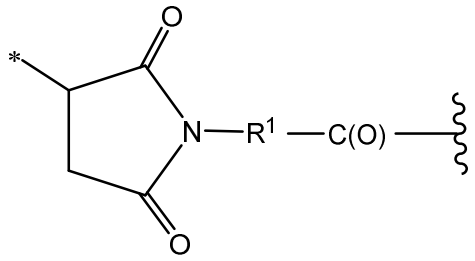
WHAT IS CLAIMED IS:

~~1.~~ An extracellular vesicle (EV) comprising a biologically active molecule covalently linked to the EV via a maleimide moiety~~.~~, wherein the maleimide moiety is linked to the biologically active molecule by a linker

~~2.~~1. ~~The extracellular vesicle of claim 1~~, wherein the maleimide moiety has the formula (I):



(I),

wherein

R^1 is selected from the group consisting of -C_{1-10} alkylene-, -C_{3-8} carbocyclo-, -O-(C_{1-8} alkylene)-, -arylene-, -C_{1-10} alkylene-arylene-, -arylene-C_{1-10} alkylene-, -C_{1-10} alkylene-(C_{3-8} carbocyclo)-, -(C_{3-8} carbocyclo)-C_{1-10} alkylene-, -C_{3-8} heterocyclo-, -C_{1-10} alkylene-(C_{3-8} heterocyclo)-, -(C_{3-8} heterocyclo)-C_{1-10} alkylene-, -$(CH_2CH_2O)_r$-, and -$(CH_2CH_2O)_r$-CH_2-;

r is an integer from 1 to 10;

* indicates the covalent attachment site of the maleimide moiety to the EV; and,

the wavy line indicates the attachment site of the maleimide moiety to the biologically active molecule,

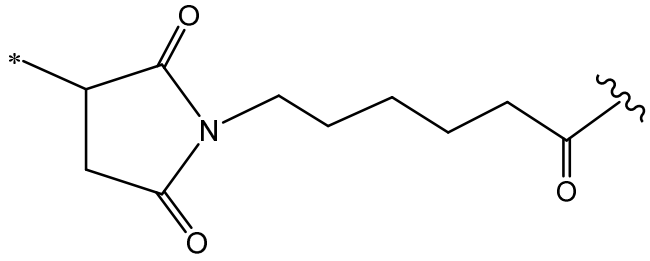
wherein the linker has the formula (IV):

$$-A_a-Y_y- \quad \text{(IV)},$$

wherein each –A- is independently an amino acid unit, a is independently an integer from 1 to 12; -Y- is a spacer unit, and y is 0, 1, or 2.

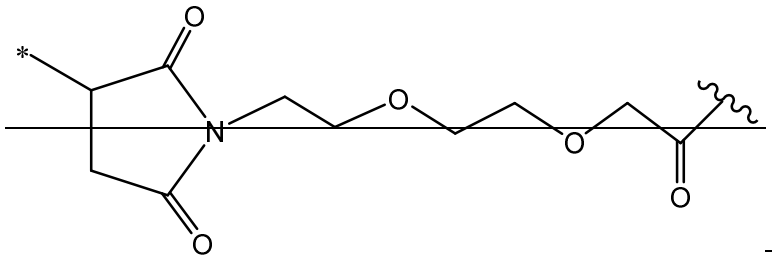
~~3. The extracellular vesicle of claim 2, wherein R^1 is $-(CH_2)_s-$, wherein s is 4, 5, or 6.~~

~~4.~~2. The extracellular vesicle of claim ~~2 or 3~~ 1, wherein the maleimide moiety has the formula (II), where R^1 is $–(CH_2)_5$-:



(II).

~~5. The extracellular vesicle of claim 2, wherein the maleimide moiety has the formula (III), where R^1 is $-(CH_2CH_2O)_r-CH_2-$, where r is 2:~~



~~(III).~~

~~6.~~3. The extracellular vesicle of ~~any one of claims 1-5~~ claim 1 or 2, wherein the maleimide moiety is covalently linked to a functional group present on the EV, wherein the functional group is a sulfhydryl group.

~~7.~~4. The extracellular vesicle of claim ~~6~~3, wherein the sulfhydryl group is on a protein on the surface of the EV.

~~8. The extracellular vesicle of any one of claims 1-7, wherein the maleimide moiety is linked to the biologically active molecule by a linker.~~

~~9.~~5. The extracellular vesicle of claim ~~8~~1, wherein the linker comprises a cleavable linker.

