Claims

1. A method for producing a retinal cell or a retinal tissue, comprising the following steps (1) - (3):
(1) a first step of culturing human pluripotent stem cells in the absence of feeder cells in a medium containing 1) a TGFβ family signal transduction pathway inhibiting substance and/or a Sonic hedgehog signal transduction pathway activating substance, and 2) a factor for maintaining undifferentiated state,
(2) a second step of culturing the cells obtained in the first step in suspension in a medium containing a Wnt signal transduction pathway inhibiting substance to form a cell aggregate, and
(3) a third step of culturing the aggregate obtained in the second step in suspension in the presence or absence of a Wnt signal transduction pathway inhibiting substance in a medium containing a BMP signal transduction pathway activating substance to obtain an aggregate containing a retinal cell or a retinal tissue,
wherein the TGFβ family signal transduction pathway inhibiting substance is selected from the group consisting of Lefty, SB431542, A-83-01, LDN193189 and Dorsomorphin, wherein the Sonic hedgehog signal transduction pathway activating substance is selected from the group consisting of Shh, SAG and Purmorphamine, wherein the Wnt signal transduction pathway inhibiting substance is selected from the group consisting of CKI-7, D4476, IWR-1-endo and IWP-2, and wherein the BMP signal transduction pathway activating substance is selected from the group consisting of BMP2, BMP4, BMP7 and GDF7.

2. The production method according to claim 1, wherein the pluripotent stem cells are cultured for 0.5 hr - 144 hr in the first step.

3. The production method according to claim 1 or 2, wherein the culturing in the first step is performed by adhesion culture.

4. The production method according to any of claims 1 to 3, wherein, in the second step, the cells obtained in the first step are dispersed, and the dispersed cells are cultured in suspension.

5. The production method according to any of claims 1 to 4, wherein the factor for maintaining undifferentiated state is one or more proteins selected from the group consisting of bFGF, FGF4, FGF8, EGF, Nodal, Activin A, Activin B, TGFβ1 and TGFβ2.

6. The production method according to claim 5, wherein the factor for maintaining undifferentiated state is bFGF.

7. The production method according to any of claims 1 to 6, wherein the medium used for suspension culture in the second step further comprises a Sonic hedgehog signal transduction pathway activating substance which is selected from the group consisting of Shh, SAG and Purmorphamine.

8. The production method according to claim 7, wherein, in the second step, the concentration of the Sonic hedgehog signal transduction pathway activating substance in the medium is a concentration corresponding to Sonic hedgehog signal transduction activity of SAG at 10 nM to 700 nM.

9. The production method according to any of claims 1 to 8, wherein, in the third step, the BMP signal transduction pathway activating substance is added to the

medium between day 1 and day 9 from the start of the second step.

10. The production method according to claim 9, wherein, in the third step, the BMP signal transduction pathway activating substance is added to the medium between day 1 and day 6 from the start of the second step.

11. The production method according to claim 10, wherein, in the third step, the BMP signal transduction pathway activating substance is added to the medium between day 1 and day 3 from the start of the second step.

12. The production method according to any of claims 1 to 11, wherein the BMP signal transduction pathway activating substance is BMP4.

13. The production method according to any of claims 1 to 12, wherein, in the third step, the concentration of the Sonic hedgehog signal transduction pathway activating substance in the medium is not more than a concentration corresponding to Sonic hedgehog signal transduction activity of SAG at 700 nM.

14. The production method according to any of claims 1 to 13, wherein the culturing is performed in a medium containing a Wnt signal transduction pathway inhibiting substance for 3 days to for 18 days from the start of the second step.

15. The production method according to any of claims 1 to 14, wherein the culturing is performed in a medium containing a Wnt signal transduction pathway inhibiting substance for 10 days from the start of the second step.

16. The production method according to any of claims 1 to

15, wherein the Wnt signal transduction pathway inhibiting substance is IWR-1-endo.

17. The production method according to any of claims 1 to 16, wherein the aggregate obtained in the third step comprises one or more cells selected from the group consisting of retinal progenitor cell, neural retinal progenitor cell, photoreceptor precursor cell, photoreceptor cell, rod photoreceptor cell, cone photoreceptor cell, horizontal cell, amacrine cell, interneuron, ganglion cell, bipolar cell, retinal pigment epithelial cell, and ciliary marginal zone cell.

18. The production method according to any of claims 1 to 17, wherein the pluripotent stem cells are induced pluripotent stem cells.

19. The production method according to any of claims 1 to 18, wherein a uniformed aggregate is formed in the second step.

20. The production method according to any of claims 1 to 19, wherein the suspension culture is performed in the absence of a basement membrane preparation.

21. The production method according to any of claims 1-20, wherein the pluripotent-like state of pluripotent stem cells is maintained throughout the first step.

22. The production method according to any of claims 1-21, wherein the cells obtained in the first step contain Oct3/4 positive stem cells at a proportion of not less than 60%.

23. A method for producing a retinal tissue comprising: producing a cell aggregate comprising a retinal tissue by

the production method according to any of claims 1 to 22, and
cutting out the retinal tissue from the cell aggregate.