CLAIMS

1. A culture chamber comprising a plurality of recesses each formed of a bottom portion and an opening portion, wherein

the bottom portion has one of a hemispherical shape and a truncated cone shape,

the opening portion is defined by a wall that surrounds an area from a boundary between the opening portion and the bottom portion to an end of each of the recesses, the wall having a taper angle in a range from 1 degree to 20 degrees,

an equivalent diameter of the boundary is in a range from 50 μm to 2 mm and a depth from a bottom of the bottom portion to the end of each of the recesses is in a range from 0.6 or more times to 3 or less times the equivalent diameter, and

the wall defining the opening portion forms a surface continuous to the bottom portion, and an inclination of the continuous surface changes at the boundary.

2. The culture chamber according to Claim 1, wherein the end of each of the recesses has one of a hemispherical shape, a trapezoidal shape, and an inverted triangular shape.

3. The culture chamber according to Claim 1 or 2, wherein an area between two adjacent recesses is flat and a distance between the two recesses is in a range from 5 μm to 50 μm.

4. The culture chamber according to any one of Claims 1 to 3, wherein the culture chamber is a resin molding formed of one or a combination of two or more selected from the group consisting of acrylic resin, polylactic acid, polyglycolic acid, styrene resin, acrylic styrene copolymer resin, polycarbonate resin, polyester resin, polyvinyl alcohol resin, ethylene vinyl alcohol copolymer resin, thermoplastic elastomer, vinyl chloride resin, and silicon resin.

5. The culture chamber according to any one of Claims 1 to 4, wherein a functional group is formed on the recesses by a surface modification treatment method of any one of plasma treatment, glass coating, corona discharge, and UV ozonation, or a combination thereof and the treatment is performed so that a water contact angle becomes 45 degrees or less.

6. The culture chamber according to any one of Claims 1 to 5, wherein a hydrophilic polymer chain that inhibits cell adhesion is immobilized in the recesses.

7. The culture chamber according to any one of Claims 1 to 6, wherein a phospholipid or a phospholipid-polymer complex is immobilized in the recesses.

8. The culture chamber according to any one of Claims 1 to 7, wherein each of the recesses has a cell non-adhesive surface on which at least one polymer of a hydrophilic polymer chain that inhibits cell adhesion, and a phospholipid, or a phospholipid-polymer complex is immobilized after a functional group is formed in the recesses by a surface modification treatment method of any one of plasma treatment, glass coating, corona discharge, and UV ozonation, or a combination thereof and the treatment is performed so that a water contact angle becomes 45 degrees or less.

9. The culture chamber according to Claim 8, wherein the hydrophilic polymer chain is poly(hydroxyethyl methacrylate).

10. The culture chamber according to Claim 9, wherein an average molecular weight of the poly(hydroxyethyl methacrylate) is 100,000 or more.

11. A culture method that uses a culture chamber according to any one of Claims 1 to 10, the culture method comprising:

dispersing cells into a medium, a total number of the cells being equal to or greater than a number N of the recesses of the culture chamber and equal to or less than a number obtained by multiplying the number N of the recesses by a value obtained by dividing a volume V1 of a space defined by each of the recesses by a volume V2 of cells to be seeded; and

adding the medium to the culture chamber.

12. The culture method according to Claim 11, wherein one spheroid is formed in one space defined by each of the recesses.

13. The culture method according to Claim 12, wherein a spheroid is formed in the space and the spheroid is allowed to grow.

14. The culture method according to Claim 12 or 13, wherein a spheroid is formed in the space and is differentiated and induced.

15. The culture method according to any one of Claims 11 to 14, wherein 60% or more of a total number of spheroids formed in the culture chamber have a diameter in a range of ±5% of an average spheroid diameter.

16. The culture method according to any one of Claims 11 to 15, wherein cells in the recesses are recovered by agitating the medium.

17. The culture method according to Claim 16, wherein the agitation of the medium is done by any one of the following means: agitation of the medium by shaking the culture chamber; agitation of the medium by sucking and discharging the medium; agitation of the medium by disposing a stirring blade in the culture chamber; and agitation of the medium by placing a stirrer in the culture chamber, or a combination thereof.

18. The culture method according to any one of Claims 11 to 17, wherein the medium is replaced at least once, and 20% or more of the medium is replaced.

19. A culture method for cell seeding, cell culture, replacement of a medium, and harvesting of cells by using a culture chamber according to any one of Claims 1 to 10, the culture method comprising the steps of:

a) dispersing cells into a medium, the number of the cells being equal to or greater than a number n of recesses of the culture chamber and equal to or less than a number obtained by multiplying the number n of the recesses by a value obtained by dividing a volume V of each of the recesses by a volume v of cells to be seeded, and adding the medium to the culture chamber;

b) culturing the cells in the culture chamber for 12 hours or more to form a spheroid;

c) sucking 20% or more of the medium and then injecting the same amount of fresh medium;

d) repeating the steps a) to c) a plurality of times to allow the spheroid to grow;

e) allowing the spheroid to grow to a desired size and then agitating the medium to suspend the cells within the recesses in the medium; and

f) sucking the medium including the cells by a suction machine to recover the cells.

CLAIMS

1. A culture chamber comprising a plurality of recesses each formed of a bottom portion and an opening portion, wherein

the bottom portion has one of a hemispherical shape, a truncated cone shape, a shape formed using a part of a spherical shape, and a linear shape,

the opening portion is defined by a wall that surrounds an area from a boundary between the opening portion and the bottom portion to an end of each of the recesses, the wall having a taper angle in a range from 1 degree to 20 degrees,

an equivalent diameter of the boundary is in a range from 50 μm to 1 mm and a depth from a bottom of the bottom portion to the upper end of each of the recesses is in a range from 0.6 or more times to 3 or less times the equivalent diameter, wherein the equivalent diameter is a diameter of an inscribed circle that is inscribed at the boundary, and

the wall defining the opening portion forms a surface continuous to the bottom portion, and an inclination of the continuous surface changes at the boundary.

