

WHAT IS CLAIMED IS:

1. A fusion protein comprising a nanocage monomer or a subunit thereof linked to a DR4 and/or DR5 antigen-binding moiety, wherein a plurality of the fusion proteins self-assemble to form a nanocage.

2. The fusion protein of claim 1, wherein the DR4 and/or DR5 antigen-binding moiety targets the DR4 and/or DR5 ectodomain.

~~3. The fusion protein of claim 1 or 2, wherein the DR4 and/or DR5 antigen-binding moiety decorates the interior and/or exterior surface, preferably the exterior surface, of the assembled nanocage.~~

~~43.~~ The fusion protein of ~~any one of claims 1 to 3~~claim 1 or 2, wherein the DR4 and/or DR5 antigen-binding moiety comprises an antibody or fragment thereof.

~~54.~~ The fusion protein of claim ~~43~~, wherein the antibody or fragment thereof comprises a Fab fragment.

~~56.~~ The fusion protein of claim ~~43~~, wherein the antibody or fragment thereof comprises a single chain Fab (scFab) fragment, a single chain Fv (scFv) fragment, a single domain antibody (sdAb) fragment, a nanobody, a VHH domain or a combination thereof.

~~67.~~ The fusion protein of claim ~~43~~, wherein the antibody or fragment thereof comprises a heavy chain and/or light chain of a Fab fragment.

~~78.~~ The fusion protein of any one of claims ~~43~~ to ~~76~~, wherein the fusion protein comprises a DR4 antigen-binding moiety.

~~89.~~ The fusion protein of claim ~~87~~, wherein the DR4 antigen-binding moiety comprises a DR4 antigen-binding moiety of CM005G08, CM059H03, CM084A02, T1014A04, T1014G03, T1014A02, T1014A12, T1014B01, T1014B11, T1014F08, T1014G04, T1015A02, T1015A07, T1006F07, 42/43, 44/45, and/or 46/47.

~~910.~~ The fusion protein of any one of claims ~~43~~ to ~~98~~, wherein the fusion protein comprises a DR5 antigen-binding moiety.

~~101.~~ The fusion protein of claim ~~109~~, wherein the DR5 antigen-binding moiety comprises an antigen-binding moiety of Tigatuzumab, Lexatumumab, Drozitumab, and/or Conatumumab.

~~112.~~ The fusion protein of claim ~~1110~~, wherein the DR5 antigen-binding moiety comprises the antigen-binding moiety of Conatumumab.

~~123.~~ The fusion protein of any one of claims 1 to ~~1211~~, wherein the DR4 and/or DR5 antigen-binding moiety is linked at the N- or C-terminus of the nanocage monomer or subunit thereof, or wherein there is a first DR4 and/or DR5 antigen-binding moiety linked at the N-terminus and a second DR4 and/or DR5 antigen-binding moiety linked at the C-terminus of the nanocage monomer or subunit thereof, wherein the first and second DR4 and/or DR5 antigen-binding moieties are the same or different.

~~14.—The fusion protein of claim 13, wherein the fusion protein comprises a nanocage monomer and the DR4 and/or DR5 antigen-binding moiety is linked at the N-terminus of the nanocage monomer.~~

~~135.~~ The fusion protein of any one of claims 1 to ~~1412~~, wherein the fusion protein comprises a first nanocage monomer subunit linked to the DR4 and/or DR5 antigen-binding moiety; wherein the first nanocage monomer subunit is capable of self-assembling with a second nanocage monomer subunit to form the nanocage monomer.

~~16.—The fusion protein of claim 15, wherein the DR4 and/or DR5 antigen-binding moiety is linked at the N- or C-terminus of the first nanocage monomer subunit, or wherein there is a first DR4 and/or DR5 antigen-binding moiety linked at the N-terminus and a second DR4 and/or DR5 antigen-binding moiety linked at the C-terminus of the first nanocage monomer subunit, wherein the first and second DR4 and/or DR5 antigen-binding moieties are the same or different.~~

~~17. The fusion protein of claim 15 or 16, in combination with the second nanocage monomer subunit.~~

~~18. The fusion protein of any one of claims 15 to 17, wherein the second nanocage monomer subunit is linked to a bioactive moiety at the N- or C-terminus.~~

