGT1_Hybrid_7_p1#12	MKNITSLVSKKKKAAASKNEDKNISSESRDIVNQOEVTENTENFEEGKKDSAFELDLHLEFTNSAQLOGSDDEDNENINETNTDDANEANSE	90
Sc_AGT1_Laeger_2	EKSMTLKQALILPKAALWMSILVSTTLVMEGYDTALINALYALPVFORKEGTLNGEGSYEITTSQWQIGLNMCVOCGEIIGLQITPMVEEF	180
Sc_AGT1_Ale_1	EKSMTLKQALILPKAALWMSILVSTTLVMEGYDTALINALYALPVFORKEGTLNGEGSYEITTSQWQIGLNMCVOCGEIIGLQITPMVEEF	180
Sc_AGT1_Hybrid_4_p1#2	EKSMTLKQALILPKAALWMSILVSTTLVMEGYDTALINALYALPVFORKEGTLNGEGSYEITTSQWQIGLNMCVOCGEIIGLQITPMVEEF	180
Sc_AGT1_Hybrid_7_p1#11	EKSMTLKQALILPKAALWMSILVSTTLVMEGYDTALINALYALPVFORKEGTLNGEGSYEITTSQWQIGLNMCVOCGEIIGLQITPMVEEF	180
Sc_AGT1_Hybrid_7_p1#12	EKSMTLKQALILPKAALWMSILVSTTLVMEGYDTALINALYALPVFORKEGTLNGEGSYEITTSQWQIGLNMCVOCGEIIGLQITPMVEEF	180
Sc_AGT1_Laeger_2	MGNRYTMITALGLLITAYFFILYYCKSLAMIAVGQVLSAMPWGCFOGLITVYASEVCPALALRYMTSYSNICWLEFGQIFASGIMKNSQENL	270
Sc_AGT1_Ale_1	MGNRYTMITALGLLITAYFFILYYCKSLAMIAVGQVLSAMPWGCFOGLITVYASEVCPALALRYMTSYSNICWLEFGQIFASGIMKNSQENL	270
Sc_AGT1_Hybrid_4_p1#2	MGNRYTMITALGLLITAYFFILYYCKSLAMIAVGQVLSAMPWGCFOGLITVYASEVCPALALRYMTSYSNICWLEFGQIFASGIMKNSQENL	270
Sc_AGT1_Hybrid_7_p1#11	MGNRYTMITALGLLITAYFFILYYCKSLAMIAVGQVLSAMPWGCFOGLITVYASEVCPALALRYMTSYSNICWLEFGQIFASGIMKNSQENL	270
Sc_AGT1_Hybrid_7_p1#12	MGNRYTMITALGLLITAYFFILYYCKSLAMIAVGQVLSAMPWGCFOGLITVYASEVCPALALRYMTSYSNICWLEFGQIFASGIMKNSQENL	270
Sc_AGT1_Laeger_2	GNSDLDYKLPALQWIMPAPLMIGTFFAPESPWWLVKRDVAAEARKSLSRILSGKGAEKDIOQDITLKQIELTTEKERLLASKSGSFFDC	360
Sc_AGT1_Ale_1	GNSDLGYKLPALQWIMPAPLMIGTFFAPESPWWLVKRDVAAEARKSLSRILSGKGAEKDIOQDITLKQIELTTEKERLLASKSGSFFDC	360
Sc_AGT1_Hybrid_4_p1#2	GNSDLDYKLPALQWIMPAPLMIGTFFAPESPWWLVKRDVAAEARKSLSRILSGKGAEKDIOQDITLKQIELTTEKERLLASKSGSFFDC	360
Sc_AGT1_Hybrid_7_p1#11	GNSDLGYKLPALQWIMPAPLMIGTFFAPESPWWLVKRDVAAEARKSLSRILSGKGAEKDIOQDITLKQIELTTEKERLLASKSGSFFDC	360
Sc_AGT1_Hybrid_7_p1#12	GNSDLGYKLPALQWIMPAPLMIGTFFAPESPWWLVKRDVAAEARKSLSRILSGKGAEKDIOQDITLKQIELTTEKERLLASKSGSFFDC	360