~~10.~~6. The extracellular vesicle of claim ~~9~~5, wherein the cleavable linker is cleaved by a protease.

~~11.~~7. The extracellular vesicle of claim ~~10~~6, wherein the protease is a cathepsin.

~~12.~~8. The extracellular vesicle of claim ~~8~~1, wherein the linker is a reduction-sensitive linker, or an acid labile linker.

~~13. The extracellular vesicle of any one of claims 8-12, wherein the linker has the formula (IV):~~

~~$-A_a-Y_y-$ (IV),~~

~~wherein each -A- is independently an amino acid unit, a is independently an integer from 1 to 12; -Y- is a spacer unit, and y is 0, 1, or 2.~~

~~14.~~9. The extracellular vesicle of claim 1~~3~~, wherein $-A_a-$ is a dipeptide, a tripeptide, a tetrapeptide, a pentapeptide, or a hexapeptide.

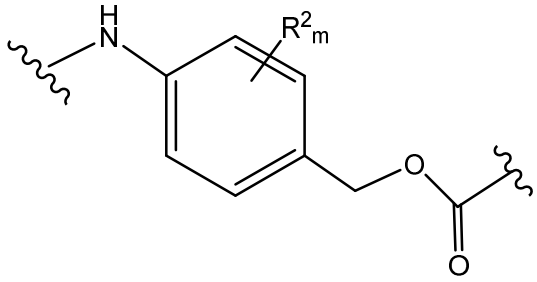
~~15.~~10. The extracellular vesicle of claim ~~14~~9, wherein a is 2 and $–A_a-$ is selected from the group consisting of valine-alanine, valine-citrulline, phenylalanine-lysine, N-methylvaline-citrulline, cyclohexylalanine-lysine, and beta-alanine-lysine.

~~16.~~11. The extracellular vesicle of claim ~~15~~10, wherein said $–A_a-$ is valine-alanine or valine-citrulline.

~~17.~~12. The extracellular vesicle of ~~any one of claims 13-16~~ claim 1, wherein y is 1.

~~18.~~13. The extracellular vesicle of ~~any one of claims 13-17~~ claim 1, wherein –Y- is a self-immolative spacer.

~~19.~~14. The extracellular vesicle of claim ~~18~~13, wherein $–Y_y-$ has the formula (V):



(V),

wherein each R^2 is independently C_{1-8} alkyl, -O-(C_{1-8} alkyl), halogen, nitro, or cyano; and m is an integer from 0 to 4.

~~20.~~15. The extracellular vesicle of claim ~~19~~14, wherein m is 0, 1, or 2.

~~21.~~16. The extracellular vesicle of claim ~~20~~15, wherein m is 0.

~~22.~~17. The extracellular vesicle of any one of claims ~~8~~5-~~21~~16, wherein the cleavable linker is valine-alanine-p-aminobenzylcarbamate or valine-citrulline-p-aminobenzylcarbamate.

~~23.~~18. The extracellular vesicle of ~~any one of claims 13-17~~ claim 1, wherein –Y- is a non self-immolative spacer.

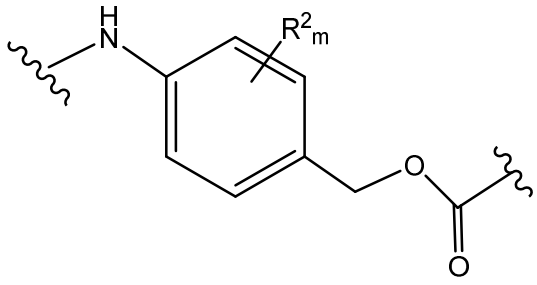
~~24.~~19. The extracellular vesicle of claim ~~23~~18, wherein the non self-immolative spacer is –Gly- or –Gly-Gly-.

~~25.~~20. The extracellular vesicle of claim ~~8~~1, wherein the linker is an acid labile linker.

~~26.~~21. The extracellular vesicle of claim ~~25~~20, wherein the acid labile linker comprises a cis-aconitic linker, a hydrazide linker, a thiocarbamoyl linker, or any combination thereof.

~~27.~~22. The extracellular vesicle of claim ~~25~~20 or ~~26~~21, wherein the acid labile linker comprises a spacer unit to link the biologically active molecule to the acid labile linker.

