in the population have intact inner and outer membranes; and/or at least about 60%, at least about 65%, at least about 70%, at least about 75%, at least about 80%, at least about 85%, at least about 90%, or at least about 95% of the mitochondria in the population are polarized as measured by a fluorescence indicator. In examples, the fluorescence indicator is selected from the group consisting of positively charged dyes such as JC-1, tetramethylrhodamine methyl ester (TMRM), and tetramethylrhodamine ethyl ester (TMRE).

[0005a] In one aspect, the present disclosure provides a population of isolated human mitochondria, wherein: (i) at least 80% of the mitochondria in the population have intact inner and outer membranes, (ii) at least 80% of the mitochondria in the population are polarized as measured by a fluorescence indicator, and/or (iii) at least 80% of the mitochondria in the population maintain functional capability in an extracellular environment, wherein the mitochondria the mitochondria exhibit decreased association with mitochondria-associated membrane (MAM) as measured by glucose regulated protein 75 (GRP75) expression ; wherein the decreased association is a decrease of at least 30% relative to the association with MAM of mitochondria in a cell; wherein the mitochondria are isolated from cells treated with a surfactant at a concentration below the critical micellar concentration (CMC); and wherein the mitochondria are free from the effects of physical disruption or chemical destruction by surfactants during their collection process, come in contact with no surfactants or the surfactant at a concentration far below the CMC that cannot be removed from the first solution and cannot damage the mitochondria; wherein the population of isolated mitochondria provided herein have a feature of maintaining functional capability even when exposed to a high calcium (Ca2+) environment of 4 mg/dL to 6 mg/dL , wherein the functional capability is the membrane potential; and wherein a physical stimulus can be added to the cells only such that the lipid bilayer of the mitochondrion does not cause mechanical disruption, in order to enhance the recovery of the mitochondria from the cells.

[0006] In examples, the present disclosure provides a population of isolated mitochondria, wherein at least about 60%, at least about 65%, at least about 70%, at least about 75%, at least about 80%, at least about 85%, at least about 90%, or at least about 95% of the mitochondria in the population maintain functional capability (e.g., are polarized) in an extracellular environment. In examples, the functional capability in an extracellular environment is measured by a fluorescence indicator of membrane potential. In examples, the fluorescence indicator is selected from the group consisting of positively charged dyes such as JC-1, TMRM, and TMRE. In examples, the extracellular environment may comprise a total calcium concentration of about 4 mg/dL to about 12 mg/dL, or about 1 mmol/L (1000 μM) to about 3 mmol/L (3000 μM). For example, in examples, the extracellular environment comprises a concentration of total calcium of about 8 mg/dL to about 12 mg/dL, or about 2 mmol/L (2000 μM) to about 3 mmol/L (3000 μM). In examples, the extracellular environment comprises a concentration of free or active calcium of about 4 mg/dL to about 6 mg/dL, or about 1 mmol/L

(1000 μM) to about 1.5 mmol/L (1500 μM). In examples, the population of mitochondria maintain functional capability in an environment having a higher calcium concentration compared to the calcium environment in a cell.

[0007] In examples, provided herein is a population of isolated mitochondria wherein at least about 60%, at least about 65%, at least about 70%, at least about 75%, at least about 80%, at least about 85%, at least about 90%, or at least about 95% of the mitochondria in the population are not undergoing dynamin-related protein 1 (drp1) – dependent division. In examples, provided herein is a population of isolated mitochondria having inner and outer membranes, wherein the inner membranes of the mitochondria comprise densely folded cristae.

[0008] In examples, provided herein is a population of isolated mitochondria wherein at least about 60%, at least about 65%, at least about 70%, at least about 75%, at least about 80%, at least about 85%, at least about 90%, or at least about 95% of the mitochondria in the population have a substantially non-filamentous, non-branched structure or shape. For example, in examples, the mitochondria provided herein appear as round, dot-like, globular,

at least about 85%, at least about 90%, or at least about 95% of the mitochondria in the population capable of being incorporated into cells and/or co-localization with and/or fusion with endogenous mitochondria in cells after the mitochondria have been stored and/or undergone one or more freeze-thaw cycle. In examples, the method of storing and thawing the population of isolated mitochondria provided herein comprises storing the population at about -20°C ± 3°C, about -80°C ± 3°C, or colder (e.g., in liquid nitrogen), and then thawing the mitochondria at about 20°C ± 3°C or colder, wherein the mitochondria are thawed within about 5 minutes, about 4 minutes, about 3 minutes, about 2 minutes, or about 1 minute. In particular examples, the population of mitochondria is thawed within about 1 minute. Thus, in examples, the mitochondria provided herein are markedly different from mitochondria isolated via conventional methods at least in that they are capable of being incorporated into cells and/or co-localization with and/or fusion with endogenous mitochondria in cells, whereas mitochondria isolated by conventional methods are incapable of or exhibit vastly reduced ability to being incorporated into cells and/or co-localize with and/or fuse with endogenous mitochondria in cells. In examples, the co-localized isolated mitochondria can form a filamentous structure, a network structure, and/or a mesh-like structure.