Sc_AGT1_Laeger_2	FKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKAGMATDKAFTFSVIQYCLGLAGTLCSMVISGRVGRWTTILTYGLAFQMWCLFTIG	395
Sc_AGT1_Ale_1	FKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKAGMATDKAFTFSVIQYCLGLAGTLCSMVISGRVGRWTTILTYGLAFQMWCLFTIG	395
Sc_AGT1_Hybrid_4_p1#2	FKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKAGMATDKAFTFSVIQYCLGLAGTLCSMVISGRVGRWTTILTYGLAFQMWCLFTIG	395
Sc_AGT1_Hybrid_7_p1#11	FKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKAGMATDKAFTFSVIQYCLGLAGTLCSMVISGRVGRWTTILTYGLAFQMWCLFTIG	395
Sc_AGT1_Hybrid_7_p1#12	FKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKGVNGRRTRLACLHWAQNTSGACLLGYSTYFFFKAGMATDKAFTFSVIQYCLGLAGTLCSMVISGRVGRWTTILTYGLAFQMWCLFTIG	395
Sc_AGT1_Laeger_2	GMGFGSGSGASNGAGGCLLLALSFFFNAGIGAVVYCIATIEIPSAELRTKTIIVLARICYNIMAVINAILTPYMLNVSDWNWGAKTGLYWGGE	540
Sc_AGT1_Ale_1	GMGFGSGSGASNGAGGCLLLALSFFFNAGIGAVVYCIATIEIPSAELRTKTIIVLARICYNIMAVINAILTPYMLNVSDWNWGAKTGLYWGGE	540
Sc_AGT1_Hybrid_4_p1#2	GMGFGSGSGASNGAGGCLLLALSFFFNAGIGAVVYCIATIEIPSAELRTKTIIVLARICYNIMAVINAILTPYMLNVSDWNWGAKTGLYWGGE	540
Sc_AGT1_Hybrid_7_p1#11	GMGFGSGSGASNGAGGCLLLALSFFFNAGIGAVVYCIATIEIPSAELRTKTIIVLARICYNIMAVINAILTPYMLNVSDWNWGAKTGLYWGGE	540
Sc_AGT1_Hybrid_7_p1#12	GMGFGSGSGASNGAGGCLLLALSFFFNAGIGAVVYCIATIEIPSAELRTKTIIVLARICYNIMAVINAILTPYMLNVSDWNWGAKTGLYWGGE	540
Sc_AGT1_Laeger_2	TAVTLAWVIIDLPETSGRTFSEIMELFNQGVPAARKPASTVDPFGKGKGTQHDSDLDESISQSSSIKQORELNADKQ	617
Sc_AGT1_Ale_1	TAVTLAWVIIDLPETSGRTFSEIMELFNQGVPAARKPASTVDPFGKGKGTQHDSDLDESISQSSSIKQORELNADKQ	617
Sc_AGT1_Hybrid_4_p1#2	TAVTLAWVIIDLPETSGRTFSEIMELFNQGVPAARKPASTVDPFGKGKGTQHDSDLDESISQSSSIKQORELNADKQ	617
Sc_AGT1_Hybrid_7_p1#11	TAVTLAWVIIDLPETSGRTFSEIMELFNQGVPAARKPASTVDPFGKGKGTQHDSDLDESISQSSSIKQORELNADKQ	617
Sc_AGT1_Hybrid_7_p1#12	TAVTLAWVIIDLPETSGRTFSEIMELFNQGVPAARKPASTVDPFGKGKGTQHDSDLDESISQSSSIKQORELNADKQ	617

