CLAIMS

What is claimed is:

1. A method of preparing an oocyte for in vitro fertilization (IVF) or artificial insemination, the method comprising transferring a composition comprising i) oogonial stem cell (OSC) mitochondria, or ii) mitochondria obtained from a progeny of an OSC, into an autologous oocyte, thereby preparing said oocyte for IVF or artificial insemination.

2. The method of claim 1, wherein the OSC is an isolated non-embryonic stem cell that is mitotically competent and expresses Vasa, Oct-4, Dazl, Stella and optionally a stage-specific embryonic antigen.

3. The method of claim 1, wherein the OSC is obtained from ovarian tissue.

4. The method of claim 1, wherein the OSC is obtained from non-ovarian tissue.

5. The method of claim 4, wherein the non-ovarian tissue is blood.

6. The method of claim 4, wherein the non-ovarian tissue is bone marrow.

7. The method of claim 1, wherein the composition comprising an OSC mitochondria, or mitochondria obtained from a progeny of an OSC, is the cytoplasm of said cells without a nucleus.

8. The method of claim 1, wherein the composition comprising i) OSC mitochondria, or ii) mitochondria obtained from a progeny of an OSC, is a purified preparation of mitochondria.

9. A composition comprising isolated OSC mitochondria or mitochondria obtained from a progeny of an OSC.

10. The composition of claim 9, wherein the composition is at least about 85%, 90%, 95% free of cells or non-functional mitochondria.

11. A composition comprising at least one isolated mitochondrion obtained from an OSC or at least one progeny of an OSC.

12. An oocyte prepared according to the method of claim 1.

13. An oocyte comprising exogenous, autologous OSC mitochondria or mitochondria obtained from the progeny of an OSC.

14. A method of in vitro fertilization, said method comprising the steps of:

a) obtaining a composition comprising i) mitochondria from an OSC, or ii) mitochondria obtained from a progeny of an OSC;

b) transferring the composition into an isolated, autologous oocyte; and

c) fertilizing the autologous oocyte in vitro to form a zygote.

15. The method of claim 14, further comprising transferring the zygote, or a preimplantation stage embryo derived from said zygote, into the uterus or oviduct of a female subject.

16. A method of isolating a population of functional mitochondria from at least one OSC, or at least one progeny of an OSC, said method comprising the steps of incubating a composition comprising at least one OSC, or at least one progeny of an OSC, with a fluorescent mitochondrial tracking probe selected from the group consisting of a non-oxidation dependent probe, accumulation dependent probe and reduced oxidative state probe under conditions sufficient to bind the probe to the functional mitochondria and sorting the functional mitochondria using fluorescence-activated cell sorting, thereby isolating the population of functional mitochondria from at least one OSC, or at least one progeny of an OSC.

17. The method of claim 16, wherein non-functional mitochondria are excluded from the population of functional mitochondria.

18. A method of identifying a population of functional mitochondria obtained from at least one OSC, or at least one progeny of an OSC, said method comprising the steps of:

a) incubating a composition comprising at least one OSC, or at least one progeny of an OSC, with a fluorescent reduced oxidative state probe and a fluorescent accumulation dependent probe under conditions sufficient to bind the fluorescent reduced oxidative state probe to functional mitochondria in the composition and bind the fluorescent accumulation dependent probe to total mitochondria in the composition;

b) obtaining a composition comprising the functional mitochondria using fluorescence-activated cell sorting, wherein said composition excludes non-functional mitochondria;

c) determining the amount of functional mitochondria and the amount of total mitochondria; and

d) calculating the ratio of functional mitochondria to total mitochondria; and

e) determining whether the ratio is greater than about 0.02, thereby identifying a population of functional mitochondria obtained from at least one OSC, or at least one progeny of an OSC.

19. The method of claim 18, wherein the fluorescent accumulation dependent probe fluoresces in the green spectrum.

20. The method of claim 18, wherein the fluorescent reduced oxidative state probe fluoresces in the red spectrum.

21. A composition comprising the functional mitochondria obtained according to steps a) and b) of claim 18.

22. A kit comprising the composition of any one of claims 9 to 11 and instructions for use.

23. A method for increasing the ATP-generating capacity of an oocyte, said method comprising the steps of:

a) obtaining a composition comprising mitochondria obtained from at least one OSC or at least one progeny of an OSC that is autologous to the oocyte; and

b) injecting the composition of mitochondria into the oocyte.

24. The method of claim 1, 14 or 23, wherein the composition comprises $1x10^3$ to $5x10^4$ mitochondria.

25. The method of claim 1, 14 or 23, wherein the OSC or progeny of an OSC produces at least 5-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

26. The method of claim 25, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

27. The method of claim 1, 14 or 23, wherein the OSC or progeny of an OSC produces at least 10-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

28. The method of claim 27, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

29. The method of claim 1, 14, or 23, wherein the OSC or progeny of an OSC produces at least 50-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

30. The method of claim 29, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

31. The method of claim 1, 14, or 23, wherein the OSC or progeny of an OSC produces at least 100-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

32. The method of claim 31, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

33. The method of claim 1, 14 or 23, wherein the oocyte is obtained from a human female of advanced maternal age.

34. The method of claim 1, 14 or 23, wherein the oocyte is obtained from a human female with low ovarian reserve.

35. The method of claim 1, 14 or 23, wherein the composition comprises mitochondria that have been isolated by centrifugation.

