Claims

1. A composition comprising chondrocytes, wherein said composition has an integrin alpha10 to integrin alpha11 expression ratio of at least 4 to 1.

2. The composition according to any one of the preceding claims, wherein the integrin alpha10 to integrin alpha11 expression ratio is at least 4 to 1, such as at least 5 to 1, such as at least 10 to 1, such as at least 15 to 1, such as at least 20 to 1, such as at least 25 to 1, such as at least 30 to 1, such as at least 35 to 1, such as at least 40 to 1, such as at least 45 to 1, such as at least 50 to 1, such as at least 60 to 1, such as at least 70 to 1, such as at least 80 to 1, such as at least 90 to 1, such as at least 100 to 1, such as at least 150 to 1, such as at least 200 to 1.

3. The composition according to any one of the preceding claims, wherein the integrin alpha10 and/or integrin alpha11 is expressed as a heterodimer in combination with an integrin beta1 subunit.

4. The composition according to any one of the preceding claims, wherein the composition comprises differentiated chondrocytes, and wherein said differentiated chondrocytes express integrin alpha10 and substantially no integrin alpha11.

5. The composition according to any one of the preceding claims, wherein the composition comprises partially dedifferentiated chondrocytes, and wherein said partially dedifferentiated chondrocytes express both integrin alpha10 and integrin alpha11.

6. The composition according to any one of the preceding claims, wherein said composition is selected from the group consisting of a cartilage tissue preparation, a tissue prepration comprising chondrocytes, a monolayer chondrocyte cell culture, and a three-dimensional chondrocyte cell culture, wherein the three-dimensional chondrocyte cell culture is a shperoid, a pellet, a cell sheet or chondrocytes cultured in scaffolds.

7. The composition according to any one of the preceding claims, wherein said composition is derived from a cartilage tissue, a chondrocyte comprising tissue, a monolayer chondrocyte cell culture, and a three-dimensional chondrocyte cell culture, wherein the three-dimensional chondrocyte cell culture is a shperoid, a pellet, a cell sheet or chondrocytes cultured in scaffolds.

8. The composition according to any one of the preceding claims, wherein the cells in the composition are mammalian cells.

9. The composition according to any one of the preceding claims, wherein the cells in the composition are human.

10. The composition according to any one of the preceding claims, wherein the cells in the composition are equine.

11. The composition according to any one of the preceding claims, wherein the cells in the composition are canine.

12. The composition according to any one of the preceding claims, wherein the integrin alpha10 and/or the integrin alpha11 is expressed on the cell surface of a mammalian chondrocyte or a fibroblast-like cell.

13. The composition according to any one of the preceding claims, wherein said composition is substantially free from contaminating cells, wherein the contaminating cells express integrin alpha11 but not integrin alpha10.

14. The composition according to any one of the preceding claims, wherein said composition comprises at least 80 %, such as at least 85 %, such as at least 90 %, such as at least 95 %, such as at least 98 %, such as at least 99 %, such as 100 % chondrocytes, out of the total composition or population of cells.

15. The composition according to any one of the preceding claims, wherein the chondrocytes further express a secondary marker selected from the group

consisting of CEP-68/CRTAC1, GP-39 cartilage glycoprotein, CD44, CD166, Collagen IIA, Collagen IIB, Aggrecan, Alizarian Red (neg), Alcian Blue, CRTAC1, CEP-68, CD146, CD90, CD49e, CD63 and Sca-1.

16. A method for determining quality of a composition comprising chondrocytes, wherein the quality determination comprises one or more of the following criteria:

- identity of the chondrocytes,
- differentiation state of the chondrocytes,
- purity of the chondrocytes, and
- potency of the chondrocytes

the method comprising the steps of:

a) providing a candidate composition comprising chondrocytes;

b) analyzing the expression levels of integrin alpha10 and integrin alpha11 in said composition;

c) comparing the expression level of integrin alpha10 to the expression level of integrin alpha11,

wherein an integrin alpha10 to integrin alpha11 expression ratio of at least 4 to 1

is indicative of

- chondrocytic identity of the chondrocytes in the chondrocyte composition,
- differentiated state of the chondrocytes in the chondrocyte composition,
- purity of the chondrocyte composition, and/or
- potency of the chondrocytes of the chondrocyte composition,

and thereby is indicative of a high quality chondrocyte composition.