2. A culture chamber comprising a plurality of recesses each formed of a bottom portion and an opening portion, wherein

the bottom portion has one of a hemispherical shape and a truncated cone shape,

the opening portion is defined by a wall that surrounds an area from a boundary between the opening portion and the bottom portion to an end of each of the recesses, the wall having a taper angle in a range from 1 degree to 20 degrees,

an equivalent diameter of the boundary is in a range from 50 μm to 2 mm and a depth from a bottom of the bottom portion to the upper end of each of the recesses is in a range from 0.6 or more times to 1.2 or less times the equivalent diameter, wherein the equivalent diameter is a diameter of an inscribed circle that is inscribed at the boundary, and

the wall defining the opening portion forms a surface continuous to the bottom portion, and an inclination of the continuous surface changes at the boundary.

3. The culture chamber according to Claim 1 or 2, wherein the end of each of the recesses has one of a hemispherical shape, a trapezoidal shape, and an inverted triangular shape.

4. The culture chamber according to any one of Claims 1 to 3, wherein an area between two adjacent recesses is flat and a distance between the two recesses is in a range from 5 μm to 50 μm.

5. The culture chamber according to any one of Claims 1 to 4, wherein the culture chamber is a resin molding formed of one or a combination of two or more selected from the group consisting of acrylic resin, polylactic acid, polyglycolic acid, styrene resin, acrylic styrene copolymer resin, polycarbonate resin, polyester resin, polyvinyl alcohol resin, ethylene vinyl alcohol copolymer resin, thermoplastic elastomer, vinyl chloride resin, and silicon resin.

6. The culture chamber according to any one of Claims 1 to 5, wherein a functional group is formed on the recesses by a surface modification treatment method of any one of plasma treatment, glass coating, corona discharge, and UV ozonation, or a combination thereof and the treatment is performed so that a water contact angle becomes 45 degrees or less.

7. The culture chamber according to any one of Claims 1 to 6, wherein a hydrophilic polymer chain that inhibits cell adhesion is immobilized in the recesses.

8. The culture chamber according to any one of Claims 1 to 7, wherein a phospholipid or a phospholipid-polymer complex is immobilized in the recesses.

9. The culture chamber according to any one of Claims 1 to 8, wherein each of the recesses has a cell non-adhesive surface on which at least one polymer of a hydrophilic polymer chain that inhibits cell adhesion, and a phospholipid, or a phospholipid-polymer complex is immobilized after a functional group is formed in the recesses by a surface modification treatment method of any one of plasma treatment, glass coating, corona discharge, and UV ozonation, or a combination thereof and the treatment is performed so that a water contact angle becomes 45 degrees or less.

10. The culture chamber according to Claim 9, wherein the hydrophilic polymer chain is poly(hydroxyethyl methacrylate).

11. The culture chamber according to Claim 10, wherein an average molecular weight of the poly(hydroxyethyl methacrylate) is 100,000 or more.

12. The culture chamber according to Claims 1 to 11, wherein the plurality of recesses are formed in the bottom of the culture chamber.

13. A culture method that uses a culture chamber according to any one of Claims 1 to 12, the culture method comprising:

dispersing cells into a medium, a total number of the cells being equal to or greater than a number N of the recesses of the culture chamber and equal to or less than a number obtained by multiplying the number N of the recesses by a value obtained by dividing a volume V1 of a space defined by each of the recesses by a volume V2 of cells to be seeded; and

adding the medium to the culture chamber.

14. The culture method according to Claim 13, wherein one spheroid is formed in one space defined by each of the recesses.

15. The culture method according to Claim 14, wherein a spheroid is formed in the space and the spheroid is allowed to grow.

16. The culture method according to Claim 14 or 15, wherein a spheroid is formed in the space and is differentiated and induced.

17. The culture method according to any one of Claims 13 to 16, wherein cells in the recesses are recovered by agitating the medium.

18. The culture method according to Claim 17, wherein the agitation of the medium is done by any one of the following means: agitation of the medium by shaking the culture chamber; agitation of the medium by sucking and discharging the medium; agitation of the medium by disposing a stirring blade in the culture chamber; and agitation of the medium by placing a stirrer in the culture chamber, or a combination thereof.

19. The culture method according to any one of Claims 13 to 18, wherein the medium is replaced at least once, and 20% or more of the medium is replaced.

20. A culture method for cell seeding, cell culture, replacement of a medium, and harvesting of cells by using a culture chamber according to any one of Claims 1 to 12, the culture method comprising the steps of:

a) dispersing cells into a medium, the number of the cells being equal to or greater than a number n of recesses of the culture chamber and equal to or less than a number obtained by multiplying the number n of the recesses by a value obtained by dividing a volume V of each of the recesses by a volume v of cells to be seeded, and adding the medium to the culture chamber;

b) culturing the cells in the culture chamber for 12 hours or more to form a spheroid;

c) sucking 20% or more of the medium and then injecting the same amount of fresh medium;

d) repeating the steps a) to c) a plurality of times to allow the spheroid to grow;

e) allowing the spheroid to grow to a desired size and then agitating the medium to suspend the cells within the recesses in the medium; and

f) sucking the medium including the cells by a suction machine to recover the cells.