

Claims

1. A method for reducing antennary fucosylation of complex N-glycans in a recombinantly expressed glycoprotein, comprising the step of overexpressing together with the glycoprotein β -galactoside α -2,6-sialyltransferase 1 (ST6Gal1) and/or α -2,3-sialyltransferase 4 (ST3Gal4).
2. The method of claim 1, wherein the glycoprotein is characterized by a significantly reduced antennary fucosylation of complex N-glycans as compared to the same recombinant glycoprotein expressed without overexpression of ST6Gal1 and/or ST3Gal4.
3. The method of claim 1 or claim 2, wherein at least 80% of the complex N-glycan antennae of the recombinantly expressed glycoprotein are not fucosylated.
4. The method of any one of claims 1 to 3, wherein the glycoprotein is selected from the group consisting of α 1-antitrypsin (AAT), hepatocyte growth factor (HGF), Factor VII (FVII), Factor VIII (FVIII), Factor IX (FIX), von Willebrand-Factor (vWF), alkaline phosphatase, and C1 esterase inhibitor (C1-inhibitor; C1 Inh).
5. The method of any one of claims 1 to 4, wherein the glycoprotein is a mammalian, preferably a human glycoprotein.
6. The method of any one of claims 1 to 5, wherein
 - (i) β -galactoside α -2,3-sialyltransferase 1 (ST3Gal1) is not overexpressed together with the glycoprotein, and/or
 - (ii) the expression of α -1,3-mannosyl-glycoprotein 4- β -N-acetylglucosaminyltransferase A (GnTIVa), α -1,3-mannosyl-glycoprotein 4- β -N-acetylglucosaminyltransferase B (GnTIVb), and α -1,6-

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mannosylglycoprotein 6- β -N-acetylglucosaminyltransferase A (GnTV) is not reduced.

7. A cell line that is genetically modified to overexpress β -galactoside α -2,6-sialyltransferase 1 (ST6Gal1) and/or α -2,3-sialyltransferase 4 (ST3Gal4).
8. The cell line of claim 7, wherein the cell line comprises endogenous gene(s) encoding ST6Gal1 and/or ST3Gal4, and further has at least one genetic element, selected from the group consisting of a promoter, an enhancing element, and a stabilizing element inserted into the genome in one or more position(s) suitable to cause overexpression of ST6Gal1 and/or ST3Gal4.
9. The cell line of claim 7 or claim 8, wherein the cell line comprises exogenous nucleic acid(s) encoding ST6Gal1 and/or ST3Gal4.
10. The cell line of any one of claims 7 to 9, wherein the cell line is derived from a cell line, selected from the group consisting of AGE.CR[®] cells, Vero cells, MDCK cells, BHK cells, CHO cells, HEK293 cells, HepG2 cells, Huh7 cells, AGE1.HN[®] cells, NC5T11 cells, Per.C6 cells, HMCLs cells, MM.1 cells, U266 cells, RPMI18226 cells, HKB11 cells, NM cells, NM-F9 cells, and CAP cells.
11. The cell line of any one of claims 7 to 9, wherein the cell line is derived from human primary amniocytes comprising at least one nucleic acid encoding the gene products of the adenoviral E1 and pIX regions.
12. The cell line of any one of claims 7 to 11, wherein the cell line is not genetically modified to
 - (i) overexpress β -galactoside α -2,3-sialyltransferase 1 (ST3Gal1), and/or
 - (ii) reduce the expression of α -1,3-mannosyl-glycoprotein 4- β -N-acetylglucosaminyltransferase A (GnTIVa), α -1,3-mannosyl-glycoprotein 4- β -N-acetylglucosaminyltransferase B (GnTIVb), and α -1,6-mannosylglycoprotein 6- β -N-acetylglucosaminyltransferase A (GnTV).

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13. A recombinant glycoprotein having complex N-glycans, wherein antennary fucosylation of the complex N-glycans is reduced, so that at least 80% of the complex N-glycan antennae of the recombinant glycoprotein are not fucosylated.
14. The recombinant glycoprotein of claim 13, wherein the glycoprotein is selected from the group consisting of α 1-antitrypsin (AAT), hepatocyte growth factor (HGF), Factor VII (FVII), Factor VIII (FVIII), Factor IX (FIX), von Willebrand-Factor (vWF), alkaline phosphatase, and C1 esterase inhibitor (C1-inhibitor; C1 Inh).
15. A method for the expression of a recombinant glycoprotein of claim 13 or claim 14, comprising the steps of:
 - (a) providing a cell line of any one of claims 7 to 12,
 - (b) expressing the recombinant glycoprotein in said cell line; and
 - (c) isolating the recombinant glycoprotein from the cells or the cell culture supernatant.