17. A method for isolating a population of high quality chondrocytes, the method comprising the steps of:

a) providing a candidate composition comprising chondrocytes;

b) analyzing expression of integrin alpha10 and integrin alpha11 in said composition; and

c) enriching chondrocytes having an integrin alpha10 to integrin alpha11 expression ratio of at least 4 to 1,

thus obtaining an isolated population of high quality chondrocytes.

18. A method for manufacturing a population of high quality chondrocytes, the method comprising the steps of:

a) providing a candidate composition comprising chondrocytes;

b) analyzing expression of integrin alpha10 and integrin alpha11 in said composition; and

c) expanding chondrocytes having an integrin alpha10 to integrin alpha11 expression ratio of at least 4 to 1,

thus manufacturing a population of high quality chondrocytes.

19. The method according to any one of claims16 to 18, wherein the integrin alpha10 to integrin alpha11 expression ratio is at least 4 to 1, such as at least 10 to 1, such as at least 15 to 1, such as at least 20 to 1, such as at least 25 to 1, such as at least 30 to 1, such as at least 35 to 1, such as at least 40 to 1, such as at least 45 to 1, such as at least 50 to 1, such as at least 60 to 1, such as at least 70 to 1, such as at least 80 to 1, such as at least 90 to 1, such as at least 100 to 1, such as at least 150 to 1, such as at least 200 to 1.

20. The method according to any one of claims 16 to 19, wherein the integrin alpha11 expression is at the most 25% of the integrin alpha10 expression, such as at the most 20% of the integrin alpha10 expression, such as at the most 15% of the integrin alpha10 expression, such as at the most 10% of the integrin alpha10 expression, such as at the most 5% of the integrin alpha10 expression, such as at the most 4% of the integrin alpha10 expression, such as at the most 3% of the integrin alpha10 expression, such as at the most 2% of

the integrin alpha10 expression, such as at the most 1% of the integrin alpha10 expression.

21. The method according to any one of claims 16 to 20, wherein expression of both integrin alpha10 and integrin alpha11 is indicative of a partially dedifferentiated chondrocyte composition.

22. The method according to any one of claims 16 to 21, wherein expression of integrin alpha10 and substantially no expression of integrin alpha11 is indicative of a differentiated chondrocyte composition.

23. The method according to any one of claims 16 to 22, wherein at the most 20% of the cells in the chondrocyte composition expressing alpha11 and expressing substantially no integrin alpha10, is indicative of purity of the chondrocyte composition.

24. The method according to any one of claims 16 to 23, wherein at the most 20%, such as at the most 15%, such as at the most 10%, such as at the most 5%, such as at the most 4%, such as at the most 3%, such as at the most 2%, such as at the most 1%, such as at the most 0.5%, of the cells in the chondrocyte composition expressing alpha11 and substantially no integrin alpha10, is indicative of purity of the chondrocyte composition.

25. The method according to any one of claims 16 to 24, wherein substantially no integrin alpha11 is expressed in the chondrocyte composition or population.

26. The method according to any one of claims 16 to 25, wherein the candidate composition is selected from the group consisting of a cartilage tissue preparation, a tissue prepration comprising chondrocytes, a monolayer chondrocyte cell culture, and a three-dimensional chondrocyte cell culture, wherein the three-dimensional chondrocyte cell culture is a shperoid, a pellet, a cell sheet or chondrocytes cultured in scaffolds.

27. The method according to any one of claims 16 to 26, wherein the candidate composition is derived from a cartilage tissue, a chondrocyte comprising tissue, a monolayer chondrocyte cell culture, and a three-dimensional chondrocyte cell culture, wherein the three-dimensional chondrocyte cell culture is a shperoid, a pellet, a cell sheet or chondrocytes cultured in scaffolds.