~~19. The fusion protein of claim 18, wherein the bioactive moiety comprises an Fc fragment.~~

~~20. The fusion protein of claim 19, wherein the Fc fragment is an IgG1 Fc fragment.~~

~~21. The fusion protein of claim 19 or 20, wherein the Fc fragment comprises one or more mutations or sets of mutations that modulate the half-life of the fusion protein from, for example, minutes or hours to several days, weeks, or months.~~

~~22. The fusion protein of claim 21, wherein the Fc fragment comprises a mutation at one or more of L234, L235, G236, G237, M252, I253, S254, T256, P329, A330, M428, N434, or a combination thereof (wherein numbering is according to the EU index), such as M428L and N434S (“LS”); M252Y, S254T and T256E (“YTE”); L234A and L235A (“LALA”); I253A, and/or L234A, L235A, and P329G (“LALAP”); G236R, G237A, A330L or a combination thereof.~~

~~23. The fusion protein of any one of claims 19 to 22, wherein the Fc fragment is an scFc fragment.~~

~~24. The fusion protein of claim 19, wherein the Fc fragment comprises or consists of a sequence at least 70% (such as at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99%, or 100%) identical to:~~

~~DKTHTCPGPCAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVK
FNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV
SKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIA
VEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQQGNVFCFSVMHE
ALHNHYTQKSLSLSPGK.~~

~~25.—The fusion protein of any one of claims 1 to 24, wherein from about 3 to about 100 nanocage monomers, such as 24, 32, 48, or 60 monomers, or from about 4 to about 200 nanocage monomer subunits, such as 4, 6, 8, 10, 12, 14, 18, 20, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 42, 44, 46, 48, 50, or more, optionally in combination with one or more whole nanocage monomers, are capable of self-assembling to form a nanocage.~~

1426. The fusion protein of any one of claims 1 to ~~25~~13, wherein the nanocage monomer is selected from ferritin, apoferritin, encapsulin, SOR, lumazine synthase, pyruvate dehydrogenase, carboxysome, vault proteins, GroEL, heat shock protein, E2P, MS2 coat protein, fragments thereof, and variants thereof.

1527. The fusion protein of claim ~~26~~14, wherein the nanocage monomer is apoferritin, optionally human apoferritin.

1628. The fusion protein of claim ~~26~~14, wherein the nanocage monomer is an apoferritin light chain, optionally human apoferritin light chain.

1729. The fusion protein of claim ~~27~~15 or ~~28~~16, wherein the fusion protein comprises a first apoferritin subunit, optionally a first human apoferritin subunit, and wherein the first apoferritin subunit is capable of self-assembling with a second apoferritin subunit.

1830. The fusion protein of claim ~~29~~17, wherein the first and second apoferritin monomer subunits interchangeably comprise the “N” and “C” regions of apoferritin.

~~31.—The fusion protein of claim 30, wherein the “N” region of apoferritin comprises or consists of a sequence at least 70% (such as at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99%, or 100%) identical to:
MSSQIRQNYSTDVEAAVNSLVNLYLQASYTYLSLGFYFDRDDVALEGVSHFF
RELAEKREGYERLLKMQNRGGRALFQDIKKPAEDEV.~~

~~32. The fusion protein of claim 30 or 31, wherein the “C” region of apoferritin comprises or consists of a sequence at least 70% (such as at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99%, or 100%) identical to:~~

~~GKTPDAMKAAMALEKKLNQALLDLHALGSARTDPHLCDFLETHFLDEEVKLI
KKMGDHLTNLHRLGGPEAGLGEYLFERLTLRHD or~~

~~GKTPDAMKAAMALEKKLNQALLDLHALGSARTDPHLCDFLETHFLDEEVKLI
KKMGDHLTNLHRLGGPEAGLGEYLFERLTLKHD.~~

1933. The fusion protein of any one of claims 1 to 32,18 further comprising a bioactive moiety and an optional linker between the nanocage monomer or subunit thereof and the bioactive moiety.

2034. The fusion protein of claim 3319, wherein the linker is flexible or rigid and comprises from ~~about~~ 1 to ~~about~~ 30 amino acid residues, such as from ~~about~~ 8 to ~~about~~ 16 amino acid residues.

2135. The fusion protein of claim 33-19 or 2034, wherein the linker comprises at least one GGGGS repeat, such as 1, 2, 3, 4, or more GGGGS repeats.

