CLAIMS

1. A method of culturing mesenchymal stromal cells for use in therapy comprising the steps of:

(i) culturing a sample of mesenchymal stromal cells in a first cell culture medium comprising a first serum, first serum substitute and/or first serum fraction for sufficient time to enable the mesenchymal stromal cells to achieve a desired level of expansion; and

(ii) prior to harvesting the mesenchymal stromal cells for use in therapy said first cell culture medium is replaced with a second cell culture medium comprising a second serum, a second serum substitute and/or second serum fraction which serum or serum fraction has the same species origin as the mesenchymal stromal cells being cultured or the second serum substitute replicates the same species origin and the mesenchymal stromal cells are cultured for less than 48 hours before harvesting for use.

2. A method of culturing mesenchymal stromal cells for use in therapy comprising the steps of:

(i) culturing a sample of mesenchymal stromal cells in a first cell culture medium comprising a first serum, first serum substitute and/or first serum fraction for sufficient time to enable the cells to achieve a desired level of expansion; and

(ii) prior to harvesting the mesenchymal stromal cells for use in therapy said first cell culture medium is replaced with a second cell culture medium comprising a second serum, a second serum substitute and/or second serum fraction which serum or serum fraction has the same species origin as the mesenchymal stromal cells being cultured or the second serum substitute replicates the same species origin and the cells are cultured for between about 24 hours and about 36 hours before the mesenchymal stromal cells are harvested for use in cell therapy.

3. A method according to claim 1 or 2, wherein the first serum, first serum substitute and/or first serum fraction is selected from the group consisting of fetal calf serum (FCS), heat inactivated fetal calf serum (HIFCS), platelet lysate, serum albumin and growth factors.

4. A method according to any one of claims 1 to 3, wherein the sample of mesenchymal stromal cells are isolated from a human, a cameline, an equine, a canine or a feline.

5. A method according to any one of claims 1 to 4, wherein the first medium in step (i) comprises a base medium selected from the group consisting of DMEM (high or low glucose), Eagle's basal medium (MEM), Ham's F10 medium (F10), Ham's F-12 medium

(F12), Iscove's modified Dulbecco's medium, M-199, Mesenchymal Stem Cell Growth Medium (MSCGM), Liebovitz's L-15 medium, MCDB, DMEM/F12, RPMI 1640, advanced DMEM (Gibco), DMEM/MCDB201 (Sigma), and CELL-GRO FREE or a combination thereof, preferably the base medium is DMEM.

6. A method according to claim 5, wherein the base medium is supplemented with heat inactivated fetal calf serum (HIFCS).

7. A method according to any one of claims 1 to 6, wherein the sample of mesenchymal stromal cells in step (i) comprises between 1.0×10^3 cells per cm^2 of culture vessel and 1.0×10^{10} cells per cm^2 of culture vessel.

8. A method according to any one of claims 1 to 7, wherein the sample of mesenchymal stromal cells in step (i) are cultured until the desired level of expansion of the cells has been reached which is approximately 70-85% confluent.

9. A method according to any one of claims 1 to 8, wherein the second medium in step (ii) comprises a base medium selected from the group consisting of DMEM (high or low glucose), Eagle's basal medium (MEM), Ham's F10 medium (F10), Ham's F-12 medium (F12), Iscove's modified Dulbecco's medium, M-199, Mesenchymal Stem Cell Growth Medium (MSCGM), Liebovitz's L-15 medium, MCDB, DMEM/F12, RPMI 1640, advanced DMEM (Gibco), DMEM/MCDB201 (Sigma), and CELL-GRO FREE or a combination thereof, optionally the base medium is DMEM phenol red-free.

10. A method according to claim 9, wherein the base medium is supplemented with human serum or human serum albumin, optionally the human serum or human serum albumin is about 2% v/v.

11. A method of culturing human mesenchymal stromal cells for use in cell therapy comprising the steps of:

(i) culturing mesenchymal stromal cells in a first cell culture medium comprising DMEM supplemented with heat inactivated fetal calf serum (HIFCS) until said cells reach about 70-85% confluence; and

(ii) prior to harvesting cells for use in therapy said first cell culture medium is removed and replaced with a second cell culture medium comprising DMEM phenol red-free and about 2% human serum albumin and the cells are then cultured for a further about 24 hours to about 36 hours.

12. A method of culturing human mesenchymal stromal cells for use in cell therapy comprising the steps of:

(i) culturing between 1.0 x 10^3 cells per cm^2 of culture vessel and 1.0 x 10^{10} cells per cm^2 of culture vessel of mesenchymal stromal cells in a first cell culture medium comprising DMEM supplemented with heat inactivated fetal calf serum (HIFCS) until said cells reach about 70-85% confluence; and

(ii) prior to harvesting cells for use in therapy said first cell culture medium is removed and replaced with a second cell culture medium comprising DMEM phenol red-free and about 2% human serum albumin and the cells are then cultured for a further about 24 hours to about 36 hours.

13. A method for preparing mesenchymal stromal cells for allogeneic cell therapy in a patient comprising the steps of:

(i) providing a sample of mesenchymal stromal cells from a healthy donor;

(ii) culturing said sample of mesenchymal stromal cells in a first cell culture medium comprising a first serum, first serum substitute and/or first serum fraction for sufficient time to enable the mesenchymal stromal cells to achieve a desired level of expansion; and

(iii) prior to harvesting mesenchymal stromal cells for use in therapy said first cell culture medium is replaced with a second cell culture medium comprising a second serum, a second serum substitute and/or second serum fraction which serum or serum fraction has the same species origin as the mesenchymal stromal cells being cultured or the second serum substitute replicates the same species origin and the mesenchymal stromal cells are cultured for about 24 hours to about 36 hours; and

(iv) harvesting said mesenchymal stromal cells.

14. A method according to claim 13, wherein the first serum, first serum substitute and/or first serum fraction is selected from the group consisting of fetal calf serum (FCS), heat inactivated fetal calf serum (HIFCS), platelet lysate, serum albumin and growth factors.

15. A method according to any one of claims 11 to 14, wherein the first medium in step (ii) comprises a base medium selected from the group consisting of DMEM (high or low glucose), Eagle's basal medium (MEM), Ham's F10 medium (F10), Ham's F-12 medium (F12), Iscove's modified Dulbecco's medium, M-199, Mesenchymal Stem Cell Growth Medium (MSCGM), Liebovitz's L-15 medium, MCDB, DMEM/F12, RPMI 1640, advanced DMEM (Gibco), DMEM/MCDB201 (Sigma), and CELL-GRO FREE or a combination thereof, optionally the base medium is DMEM.

16. A method according to any one of claims 11 to 15, wherein the second medium in step (iii) comprises the base medium DMEM phenol red-free and is supplemented with human serum or human serum albumin, optionally the human serum or human serum albumin is about 2%.