28. The method according to any one of claims 16 to 27, wherein the expression analysis is conducted by a method selected from the group consisting of flow cytometry, ELISA, Western Blot, immunoprecipitation, dot blot, qPCR, immunoassay, immunofluorescence, gene expression analysis and any other suitable method.

29. The method according to any one of claims 16 to 28, wherein the analysis comprises quantitative determination of the expression of integrin alpha10 and integrin alpha11.

30. The method according to any one of claims 16 to 29, wherein the integrin alpha10 and/or the integrin alpha11 is expressed on the cell surface of a mammalian chondrocyte and/or a fibroblast-like cells.

31. The method according to any one of claims 16 to 30, wherein the integrin alpha10 and/or integrin alpha11 is expressed as a heterodimer in combination with an integrin beta1 subunit.

32. The method according to any one of claims 16 to 31, further comprising detecting expression of a secondary marker selected from the group consisting of CEP-68/CRTAC1, GP-39 cartilage glycoprotein, CD44, CD166, CollagenIIA, Collagen IIB, Aggrecan, Alizarian Red (neg), Alcian Blue, CRTAC1, CEP-68, CD49e, CD63, CD146, CD90 and Sca-1.

33. The method according to any one of claims 16 to 32, wherein the analysis of expression of an integrin alpha10 by a cell comprises measuring integrin alpha10 protein expression.

34. The method according to any one of claims 16 to 33, wherein the analysis of expression of an integrin alpha11 by a cell comprises measuring integrin alpha11 protein expression.

35. The method according to claim 33, wherein the integrin alpha10 protein expression is detected by an anti-integrin alpha10 antibody.

36. The method according to claim 34, wherein the integrin alpha11 protein expression is detected by an anti-integrin alpha11 antibody.

37. The method according to any one of claims 16 to 36, wherein the analysis of expression of an integrin alpha10 comprises measuring integrin alpha10 mRNA expression.

38. The method according to any one of claims 16 to 37, wherein the analysis of expression of an integrin alpha11 comprises measuring integrin alpha11 mRNA expression.

39. The method according to any one of claims 16 to 38, wherein the antibody used for detection of expression is a monoclonal antibody, polyclonal antibody, a single chain antibody or fragment thereof.

40. The method according to any one of claims 16 to 39, wherein the antibody is a non-human antibody, a bispecific antibody, a chimeric antibody, a humanized antibody or a human antibody.

41. The method according to any one of claims 16 to 40, wherein the antibody is covalently bound to a detectable moiety, such as a detectable moiety selected from the group consisting of a fluorophore, an enzyme, a protein or a radioactive tracer or radioisotope.

42. The method according to any one of claims 16 to 41, wherein the antibody has an isotype selected from the group consisting of IgA, IgD, IgG and IgM.

43. The method according to any one of claims 16 to 42, wherein the anti-integrin alpha10 antibody is:

a) a monoclonal antibody, produced by the hybridoma cell line deposited at the Deutsche Sammlung von Microorganismen und Zellkulturen GmbH under the accession number DSM ACC2583; or

b) an antibody which competes for binding to the same epitope as the epitope bound by the monoclonal antibody produced by the hybridoma deposited at the Deutsche Sammlung von Microorganismen und Zellkulturen GmbH under the accession number DSM ACC2583; or

c) a fragment of a) or b), wherein said fragment is capable of binding specifically to the extracellular I-domain and/or to the cytoplasmic domain of the integrin alpha10 chain.

44. The method according to any one of claims 16 to 43, wherein the antibody is attached to a solid support, such as to magnetic beads.

45. The method according to any one of claims 16 to 44, wherein the antibody is covalently bound to biotin.

46. The method according to any one of claims 16 to 45, wherein the antibody is labeled with one or more fluorophore(s).

47. The method according to any one of claims 16 to 46, wherein the fluorophore is selected from the group consisting of phycoerythrin, allophycocyanin, fluorescein, Texas red, Alexa Fluor 647, and brilliant dyes.

48. The method according to any one of claims 16 to 47, wherein the analysis of the expression of integrin alpha10 and integrin alpha11 is conducted simultaneously.