~~28.~~23. The extracellular vesicle of claim ~~27~~22, wherein the spacer unit has the formula (V):



(V),

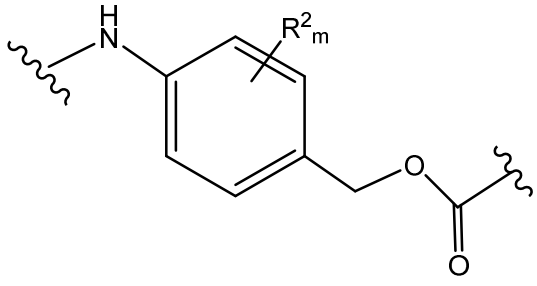
wherein each R^2 is independently C_{1-8} alkyl, -O-(C_{1-8} alkyl), halogen, nitro, or cyano; and m is an integer from 0 to 4.

~~29.~~24. The extracellular vesicle of claim ~~8~~1, wherein the linker is a non-cleavable linker.

~~30.~~25. The extracellular vesicle of claim ~~29~~24, wherein the non-cleavable linker comprises tetraethylene glycol (TEG), polyethylene glycol (PEG), succinimide, or any combination thereof.

~~31.~~26. The extracellular vesicle of claim ~~29~~24 or ~~30~~25, wherein the non-cleavable linker comprises a spacer unit to link the biologically active molecule to the non-cleavable linker.

~~32.~~27. The extracellular vesicle of claim ~~31~~26, wherein the spacer unit has the formula (V):



(V),

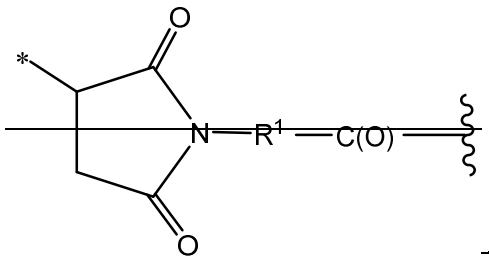
wherein each R^2 is independently C_{1-8} alkyl, -O-(C_{1-8} alkyl), halogen, nitro, or cyano; and m is an integer from 0 to 4.

~~33.~~28. ~~An~~ The extracellular vesicle of claim 1 comprising ~~a~~ the biologically active molecule and a cleavable linker, wherein the cleavable linker connects the EV to the biologically active molecule.

~~34.~~29. The extracellular vesicle of claim ~~33~~28, wherein the cleavable linker comprises valine-alanine-p-aminobenzylcarbamate or valine-citrulline-p-aminobenzylcarbamate.

~~35.~~30. The extracellular vesicle of claim ~~33~~28 or ~~34~~29, further comprising a maleimide moiety, which links the EV to the cleavable linker via a functional group present on the EV.

~~36. The extracellular vesicle of claim 35, wherein the maleimide moiety has the formula (I):~~



~~(I), wherein~~

~~R^1 is selected from the group consisting of -C_{1-10} alkylene-, -C_{3-8} carbocyclo-, -O-(C_{1-8} alkylene)-, -arylene-, -C_{1-10} alkylene-arylene-, -arylene-C_{1-10} alkylene-, -C_{1-10} alkylene-(C_{3-8} carbocyclo)-, -(C_{3-8} carbocyclo)-C_{1-10} alkylene-, -C_{3-8} heterocyclo-, -C_{1-10} alkylene-(C_{3-8} heterocyclo)-, -(C_{3-8} heterocyclo)-C_{1-10} alkylene-, -$(CH_2CH_2O)_r$-, and -$(CH_2CH_2O)_r$-CH_2-;~~

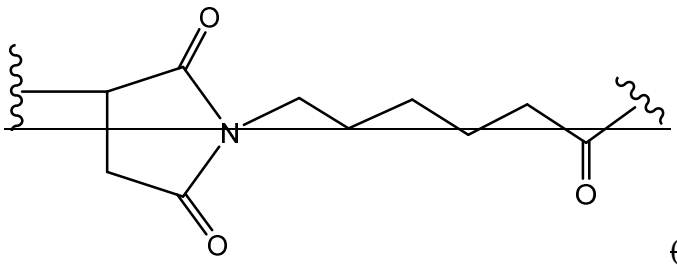
~~r is an integer from 1 to 10; and~~

~~* indicates the covalent attachment site of the maleimide moiety to the EV; and,~~

