CLAIMS

[Claim 1]

A method for evaluating the quality of a transplant neural retina,

the method comprising:

sampling a part or the whole of a cell aggregate containing a neural retina having an epithelial structure derived from a pluripotent stem cell as a sample for quality evaluation;

detecting the expression of a neural retina-related cell-related gene and a non-neural retina-related cell-related gene in the sample for quality evaluation; and

when the expression of the neural retina-related cell-related gene is found and the expression of the non-neural retina-related cell-related gene is not found, determining that

(1) the neural retina (transplant neural retina) in the same cell aggregate as the cell aggregate containing the sample for quality evaluation being the part,

(2) the neural retina (transplant neural retina) in a cell aggregate of the same lot as the cell aggregate containing the sample for quality evaluation being the part, or

(3) the neural retina (transplant neural retina) in a cell aggregate of the same lot as the cell aggregate of the sample for quality evaluation being the whole,

is applicable as the transplant neural retina, wherein

the non-neural retina-related cell-related gene comprises one or more genes selected from the group consisting of a brain and spinal cord tissue marker gene and an eyeball-related tissue marker gene, wherein

the brain and spinal cord tissue marker gene is one or more genes selected from the group consisting of a telencephalon marker gene, a diencephalon/midbrain marker gene, and a spinal cord marker gene, and

the eyeball-related tissue marker gene is one or more genes selected from the group consisting of an optic stalk marker gene, a ciliary body marker gene, a lens marker gene and a retinal pigment epithelium marker gene, and, wherein

the neural retina-related cell-related gene is a gene highly expressed in a neural retina-related cell compared with a non-neural retina-related cell.

[Claim 2]

The method according to claim 1, wherein the non-neural retina-related cell-related gene comprises a spinal cord marker gene.

[Claim 3]

The method according to claim 1 or 2, wherein the non-neural retina-related cell-related gene comprises a telencephalon marker gene, a spinal cord marker gene, an optic stalk marker gene, a ciliary body marker gene, and a retinal pigment epithelium marker gene.

[Claim 4]

The method according to any one of claims 1 to 3, wherein

the telencephalon marker gene comprises one or more genes selected from the group consisting of FoxG1, Emx2, Dlx2, Dlx1 and Dlx5,

the diencephalon/midbrain marker gene comprises one or more genes selected from the group consisting of DMBX1, Rx, Nkx2.1, OTP, FGFR2, EFNA5 and GAD1,

the spinal cord marker gene comprises one or more genes selected from the group consisting of HOXD4, HOXD3, HOXD1, HOXC5, HOXA5 and HOXB2,

the optic stalk marker gene comprises one or more genes selected from the group consisting of GREM1, GPR17, ACVR1C, CDH6, Pax2, Pax8, GAD2 and SEMA5A,

the ciliary body marker gene comprises one or more genes selected from the group consisting of Zic1, MAL, HNF1beta, FoxQ1, CLDN2, CLDN1, GPR177, AQP1 and AQP4,

the lens marker gene comprises one or more genes selected from the group consisting of CRYAA and CRYBA1, and

the retinal pigment epithelium marker gene comprises one or more genes selected from the group consisting of MITF, TTR and BEST1.

[Claim 5]

The method according to any one of claims 1 to 4, wherein the neural retina-related cell-related gene is one or more genes selected from the group consisting of RAX, Chx10, SIX3, SIX6, RCVRN, CRX, NRL and NESTIN

[Claim 6]

The method according to any one of claims 1 to 5, wherein the non-neural retina-related cell-related gene further comprises an undifferentiated pluripotent stem cell marker gene.

[Claim 7]

The method according to claim 6, wherein the undifferentiated pluripotent stem cell marker gene comprises one or more genes selected from the group consisting of Oct3/4, Nanog and lin28.

[Claim 8]

The method according to any one of claims 1 to 7, wherein the cell aggregate of the same lot as the cell aggregate of the sample for quality evaluation is a cell aggregate produced under a condition exhibiting a gene expression profile equivalent to that of the transplant neural retina.

[Claim 9]

The method according to any one of claims 1 to 8, wherein

the sample for quality evaluation is a part of the cell aggregate, and

when the expression of the neural retina-related cell-related gene is found and the expression of the non-neural retina-related cell-related gene is not found, it is determined that a neural retina continuous or adjacent at least partially to the part in the same cell aggregate as the cell aggregate containing the sample for quality evaluation being the part is applicable as the transplant neural retina.

