

5 CLAIMS

What is claimed is:

1. Method of preparing D-psicose comprising the steps of:
 - 10 (i) providing a mixture of D-psicose and D-fructose; and
 - (ii) contacting the mixture of D-psicose and D-fructose with a probiotic microorganism that is capable of metabolizing D-fructose but not D-psicose and capable of converting D-fructose into L-lactic acid and subjecting the microorganism to culture conditions that allow fermentative removal of D-fructose from the mixture of D-psicose and D-fructose with concomitant production
15 of L-lactic acid.
2. The method of claim 1, wherein the mixture of D-psicose and D-fructose is provided by enzymatically converting D-fructose into D-psicose by a D-psicose 3-epimerase (DPEase) [EC 5.1.3.30], D-tagatose 3-epimerase (DTEase) [EC 5.1.3.31], or a cell comprising said DPEase or
20 DTEase.
3. The method of claim 2, wherein the cell comprising said DPEase or DTEase is a recombinant cell.
- 25 4. The method of claim 2 or 3, wherein the DPEase is of *Agrobacterium tumefaciens* origin.
5. The method of any one of claims 2-4, wherein the DPEase has the amino acid sequence set forth in any one of SEQ ID NOs:1-3.
- 30 6. The method of any one of claims 1-5, wherein the probiotic microorganism that is capable of metabolizing D-fructose but not D-psicose and capable of converting D-fructose into L-lactic acid is of the *Lactobacillus* or *Saccharomyces* genus, preferably *Lactobacillus rhamnosus* GG or *Saccharomyces boulardii*, more preferably *Lactobacillus rhamnosus* GG.
- 35 7. The method of claims 1-6, wherein the fermentative removal of D-fructose is carried out
 - (1) at a pH range between 6.5 and 7.5, preferably at pH 7.5; and/or
 - (2) at an initial total concentration of D-psicose and D-fructose of 200 g/L or less, preferably 180 g/L or less.
- 40 8. The method of any one of claims 1-7, wherein in the mixture of D-fructose and D-psicose the weight ratio of D-fructose to D-psicose is at least 1:1, preferably at least 1.5:1, more preferably at least 2:1.

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9. The method of any one of claims 1 to 8, wherein step (ii) is carried out under conditions and for a period of time that allows the microorganism to metabolize substantially all D-fructose from the mixture.

10 10. The method of any one of claims 1 to 9, wherein the method further comprises the step (iii) of isolating the D-psicose, preferably by centrifugation and/or chromatography.

11. Use of a probiotic microorganism that is capable of metabolizing D-fructose but not D-psicose and capable of converting D-fructose into L-lactic acid for fermentative removal of D-
15 fructose from a mixture of D-psicose and D-fructose.

12. The use of claim 11, wherein the probiotic microorganism that is selective for D-fructose and capable of converting D-fructose into L-lactic acid is *Lactobacillus rhamnosus* GG or *Saccharomyces boulardii*, preferably *Lactobacillus rhamnosus* GG.

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