[0014] In another aspect, the present disclosure provides compositions comprising the isolated mitochondria provided herein. The compositions, in examples, further comprise one or more pharmaceutically acceptable carrier.

[0014a] In another aspect, the present disclosure provides a formulation comprising the composition as disclosed herein and a pharmaceutically acceptable carrier.

In examples, the present disclosure provides methods for isolating mitochondria from cells which differs from conventionally known methods and results in mitochondria having the superior functionality and other characteristics provided herein. In another aspect, the present disclosure provides a method for isolating human mitochondria from human cells, the method comprising: (i) treating human cells in a first solution with a surfactant at a concentration below the critical micellar concentration for the surfactant, (ii) removing the surfactant to form a second solution, (iii) incubating the cells in the second solution and allow mitochondria inside the cells to be released into the second solution, and (iv) recovering mitochondria from the second solution, wherein is a surfactant-free solution or a solution with a negligible and/or undetectable amount of surfactant, and wherein a physical stimulus can be added to the cells only such that the lipid bilayer of the mitochondrion does not cause mechanical disruption, in order to enhance the recovery of the mitochondria from the cells. In another example, the present disclosure provides the method for isolating mitochondria from cells comprises treating cells in a first solution with a surfactant at a concentration below the critical micelle concentration (CMC) for the surfactant, removing the surfactant to form a second solution, incubating the cells in the second solution, and recovering mitochondria from the second solution. In examples the concentration of the surfactant in the first solution is about 50% or less of the CMC for the surfactant. For example, in examples, the concentration of the surfactant in the first solution is about 40% or less, about 30% or less, about 20% or less, or about 10% or less of the CMC for the surfactant.

[0015] In examples, the surfactant is a non-ionic surfactant. In examples, the surfactant is selected from the group consisting of Triton-X 100, Triton-X 114, Nonidet P-40, n-Dodecyl-D-maltoside, Tween-20, Tween-80, saponin and digitonin. In examples, the surfactant is saponin or digitonin. In examples, the concentration of the surfactant is less than about 400

detaching the cell from the surface. In examples, the step of recovering the mitochondria from the second solution comprises collecting the supernatant to recover the isolated mitochondria, which can be optionally followed by washing the remaining cell on the culture surface with the second solution or another second solution to combine it with the supernatant.

[0022] In examples, the methods provided herein further comprise freezing the isolated mitochondria. In examples, the methods comprise freezing the mitochondria in a buffer comprising a cryoprotectant (e.g., glycerol). In examples, the methods comprise freezing the mitochondria in the buffer in liquid nitrogen. In examples, the methods further comprise thawing the mitochondria after freezing. In examples, the methods for thawing the mitochondria comprise rapidly thawing the mitochondria, for example, within about 5 minutes or within about 1 minute. In examples, the mitochondria are thawed in a warm bath having a temperature of about 20°C ± 3°C to about 37°C ± 3°C. In examples, the mitochondria are thawed at a temperature of about 20°C ± 3°C or colder.

[0023] In another aspect, the present disclosure provides a population of isolated mitochondria obtained by the method provided herein. In examples, the method provided herein is the "iMIT" method and the mitochondria obtained by this method are referred to herein as "Q" mitochondria. In examples, the present disclosure provides compositions and/or formulations comprising the population of isolated mitochondria obtained by the methods provided herein.

[0024a] In another aspect, the present disclosure provides use of a population of isolated mitochondria as disclosed herein or a composition as disclosed herein or a formulation as disclosed herein in the manufacture of a medicament for treating a disease or disorder, wherein the disease or disorder is selected from the group consisting of diabetes (Type I and Type II), metabolic disease, ocular disorders associated with reduced mitochondrial function, hearing loss, mitochondrial toxicity associated with therapeutic agents, cardiotoxicity associated with chemotherapy or other therapeutic agents, a reduced mitochondrial function disorder, and migraine. In yet another aspect, the present disclosure provides use of a population of isolated mitochondria as disclosed herein or a composition as disclosed herein or a formulation as disclosed herein in the manufacture of a medicament for treating a disease or disorder associated with reduced mitochondrial function.