~~the wavy line indicates the attachment site of the maleimide moiety to the biologically active molecule.~~

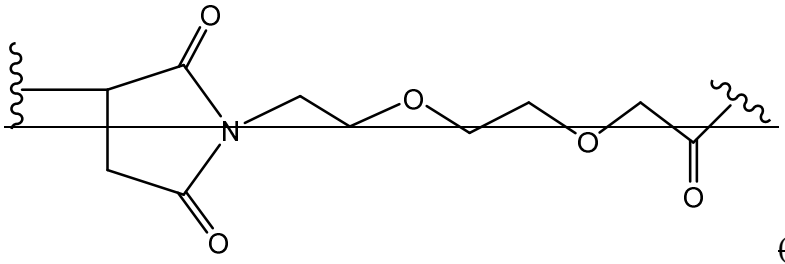
~~37. The extracellular vesicle of claim 36, wherein R^1 is $-(CH_2)_s-$, wherein s is 4, 5, or 6.~~

~~38. The extracellular vesicle of claim 36 or 37, wherein the maleimide moiety has the formula (II), where R^1 is $-(CH_2)_5-$:~~



~~(II).~~

~~39. The extracellular vesicle of claim 36, wherein the maleimide moiety has the formula (III), where R^1 is $-(CH_2CH_2O)_r-CH_2-$, where r is 2:~~



~~(III).~~

~~40.~~31. The extracellular vesicle of ~~any one of claims 34-39~~ claim 1, wherein the maleimide moiety is covalently linked to a functional group present on the EV.

~~41.~~32. The extracellular vesicle of claim ~~40~~31, wherein the functional group is on a glycan on the EV.

~~42.~~33. The extracellular vesicle of claim ~~40~~31, wherein the functional group is sulfhydryl.

~~43.~~34. The extracellular vesicle of claim ~~41~~32 or ~~42~~33, wherein the functional group is on a protein on the surface of the EV.

~~44.~~35. The extracellular vesicle of claim ~~43~~34, wherein the protein is a scaffold moiety.

~~45.~~36. The extracellular vesicle of claim ~~44~~35, wherein the protein is a PTGFRN polypeptide, a BSG polypeptide, a IGSF2 polypeptide, a IGSF3 polypeptide, a IGSF8 polypeptide, a ITGB1 polypeptide, a ITGA4 polypeptide, a SLC3A2 polypeptide, a ATP transporter polypeptide, or a fragment thereof.

~~46.~~37. ~~An~~ The extracellular vesicle of claim 1 comprising a maleimide moiety, a cleavable linker, and a biologically active molecule, wherein the maleimide moiety links the EV to the cleavable linker, and the cleavable linker connects the maleimide moiety to the biologically active molecule.

~~47.~~38. The extracellular vesicle of any one of claims 1-~~46~~37, wherein the biologically active molecule is a polypeptide, a peptide, a polynucleotide (DNA and/or RNA), a chemical compound, or any combination thereof.

~~48.~~39. The extracellular vesicle of claim ~~47~~38, wherein the biologically active molecule is a chemical compound.

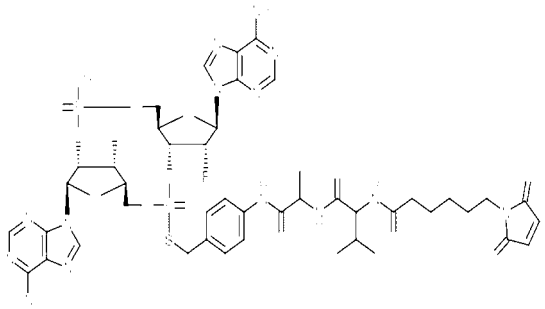
~~49.~~40. The extracellular vesicle of claim ~~48~~39, wherein the chemical compound is a small molecule.

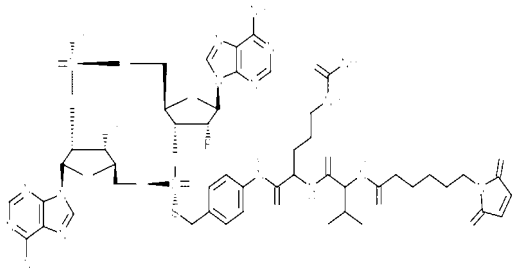
~~50.~~41. The extracellular vesicle of claim ~~49~~40, wherein the small molecule is a proteolysis-targeting chimera (PROTAC).