[Claim 10]

The method according to claim 9, wherein the transplant neural retina is contained in the same epithelial tissue as that of the sample for quality evaluation.

[Claim 11]

The method according to claim 10, wherein the transplant neural retina contains the center and/or its neighborhood of the same epithelial tissue.

[Claim 12]

The method according to claim 11, wherein the transplant neural retina is continuous epithelial tissue.

[Claim 13]

The method according to any one of claims 9 to 12, wherein

the cell aggregate containing a neural retina contains first epithelial tissue containing the transplant neural retina, and second epithelial tissue having the continuity of the slope of a tangent line to a surface different from the continuity of the slope of a tangent line to the surface of the first epithelial tissue, and containing a non-neural retina-related cell,

the transplant neural retina contains a region on the first epithelial tissue most distant from the second epithelial tissue, and

the sample for quality evaluation is a part present between the second epithelial tissue and the transplant neural retina.

[Claim 14]

The method according to claim 13, wherein the second epithelial tissue is eyeball-related tissue and/or brain and spinal cord tissue.

[Claim 15]

The method according to claim 14, wherein the eyeball-related tissue contains a retinal pigment epithelial cell and ciliary body.

[Claim 16]

The method according to any one of claims 1 to 15, comprising performing the detection of the expression of the neural retina-related cell-related gene and the non-neural retina-related cell-related gene by quantitative PCR.

[Claim 17]

The method according to claim 16, comprising determining as being applicable as the transplant neural retina when the following reference 1 and reference 2 are satisfied:

reference 1: the difference between the threshold cycle (Ct) value of the neural retina-related cell-related gene and the Ct value of an internal standard gene (ΔCt value) is 10 or less, and

reference 2: the difference between the Ct value of the non-neural retina-related cell-related gene and the Ct value of the internal standard gene (ΔCt value) is 5 or more.

[Claim 18]

The method according to claim 16 or 17, wherein the quantitative PCR is performed by a method comprising the following steps (1) to (4), thereby

simultaneously detecting the respective expression levels of neural retina-related cell-related gene and non-neural retina-related cell-related gene in two or more of the samples for quality evaluation:

(1) providing a flow channel plate having one sample well group consisting of 8 or more and 800 or less independent sample wells, one or more primer well groups consisting of 8 or more and 800 or less independent primer wells, and flow channels connecting the independent sample wells in the sample well group with the independent primer wells in each primer well group, solutions containing nucleic acids obtained from the two or more of the samples for quality evaluation (sample solutions), and a solution containing one or a plurality of primers specific for each of one or more of the neural retina-related cell-related genes or the non-neural retina-related cell-related genes (primer solution);

(2) adding the sample solutions at one sample solution/one sample well for each of the samples for quality evaluation to the sample well group;

(3) adding the primer solution to one or more primer wells in the one or more primer well groups so as to be different primer well groups;

(4) separately mixing the primers with the nucleic acids via the flow channels; and

(5) performing quantitative PCR using the mixture obtained in (4).

[Claim 19]

A method for producing a neural retina sheet, comprising:

selecting a transplant neural retina determined as being applicable as the transplant neural retina by evaluating a cell aggregate containing a neural retina having an epithelial structure derived from a pluripotent stem cell by use of the method according to any one of claims 1 to 18; and

isolating the selected transplant neural retina.

[Claim 20]

A method for producing a neural retina sheet, comprising:

sampling a sample for quality evaluation from each of 2 or more and 800 or less cell aggregates containing a neural retina having an epithelial structure derived from a pluripotent stem cell, the sample for quality evaluation being a part of the cell aggregate;

selecting a transplant neural retina determined as being applicable as the transplant neural retina by evaluating the sampled 2 or more and 800 or less samples for quality evaluation by use of the method according to any one of claims 1 to 18; and

isolating the selected transplant neural retina.

[Claim 21]

The method according to claim 19 or 20, wherein

the cell aggregate is a cell aggregate containing at least first epithelial tissue and second epithelial tissue, obtained by differentiating a pluripotent stem cell, wherein the first epithelial tissue contains a human neural retina, and the second epithelial tissue has the continuity of the slope of a tangent line to a surface

different from the continuity of the slope of a tangent line to the surface of the first epithelial tissue, and contains a non-neural retina-related cell, and

the isolation of the transplant neural retina is isolation from the cell aggregate such that the transplant neural retina contains a region on the first epithelial tissue most distant from the second epithelial tissue.