[0024] In examples, the present disclosure provides methods for treating or preventing a disease or disorder associated with mitochondrial dysfunction, the method comprising contacting cells of a subject with a population of isolated mitochondria provided herein, e.g., the Q mitochondria. In examples, the disease or disorder is an ischemia-related disease or disorder. For example, in examples, the ischemia-related disease or disorder is selected from the group consisting of cerebral ischemic reperfusion, hypoxia ischemic encephalopathy, acute coronary syndrome, a myocardial infarction, a liver ischemia-reperfusion injury, an ischemic injury-compartmental syndrome, a blood vessel blockage, wound healing, spinal cord injury, sickle cell disease, and reperfusion injury of a transplanted organ. In examples, the disease or disorder is a genetic disorder. In examples, the disease or disorder is a cancer, cardiovascular disease, ocular disorder, otic disorder, autoimmune disease, inflammatory disease, or fibrotic disorder. In examples, the disease is acute respiratory distress syndrome (ARDS). In examples, the disease or disorder is an aging disease or disorder, or a condition

associated with aging. In examples, the disease or disorder is pre-eclampsia or intrauterine growth restriction (IUGR).

[0025] In examples, the present disclosure provides methods for treating or preventing a disease or disorder provided herein, wherein the method comprises administering the population of isolated mitochondria or the composition to a subject in need thereof. In examples, the route of administration of the isolated mitochondria is via an intravenous, intra-arterial, intra-tracheal, subcutaneous, intramuscular, inhalation, or intrapulmonary route of administration. In examples, the subject is a mammal, e.g., a human.

[0026] In another aspect, the present disclosure provides an isolated mitochondrion having intact inner and outer membranes, wherein the inner membrane comprises folded cristae, wherein the mitochondrion has been isolated from a cell, wherein the mitochondrion is polarized as measured by a fluorescence indicator, and wherein the mitochondrion is capable of maintaining polarization in an extracellular environment. In another aspect, the present disclosure provides a composition comprising the isolated mitochondrion as disclosed herein. In examples, the present disclosure provides an isolated mitochondrion having intact inner and outer membranes, wherein the inner membrane comprises folded cristae, wherein the mitochondrion has been isolated from a cell, wherein the mitochondrion is polarized as measured by a fluorescence indicator (e.g., JC-1, TMRM, or TMRE), and wherein the mitochondrion is capable of maintaining polarization in an extracellular environment. In examples, the folded cristae are densely folded cristae. In examples, the mitochondrion has a substantially non-filamentous shape. In examples, the mitochondrion comprises voltage dependent anion channels (VDAC) on its surface that are associated with tubulin. For example, in examples, the isolated mitochondrion comprises dimeric tubulin associated with VDAC on the surface. In examples, the tubulin comprises at least α-tubulin. In examples, the tubulin is a heterodimer comprising α-tubulin and β-tubulin. In examples, the tubulin is a homodimer. In examples, the isolated mitochondrion exhibits decreased association with MAM as measured by GRP75 expression. For example, in examples, isolated mitochondrion exhibits about 70%, about 60%, about 50%, about 40%, about 30%, or less association with MAM when compared to mitochondrion that is present in a cell (i.e. has not been isolated), and/or a mitochondrion that has been obtained by a conventional method of isolation such as one involving homogenization and/or high levels of detergent, as further described herein. In examples, the isolated mitochondrion provided herein exhibits a decrease in association with MAM, wherein the decrease is at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, or more relative to the association with MAM of a mitochondrion that is present in a cell (i.e., has not been isolated) and/a mitochondrion that has been isolated by a conventional method of isolation.