~~51.~~42. The extracellular vesicle of claim ~~47~~38, wherein the biologically active molecule is nucleotide, wherein the nucleotide is a stimulator of interferon genes protein (STING) agonist.

~~52.~~43. The extracellular vesicle of claim ~~51~~42, wherein the STING agonist comprises a cyclic dinucleotide STING agonist or a non-cyclic dinucleotide STING agonist.

~~53.~~44. The extracellular vesicle of claim ~~52~~43, comprising a (maleimide moiety)-(cleavable linker)-(biologically active molecule) having the formula (VI) or (VII):

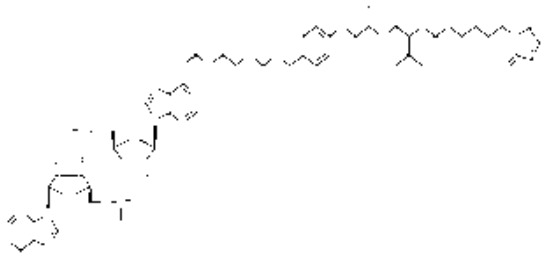


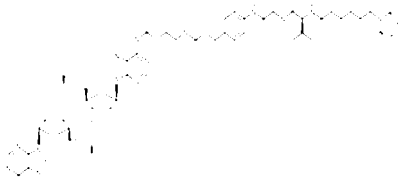


(VI),

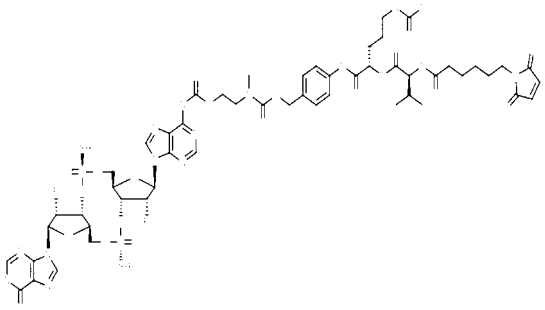
(VII), or a pharmaceutically acceptable salt thereof.

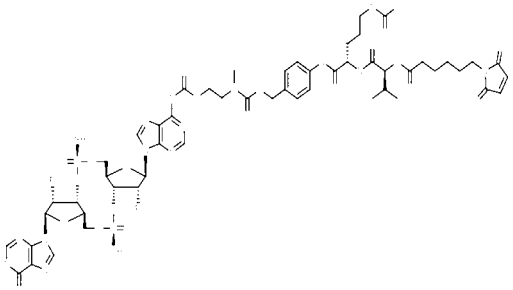
~~54.~~45. The extracellular vesicle of claim ~~52~~43, comprising a (maleimide moiety)-(cleavable linker)-(biologically active molecule) having the formula (VIII), (IX), (X), or (XI):





(VIII), (IX),





(X), (XI),

or a pharmaceutically acceptable salt thereof.

~~55.~~46. The extracellular vesicle of any one of claims 1-~~5~~2, ~~8~~5-~~32~~27, and ~~35~~30-~~45~~36, wherein the EV is modified to expose a functional group on the surface to covalently link the maleimide moiety.

~~56.~~47. The extracellular vesicle of claim ~~55~~46, wherein the functional group is a sulfhydryl group.

~~57.~~48. The extracellular vesicle of claim ~~56~~47, wherein the functional group is exposed by treating the EV with a reducing agent.

~~58.~~49. The extracellular vesicle of claim ~~57~~48, wherein the reducing agent comprises TCEP (Tris(2-carboxyethyl)phosphine), DTT (dithiothreitol), BME (2-mercaptoethanol), a thiolating agent, or any combination thereof.

~~59.~~50. The extracellular vesicle of claim ~~58~~49, wherein the thiolating agent comprises Traut's reagent (2-iminothiolane).

~~60.~~51. The extracellular vesicle of any one of claims 1 to ~~59~~50, wherein the EV is an exosome.

~~61.~~52. A pharmaceutical composition comprising the extracellular vesicle of any one of claims 1 to ~~60~~51 and a pharmaceutically acceptable carrier.

~~62.~~53. A method of conjugating a biologically active molecule to an EV, comprising linking a maleimide moiety to the EV.

~~63.~~54. The method of claim ~~62~~53, wherein the linking comprises treating the EV with a reducing agent.