49. The method according to any one of claims 16 to 48, wherein the anti-integrin alpha10 antibody is labeled with a fluorophore, and the anti-integrin alpha11 antibody is labeled with another fluorophore, and wherein the emission spectra of the two fluorophores are non-overlapping.

50. The method according to any one of claims 16 to 49, wherein the chondrocyte composition or population obtained comprises at least 80%, such as at least 85%, such as at least 90%, such as at least 95%, such as at least 98%, such as at least 99%, such as 100% chondrocytes, out of the total population of cells.

51. The method according to any one of claims 16 to 50, wherein the chondrocyte composition or population obtained comprises at the most 20% contaminating cells, such as at the most 15% contaminating cells, such as at the most 10% contaminating cells, such as at the most 5% contaminating cells, such as the most 4% contaminating cells, such as at the most 3% contaminating cells, such as at the most 2% contaminating cells, such as at the most 1% contaminating cells, such as at the most 0.5% contaminating cells, wherein the contaminating cells express integrin alpha11 but not integrin alpha10.

52. The method according to any one of claims 16 to 51, wherein the chondrocyte composition or population obtained is substantially free from contaminating cells, wherein the contaminating cells express integrin alpha11 but not integrin alpha10.

53. The method according to any one of claims 16 and 52, wherein the chondrocyte composition or population obtained is an enriched and/or purified population of chondrocytes.

54. The method according to any one of claims 16 to 53, wherein the method further comprises cultivating the composition of chondrocytes under conditions that induce integrin alpha10 expression.

55. The method according to any one of claims 16 to 54, wherein the method further comprises selecting cells expressing integrin alpha10.

56. The method according to any one of claims 16 to 55, wherein the method further comprises depletion of cells expressing integrin alpha11.

57. A method for optimizing chondrocytic potency of a composition comprising chondrocytes, the method comprising inducing expression of integrin alpha10.

58. A method of increasing proliferation rate of chondrocytes, the method comprising inducing expression of integrin alpha10 of said chondrocytes.

59. The method according to any one of claims 57 and 58, further comprising suppressing expression of integrin alpha11.

60. The method according to any one of claims 57 to 59, wherein the induction of integrin alpha10-expressing cells is conducted in a medium comprising ascorbic acid and/or platelet lysate.

61. The method according to any one of claims 57 to 60, wherein the integrin alpha10 to integrin alpha11 expression ratio thus obtained is at least 4 to 1, such as at least 5 to 1, such as at least 10 to 1, such as at least 15 to 1, such as at least 20 to 1, such as at least 25 to 1, such as at least 30 to 1, such as at least 35 to 1, such as at least 40 to 1, such as at least 45 to 1, such as at least 50 to 1, such as at least 60 to 1, such as at least 70 to 1, such as at least 80 to 1, such as at least 90 to 1, such as at least 100 to 1, such as at least 150 to 1, such as at least 200 to 1.

62. The method according to any one of claims 57 to 61, wherein the obtained integrin alpha11 expression is at the most 25% of the obtained integrin alpha10 expression, such as at the most 20% of the obtained integrin alpha10 expression, such as at the most 15% of the obtained integrin alpha10 expression, such as at the most 10% of the obtained integrin alpha10 expression, such as at the most 5% of the obtained integrin alpha10 expression, such as at the most 4% of the obtained integrin alpha10 expression, such as at the most 3% of the obtained integrin alpha10

expression, such as at the most 2% of the obtained integrin alpha10 expression, such as at the most 1% of the obtained integrin alpha10 expression, such as at the most 0.5% of the obtained integrin alpha10 expression.

63. Use of the method according to any one of claims 16 to 56, for

a) quality assuring chondrocyte compositions;

b) monitoring of consistency and/or reproducibility of chondrocyte compositions;

c) predicting potency of chondrocyte compositions;

d) in-process control chondrocyte compositions;

e) in-process control of expansion of chondrocyte compositions;

f) conducting release test of chondrocyte compositions;

g) conducting a potency assay of a chondrocyte composition; and/or

h) prediction of clinical outcome of chondrocyte compositions.