Fig. 11B

Non-Sc_AGT1_Lager1	MKNI LSLVGRKENTPEDVTANLADTSSTVMQAKDLVI	EDFEERKKNDAFELNHL	ELTTNATQ	SDSDEKENV	RVAEA	80				
Non-Sc_AGT1_Hybrid1	MKNI LSLVGRKENTPEDVTANLADTSSTVMQAKDLVI	EDFEERKKNDAFELNHL	ELTTNATQ	SDSDEKENV	RVAEA	80				
Non-Sc_AGT1_Lager1	TDANEANNEEKSMTLRQALRKYPKAALWSI	LVSITLVM	EGYDTALLSAL	YALPVFOR	KFGTMNAEGSYEITSQWQ	GLN160				
Non-Sc_AGT1_Hybrid1	TDANEANNEEKSMTLRQALRKYPKAALWSI	LVSITLVM	EGYDTALLSAL	YALPVFOR	KFGTMNAEGSYEITSQWQ	GLN160				
Non-Sc_AGT1_Lager1	MCVLGCEMI	GLQITTYWVEFMGNRYTMTALSLLTAYIFI	LYYCKSLAMI	AVGQ	LSAMPWGC	QSLAVTYASEVCPLAL240				
Non-Sc_AGT1_Hybrid1	MCVLGCEMI	GLQITTYWVEFMGNRYTMTALSLLTAYIFI	LYYCKSLAMI	AVGQ	LSAMPWGC	QSLAVTYASEVCPLAL240				
Non-Sc_AGT1_Lager1	RYYMTSYSNI	CWLFQGI	FASGI	MKNSQENL	GNSDLGYKLPALQW	WPAPLIIGIFFAPESPWWLVRKNKI	VEAKKSLNR320			
Non-Sc_AGT1_Hybrid1	RYYMTSYSNI	CWLFQGI	FASGI	MKNSQENL	GNSDLGYKLPALQW	WPAPLIIGIFFAPESPWWLVRKNKI	VEAKKSLNR320			
Non-Sc_AGT1_Lager1	ILSGIVTEKEI	QVDTILKQI	EMTI	EKERLRASKSGSF	SCFKGVDGRTRLACL	TWACNSSGAVLLGYSTYFFERAGMA	400			
Non-Sc_AGT1_Hybrid1	ILSGIVTEKEI	QVDTILKQI	EMTI	EKERLRASKSGSF	SCFKGVDGRTRLACL	TWACNSSGAVLLGYSTYFFERAGMA	400			
Non-Sc_AGT1_Lager1	TDKAFTFSLI	QYCLGLAGTLGSWI	SGRVGRWTI	LYGLSFQMVCLFI	GGMGFASGSSASNAAGLLALSFFYNAGI	G480				
Non-Sc_AGT1_Hybrid1	TDKAFTFSLI	QYCLGLAGTLGSWI	SGRVGRWTI	LYGLSFQMVCLFI	GGMGFASGSSASNAAGLLALSFFYNAGI	G480				
Non-Sc_AGT1_Lager1	AVVYCV	VAEI	PSAELRTKTI	VLARI	CYNLMAVFNAI	LTPYMLN	NSDWNWGAKTGLYWGFTALTAWWI	DLPETTGRTF560		
Non-Sc_AGT1_Hybrid1	AVVYCV	VAEI	PSAELRTKTI	VLARI	CYNLMAVFNAI	LTPYMLN	NSDWNWGAKTGLYWGFTALTAWWI	DLPETTGRTF560		
Non-Sc_AGT1_Lager1	SEI	NEL	FSQGV	PARKFAST	VDPF	GKRGL	QNR	PQVDNI	DRFSSASQAL	611
Non-Sc_AGT1_Hybrid1	SEI	NEL	FSQGV	PARKFAST	VDPF	GKRGL	QNR	PQVDNI	DRFSSASQAL	610

Decoration": Stride (with solid black) residues that differ from the Consensus.

Decoration "": Shade (with solid black) residues that differ from the Consensus.

Fig. 12A

[illegible]