~~64.~~55. The method of claim ~~63~~54, wherein the reducing agent ~~is~~ comprises TCEP (Tris(2-carboxyethyl)phosphine), DTT (dithiothreitol), BME (2-mercaptoethanol), a thiolating agent, or any combination thereof.

~~65.~~56. The method of claim ~~64~~55, wherein the thiolating agent comprises Traut's reagent (2-iminothiolane).

~~66.~~57. The method of any one of claims ~~62~~53 to ~~65~~56, wherein the linking further comprises bringing the reduced EV in contact with the maleimide moiety.

~~67.~~58. The method of claim ~~66~~57, wherein the maleimide moiety is linked to a biologically active molecule prior to the linking to the EV.

~~68.~~59. The method of claim ~~67~~58, wherein the maleimide moiety is further attached to a linker to connect the maleimide moiety to the biologically active molecule.

~~69.~~60. A kit comprising the EV of any one of claim 1 to ~~60~~51 and instructions for use.

~~70.~~61. A kit comprising reagents to conjugate a biologically active molecule to an EV, and instructions to conduct the conjugation, thereby making the EV of any one of claims 1 to ~~60~~51.

~~71~~62. Use of the EV of any one of claims 1 to ~~60~~51 in the manufacture of a medicament for treating or preventing a disease or disorder in a subject in need thereof, wherein the disease or disorder is a cancer, an inflammatory disorder, a neurodegenerative disorder, a central nervous disease~~s~~, or a metabolic disease.

~~72.~~ 63. The use of claim ~~71~~62, wherein the EV is to be administered intravenously, intraperitoneally, nasally, orally, intramuscularly, subcutaneously, parenterally, intratumorally, intrathecally, or intraocularly.

~~73.~~ 64. The EV of any one of claims 1 to ~~60~~51 for use in treating or preventing a disease or disorder in a subject in need thereof.

~~74.~~ 65. ~~An~~ The extracellular vesicle (EV) of claim 1 comprising at least one biologically active molecule covalently linked to a scaffold moiety via ~~a~~ the maleimide moiety.

~~75.~~ 66. The extracellular vesicle of claim ~~74~~65, wherein the maleimide moiety is a bifunctional molecule.

~~76.~~ 67. The extracellular vesicle of claims ~~74~~65 or ~~75~~66, wherein the maleimide moiety comprises at least one linker or spacer.

~~77.~~ 68. The extracellular vesicle of claim ~~76~~67, wherein the linker is a cleavable linker.

~~78.~~ 69. The extracellular vesicle of any one of claims ~~74~~65 to ~~77~~68, wherein the scaffold moiety is a scaffold protein or a scaffold lipid.

~~79.~~ 70. The extracellular vesicle of claim ~~78~~69, wherein the scaffold protein is a Scaffold X protein.

~~80.~~ 71. The extracellular vesicle of claim ~~79~~70, wherein the Scaffold X protein is a PTGFRN polypeptide, a BSG polypeptide, a IGSF2 polypeptide, a IGSF3 polypeptide, a IGSF8 polypeptide, a ITGB1 polypeptide, a ITGA4 polypeptide, a SLC3A2 polypeptide, a ATP transporter polypeptide, or a fragment thereof.

~~81.~~ 72. The extracellular vesicle of any one of claims ~~74~~65 to ~~80~~71, wherein the biologically active molecule comprises a vaccine antigen, a vaccine adjuvant, or any combination thereof.

~~82.~~ 73. The extracellular vesicle of any one of claims ~~74~~65 to ~~80~~71, wherein the biological active molecule comprises a STING, an ASO, a synthetic antineoplastic agent (e.g., MMAE), a cytokine release inhibitor (e.g., MCC950), an mTOR inhibitor (e.g~~,~~., Rapamycin), an autotaxin inhibitor (e.g., PAT409), an LPA1 antagonist (e.g., AM152), or any combination thereof.

~~83.~~ 74. The extracellular vesicle of any one of claims ~~74~~65 to ~~82~~73, wherein the extracellular vesicle further comprises a targeting moiety, a tropism moiety, an anti-phagocytic signal, or any combination thereof.

~~84.~~ 75. The extracellular vesicle of claim ~~83~~74, wherein the targeting moiety, tropism moiety, anti-phagocytic signal, or combination thereof is linked to the extracellular vesicle via a maleimide moiety.