36. The method of claim 1, 14 or 23, wherein the composition comprises mitochondria that have been isolated by mitochondrial membrane potential-dependent cell sorting.

37. The method of claim 23, wherein the at least one OSC is obtained from ovarian tissue.

38. The method of claim 23, wherein the at least one OSC is obtained from a non-ovarian tissue.

39. The method of claim 38, wherein the non-ovarian tissue is blood.

40. The method of claim 38, wherein the non-ovarian tissue is bone marrow.

41. The method of claim 23, wherein the composition comprising mitochondria obtained from at least one OSC or at least one progeny of an OSC, is the cytoplasm of said cells without a nucleus.

42. The method of claim 23, wherein the composition comprising mitochondria obtained from at least one OSC or at least one progeny of an OSC is a purified preparation of mitochondria.

43. The composition of claim 9 or 11, wherein the composition comprises $1x10^3$ to $5x10^4$ mitochondria.

44. The composition of claim 9 or 11, wherein the OSC or progeny of an OSC produces at least 5-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

45. The composition of claim 44, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

46. The composition of claim 9 or 11, wherein the OSC or progeny of an OSC produces at least 10-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

47. The composition of claim 46, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

48. The composition of claim 9 or 11, wherein the OSC or progeny of an OSC produces at least 50-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

49. The composition of claim 48, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

50. The composition of claim 9 or 11, wherein the OSC or progeny of an OSC produces at least 100-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

51. The composition of claim 50, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

52. The composition of claim 9 or 11 wherein the OSC or progeny of an OSC is obtained from a human female of advanced maternal age.

53. The composition of claim 9 or 11 wherein the OSC or progeny of an OSC is obtained from a human female with low ovarian reserve.

54. The composition of claim 9 or 11, wherein the composition comprises mitochondria that have been isolated by centrifugation.

55. The composition of claim 9 or 11, wherein the composition comprises mitochondria that have been isolated by mitochondrial membrane potential-dependent cell sorting.

56. An oocyte prepared according to the method of claim 23.

57. A composition of mitochondria obtained from at least one OSC or at least one progeny of an OSC, wherein the composition comprises a population of mitochondria in which greater than about 75%, 85%, 90%, or about 99% of the mitochondria are high ATP-generating capacity mitochondria.

CLAIMS

What is claimed is:

1. A method of preparing an oocyte for in vitro fertilization (IVF) or artificial insemination, the method comprising transferring a composition comprising i) oogonial stem cell (OSC) mitochondria, or ii) mitochondria isolated from a progeny of an OSC, the OSC isolated from ovarian tissue, into an autologous oocyte, thereby preparing said oocyte for IVF or artificial insemination, wherein the OSC is an isolated non-embryonic stem cell that is mitotically competent and expresses Vasa, Oct-4, Dazl, Stella and optionally a stage-specific embryonic antigen.

2. A method of in vitro fertilization, said method comprising the steps of: a) providing a composition comprising i) mitochondria isolated from an OSC, or ii) mitochondria isolated from a progeny of an OSC, the OSC isolated from ovarian tissue; b) transferring the composition into an isolated, autologous oocyte; and c) fertilizing the autologous oocyte in vitro to form a zygote, wherein the OSC is an isolated non-embryonic stem cell that is mitotically competent and expresses Vasa, Oct-4, Dazl, Stella and optionally a stage-specific embryonic antigen, and wherein the step of transferring the composition and fertilizing the autologous oocyte are performed simultaneously in vitro.

3. A method for increasing the ATP-generating capacity of an oocyte, said method comprising the steps of: a) providing a composition comprising mitochondria isolated from at least one OSC or at least one progeny of an OSC that is autologous to the oocyte, the at least one OSC isolated from ovarian tissue; and b) injecting the composition comprising mitochondria into the oocyte, wherein the OSC is an isolated non-embryonic stem cell that is mitotically competent and expresses Vasa, Oct-4, Dazl, Stella and optionally a stage-specific embryonic antigen, and wherein the composition comprises mitochondria that have been isolated by centrifugation or by mitochondrial membrane potential-dependent cell sorting, and wherein the OSC or progeny of an OSC produces at least 5-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

4. The method of claim 3, wherein the composition comprises 1×10^3 to 5×10^4 mitochondria.

5. The method of claim 3, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

6. The method of claim 1, wherein the composition comprises 1×10^3 to 5×10^4 mitochondria.

7. The method of claim 1, wherein the OSC or progeny of an OSC produces at least 5-fold more ATP per fg mtDNA than an ovarian somatic cell or mesenchymal stem cell.

8. The method of claim 7, wherein the ovarian somatic cell or mesenchymal stem cell is autologous.

9. The method of claim 1, wherein the OSC or progeny of an OSC is isolated from a human female of advanced maternal age.

10. The method of claim 1, wherein the composition comprises mitochondria that have been isolated by centrifugation.

11. The method of claim 1, wherein the composition comprises mitochondria that have been isolated by mitochondrial membrane potential-dependent cell sorting.