Fig. 12B

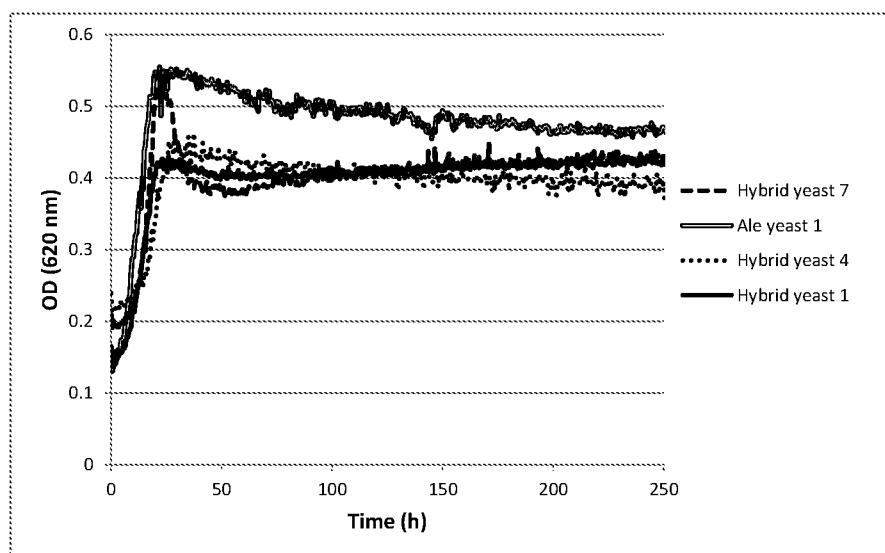
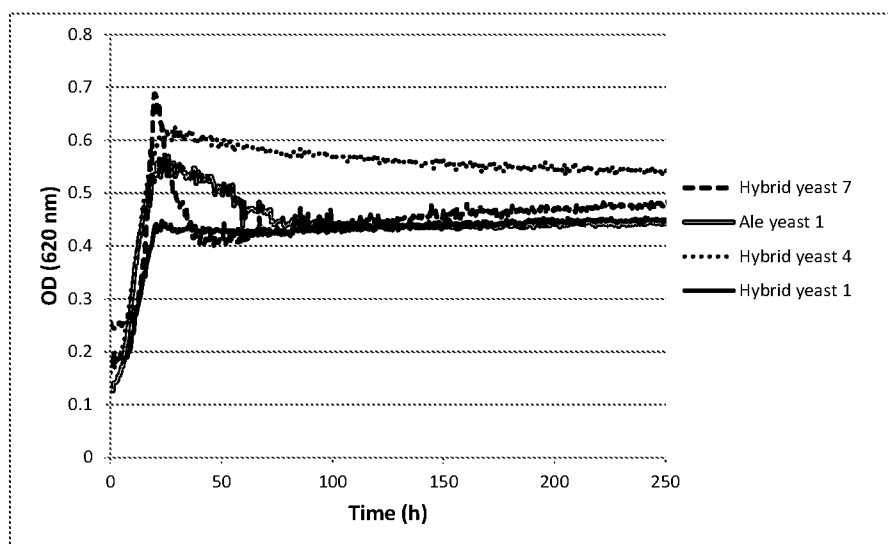


Fig. 13

A)



B)