[0027] In examples, the isolated mitochondrion provided herein has a membrane potential of between about -30 mV and about -220 mV. In examples, the isolated mitochondrion is non-filamentous in shape. In examples, the isolated mitochondrion is not undergoing drp1-

CLAIMS

[Claim 1] A population of isolated human mitochondria, wherein:

(i) at least 80% of the mitochondria in the population have intact inner and outer membranes,

(ii) at least 80% of the mitochondria in the population are polarized as measured by a fluorescence indicator,

and/or

(iii) at least 80% of the mitochondria in the population maintain functional capability in an extracellular environment,

wherein the mitochondria exhibit decreased association with mitochondria-associated membrane (MAM) as measured by glucose regulated protein 75 (GRP75) expression ; wherein the decreased association is a decrease of at least 30% relative to the association with MAM of mitochondria in a cell;

wherein the mitochondria are isolated from cells treated with a surfactant at a concentration below the critical micellar concentration (CMC); and wherein the mitochondria are free from the effects of physical disruption or chemical destruction by surfactants during their collection process, come in contact with no surfactants or the surfactant at a concentration far below the CMC that cannot be removed from the first solution and cannot damage the mitochondria ;

wherein the population of isolated mitochondria provided herein have a feature of maintaining functional capability even when exposed to a high calcium (Ca2+) environment of 4 mg/dL to 6 mg/dL , wherein the functional capability is the membrane potential; and

wherein a physical stimulus can be added to the cells only such that the lipid bilayer of the mitochondrion does not cause mechanical disruption, in order to enhance the recovery of the mitochondria from the cells.

[Claim 2] The population of isolated mitochondria of claim 1, wherein the functional capability in an extracellular environment of (iii) is measured by a fluorescence indicator of membrane potential.

[Claim 3] The population of isolated mitochondria of claim 1, wherein at least 80% of the mitochondria in the population are not undergoing dynamin-related protein 1 (drp1)-dependent division.

[Claim 4] The population if isolated mitochondria of claim 1, wherein the inner membranes of the mitochondria comprise densely folded cristae.

[Claim 5] The population of isolated mitochondria of any one of claims 1-4, wherein at least 80% of the mitochondria in the population have a non-filamentous shape.

[Claim 6] The population of isolated mitochondria of claim 5, wherein at least 85% of the mitochondria have a non-filamentous shape.

[Claim 7] The population of isolated mitochondria of claim 6, wherein at least 90% of the mitochondria have a non-filamentous shape.

[Claim 8] The population of isolated mitochondria of claim 1, wherein the decreased association is a decrease of at least 50%.

[Claim 9] The population of isolated mitochondria of any one of claims 1-8, wherein

(i) at least 85% of the mitochondria in the population have intact inner and outer membranes,

(ii) at least 85% of the mitochondria in the population are polarized as measured by a fluorescence indicator,

and/or

(iii) at least 85% of the mitochondria in the population maintain functional capability in an extracellular environment.

[Claim 10] The population of isolated mitochondria of any one of claims 1-8, wherein

(i) at least 90% of the mitochondria in the population have intact inner and outer membranes,

(ii) at least 90% of the mitochondria in the population are polarized as measured by a fluorescence indicator,

and/or

(iii) at least 90% of the mitochondria in the population maintain functional capability in an extracellular environment.

[Claim 11] The population of isolated mitochondria of any one of claims 1-10, wherein the fluorescence indicator is selected from the group consisting of JC-1, tetramethylrhodamine methyl ester (TMRM), and tetramethylrhodamine ethyl ester (TMRE).

[Claim 12] The population of isolated mitochondria of any one of claims 1-11, wherein at least 80% of the mitochondria in the population are between 500 nm and 3500 nm in size.

[Claim 13] The population of isolated mitochondria of any one of claims 1-12, wherein the polydispersity index (PDI) of the population is 0.2 to 0.8,

[Claim 14] The population of isolated mitochondria of any one of claims 1-12, wherein the polydispersity index (PDI) of the population is 0.2 to 0.3.

[Claim 15] The population of isolated mitochondria of any one of claims 1-14, wherein the zeta potential of the population of mitochondria is between -15 mV and -40 mV.

[Claim 16] The population of isolated mitochondria of any one of claims 1-15, wherein upon contact of the population of isolated mitochondria with a population of cells, the isolated mitochondria are capable co-localization with endogenous mitochondria in the cells.

[Claim 17] The population of isolated mitochondria of any one of claims 1-15, wherein upon contact of the population of isolated mitochondria with a population of cells, the mitochondria are capable of fusing with endogenous mitochondria in the cells.

[Claim 18] The population of isolated mitochondria of claim 16, wherein the mitochondria are capable of co-localization with the endogenous mitochondria after storage at 4°C for at least 12 hours.

[Claim 19] The population of isolated mitochondria of claim 17, wherein the mitochondria are capable of fusing with the endogenous mitochondria after storage at 4°C for at least 12 hours.

[Claim 20] The population of isolated mitochondria of any one of claims 1-19, wherein at least 70% of the isolated mitochondria in the population are polarized as measured by a fluorescence indicator, after the population undergoes one or more freeze-thaw cycle.

[Claim 21] The population of isolated mitochondria of any one of claims 1-20, wherein the mitochondria are capable of co-localization with endogenous mitochondria, after the population undergoes one or more freeze-thaw cycle.

[Claim 22] The population of isolated mitochondria of claim 20 or 21, wherein the population is frozen at -80°C or colder for at least two weeks and then thawed at 20°C or colder within 5 minutes.

[Claim 23] The population of isolated mitochondria of claim 22, wherein the population is thawed within 1 minute.

[Claim 24] The population of isolated mitochondria of claim 22 or 23, wherein the population is frozen in liquid nitrogen for the at least two weeks.

[Claim 25] The population of isolated mitochondria of claim 24, wherein the population is frozen in liquid nitrogen for at least two months.

[Claim 26] The population of isolated mitochondria of any one of claims 20 to 25, wherein upon contact of the thawed population of mitochondria with a population of cells, the isolated mitochondria in the population are capable of fusion with endogenous mitochondria in the cells.

[Claim 27] A composition comprising the population of isolated mitochondria of any one of claims 1-26.

[Claim 28] A formulation comprising the composition of claim 27 and a pharmaceutically acceptable carrier.

[Claim 29] A method for isolating human mitochondria from human cells, the method comprising:

(i) treating human cells in a first solution with a surfactant at a concentration below the critical micellar concentration for the surfactant,

(ii) removing the surfactant to form a second solution,

(iii) incubating the cells in the second solution and allow mitochondria inside the cells to be released into the second solution, and

(iv) recovering mitochondria from the second solution,

wherein the second solution is a surfactant-free solution or a solution with a negligible and/or undetectable amount of surfactant , and

wherein a physical stimulus can be added to the cells only such that the lipid bilayer of the mitochondrion does not cause mechanical disruption, in order to enhance the recovery of the mitochondria from the cells.

[Claim 30] The method of claim 29, wherein the concentration of the surfactant in the first solution is 50% or less of the critical micelle concentration for the surfactant.

[Claim 31] The method of claim 29 or 30, wherein the concentration of the surfactant in the first solution is 10% or less of the critical micelle concentration for the surfactant.

[Claim 32] The method of any one of claims 29-31, wherein the surfactant is a nonionic surfactant.

[Claim 33] The method of any one of claims 29-32, wherein the surfactant is selected from the group consisting of Triton-X 100, Triton-X 114, Nonidet P-40, n-Dodecyl-D-maltoside, Tween-20, Tween-80, saponin and digitonin.

[Claim 34] A population of isolated mitochondria obtained by the method according to any one of claims 29-33.

[Claim 35] Use of a population of isolated mitochondria according to any one of claims 1-30 or a composition of claim 27 or a formulation of claim 28 in the manufacture of a medicament for treating a disease or disorder, wherein the disease or disorder is selected from the group consisting of diabetes (Type I and Type II), metabolic disease, ocular disorders associated with reduced mitochondrial function, hearing loss, mitochondrial toxicity associated with therapeutic agents, cardiotoxicity associated with chemotherapy or other therapeutic agents, a reduced mitochondrial function disorder, and migraine.

[Claim 36] Use of a population of isolated mitochondria according to any one of claims 1-26 or a composition of claim 27 or a formulation of claim 28 in the manufacture of a medicament for treating a disease or disorder associated with reduced mitochondrial function.

[Claim 37] The use of claim 36, wherein the disease or disorder is selected from the group consisting of mitochondrial myopathy, diabetes and deafness (DAD) syndrome, Barth Syndrome, Leber's hereditary optic neuropathy (LHON), Leigh syndrome, NARP (neuropathy, ataxia, retinitis pigmentosa and ptosis syndrome), myoneurogenic gastrointestinal encephalopathy (MNGIE), MELAS (mitochondrial encephalopathy, lactic acidosis, and stroke-like episodes) syndrome, myoclonic epilepsy with ragged red fibers (MERRF) syndrome, Kearns-Sayre syndrome, and mitochondrial DNA depletion syndrome.

[Claim 38] The use of claim 36, wherein the disease or disorder is an ischemia-related disease or disorder.