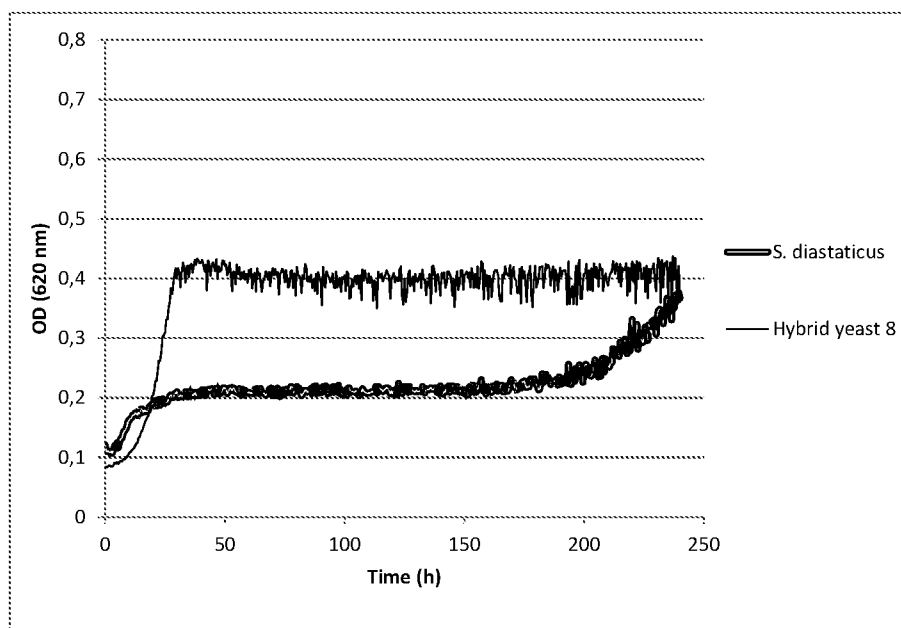
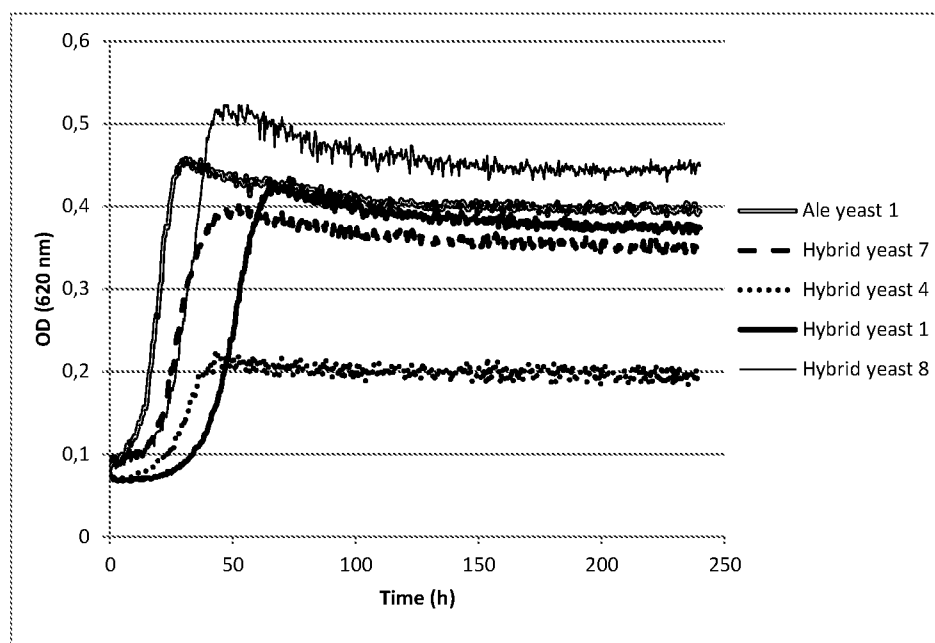


Fig. 14

A)



B)