[Claim 39] The use of claim 38, wherein the ischemia-related disease or disorder is selected from the group consisting of cerebral ischemic reperfusion, hypoxia ischemic encephalopathy, acute coronary syndrome, a myocardial infarction, a liver ischemia-reperfusion injury, an ischemic injury-compartmental syndrome, a blood vessel blockage, wound healing, spinal cord injury, sickle cell disease, and reperfusion injury of a transplanted organ.

[Claim 40] The use of claim 36, wherein the disease or disorder is a genetic disorder.

[Claim 41] The use of claim 36, wherein the disease or disorder is an aging disease or disorder.

[Claim 42] The use of claim 36, wherein the disease or disorder is a neurodegenerative condition or cardiovascular condition.

[Claim 43] The use of claim 42, wherein the neurodegenerative condition is selected from the group consisting of dementia, Friedrich's ataxia, amyotrophic lateral sclerosis, mitochondrial myopathy, encephalopathy, lactacidosis, stroke (MELAS), myoclonic epilepsy with ragged red fibers (MERFF), epilepsy, Parkinson's disease, Alzheimer's disease, or Huntington's Disease. Exemplary neuropsychiatric disorders include bipolar disorder, schizophrenia, depression, addiction disorders, anxiety disorders, attention deficit disorders, personality disorders, autism, and Asperger's disease.

[Claim 44] The use of claim 42, wherein the cardiovascular condition is selected from the group consisting of coronary heart disease, myocardial infarction, atherosclerosis, high blood pressure, cardiac arrest, cerebrovascular disease, peripheral arterial disease, rheumatic heart disease, congenital heart disease, congestive heart failure, arrhythmia, stroke, deep vein thrombosis, and pulmonary embolism.

[Claim 45] The use of claim 36, wherein the disease or disorder is a cancer, autoimmune disease, inflammatory disease, or fibrotic disorder.

[Claim 46] The use of claim 36, wherein the disease is acute respiratory distress syndrome (ARDS).

[Claim 47] The use of claim 36, wherein the disease or disorder is pre-eclampsia or intrauterine growth restriction (IUGR).

[Claim 48] The use of any one of claims 36-47, wherein the use comprises that the population of isolated mitochondria or the composition is to be administered to the subject via an intravenous, intra-arterial, intra-tracheal, subcutaneous, intramuscular, inhalation, or intrapulmonary route of administration.

[Claim 49] An isolated mitochondrion having intact inner and outer membranes, wherein the inner membrane comprises folded cristae, wherein the mitochondrion has been isolated from a cell, wherein the mitochondrion is polarized as measured by a fluorescence indicator, and wherein the mitochondrion is capable of maintaining polarization in an extracellular environment.

[Claim 50] The isolated mitochondrion of claim 49, wherein the mitochondrion has a non-filamentous shape.

[Claim 51] The isolated mitochondrion of claim 49 or 50, wherein voltage dependent anion channels (VDAC) on the surface of the mitochondrion are associated with tubulin at the surface.

[Claim 52] The isolated mitochondrion of claim 49, wherein the tubulin is dimeric tubulin.

[Claim 53] The isolated mitochondrion of claim 52, wherein the tubulin is a heterodimer comprising α-tubulin and β-tubulin.

[Claim 54] The isolated mitochondrion of any one of claims 49-53, wherein the fluorescence indicator is selected from the group consisting of JC-1, tetramethylrhodamine methyl ester (TMRM), and tetramethylrhodamine ethyl ester (TMRE).

[Claim 55] The isolated mitochondrion of any one of claims 49-54, wherein the isolated mitochondrion exhibits decreased association with mitochondria-associated membrane (MAM) as measured by glucose regulated protein 75 (GRP75) expression.

[Claim 56] The isolated mitochondrion of claim 55, wherein the decreased association is a decrease of at least 30% relative to the association with MAM of a mitochondrion in a cell or of an isolated mitochondrion obtained by a method comprising homogenization of cells.

[Claim 57] The isolated mitochondrion of claim 56, wherein the decreased association is a decrease of at least 50%.

[Claim 58] The isolated mitochondrion of any one of claims 49-57, wherein the membrane potential of the isolated mitochondrion is between -30 mV and -220 mV.

[Claim 59] The isolated mitochondrion of any one of claims 49-58, wherein the isolated mitochondrion is not undergoing drp1 dependent division.

[Claim 60] The isolated mitochondrion of any one of claims 49-59, wherein the isolated mitochondrion is between 500 nm and 3500 nm in size.

[Claim 61] A composition comprising the isolated mitochondrion of any one of claims 49-60.