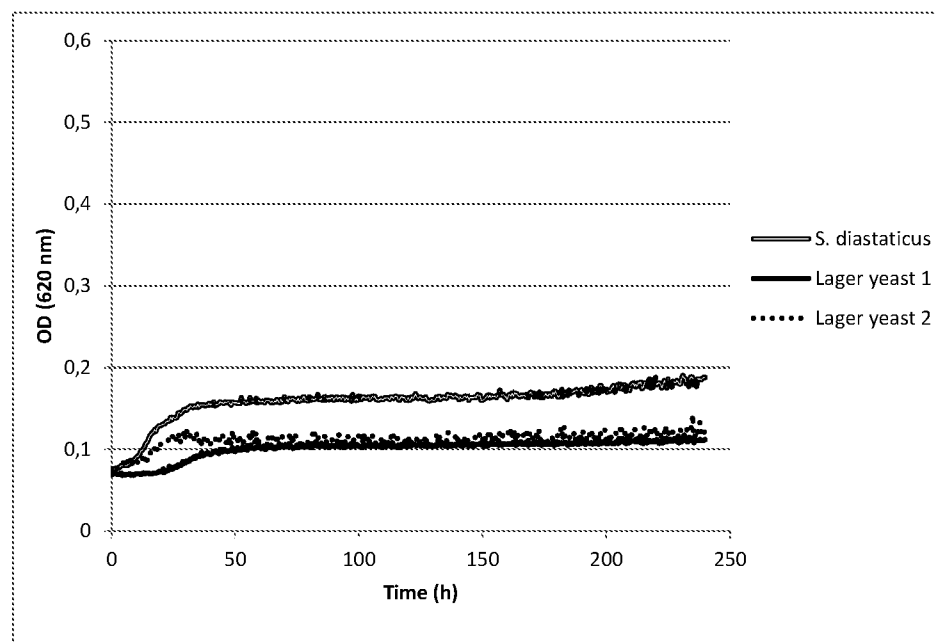


Fig. 15

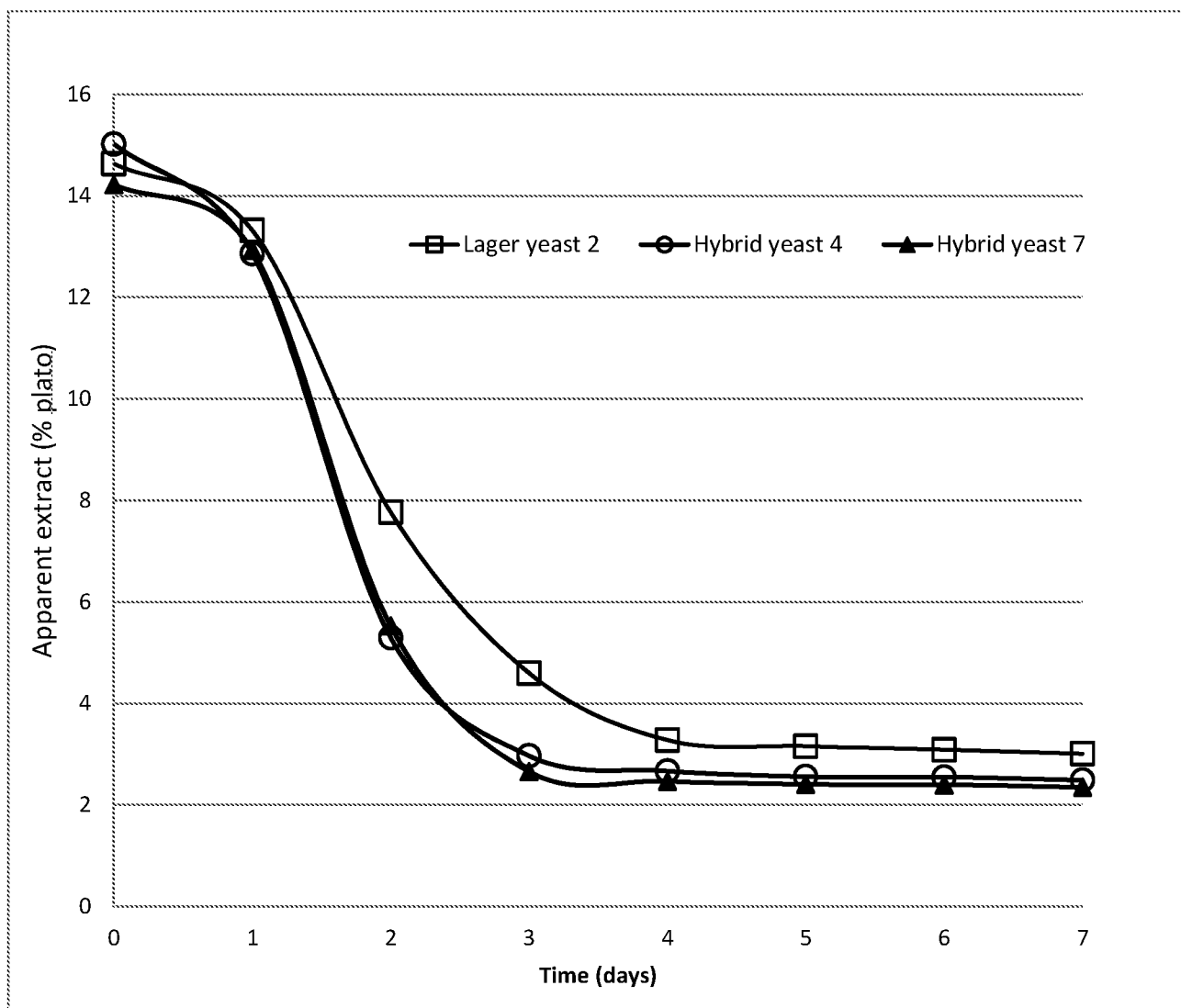


Fig. 16