~~36. The fusion protein of claim 35, wherein the linker comprises or consists of a sequence at least 70% (such as at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99%, or 100%) identical to:~~

~~GGGSGGGSGGGSGGGSGGGSGGGSGG.~~

2237. The fusion protein of any one of claims 1 to 3621, further comprising a C-terminal linker.

~~38. The fusion protein of claim 37, wherein the C-terminal linker comprises a GGS repeat.~~

~~39. The fusion protein of claim 38, wherein a C-terminal linker comprises or consists of a sequence at least 70% (such as at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99%, or 100%) identical to:~~

2747. The nanocage of any one of claims 40-23 to 2646, wherein at least one DR4 and/or DR5 antigen-binding moiety decorates the exterior surface of the nanocage and at least one Fc fragment decorates the exterior surface of the nanocage.

2848. The nanocage of claim 2747, wherein at least two DR4 and/or DR5 antigen-binding moieties decorate the exterior surface of the nanocage and at least two Fc fragments decorate the exterior surface of the nanocage.

2949. The nanocage of any one of claims 40-23 to 2848, comprising a 4:1:1 ratio of an antigen-binding moiety, such as an Fab fragment, of Conatumumab fused to a first full length human ferritin light chain; an Fc fragment (optionally an scFc fragment) fused to a second full length human ferritin light chain; and a third human ferritin light chain.

3050. The nanocage of any one of claims 40-23 to 4928, comprising at least one DR4 and/or DR5 antigen-binding moiety fused to the N-terminus of a full ferritin monomer, at least one DR4 and/or DR5 antigen-binding moiety fused to the N-terminus of an N-ferritin monomer subunit, and an Fc fragment fused to the N-terminus of a C-ferritin monomer subunit.

3151. The nanocage of claim 5030, comprising a 2:1:1 ratio of the DR4 and/or DR5 antigen-binding moiety fused to the N-terminus of the full ferritin monomer: the DR4 and/or DR5 antigen-binding moiety fused to the N-terminus of the N-ferritin monomer subunit: the Fc fragment fused to the N-terminus of the C-ferritin monomer subunit.

~~52. The nanocage of any one of claims 40 to 51, comprising at least about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, or 48 DR4 and/or DR5 antigen-binding moieties.~~

~~53. The nanocage of any one of claims 40 to 52, comprising at least about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, or 48 bioactive moieties.~~

3254. The nanocage of any one of claims 40-23 to 5331, carrying a cargo molecule, such as a pharmaceutical agent, a diagnostic agent, and/or an imaging agent.

~~55. The nanocage of claim 54, wherein the cargo molecule is not fused to the fusion protein and is contained in the nanocage internally.~~

~~56. The nanocage of claim 54, wherein the cargo molecule is a protein and is fused to the fusion protein such that the cargo molecule is contained in the nanocage internally.~~

~~57. The nanocage of any one of claims 54 to 56, wherein the cargo molecule comprises a fluorescent protein, such as GFP, EGFP, Ametrine, and/or a flavin-based fluorescent protein, such as a LOV protein, such as iLOV.~~

~~58. The nanocage of any one of claims 40 to 57, wherein the nanocage is capable of killing DR4 and/or DR 5 positive cancer cells with an IC₅₀ value of less than about 0.1 µg/ml, less than about 0.01 µg/ml, or less than about 0.001 µg/ml, as determined in an *in vitro* cell killing assay.~~

~~59. The nanocage of any one of claims 40 to 58, wherein the nanocage is capable of killing DR4 and/or DR 5 positive cancer cells with an IC₅₀ value of less than about 10 pM, less than about 1 pM, or less than about 0.1 pM, as determined in an *in vitro* cell killing assay.~~

~~60. The nanocage of any one of claims 40 to 59, wherein the nanocage is capable of killing DR4 and/or DR 5 positive cancer cells with an IC₅₀ value that is at least about 10, at least about 100, at least about 1000, at least about 10,000, or at least about 100,000 more potent than the corresponding IgG on a mass and/or molar basis.~~

3361. A DR4 and/or DR5 therapeutic or prophylactic composition comprising the nanocage of any one of claims 40-23 to 6032.

3462. A nucleic acid molecule encoding the fusion protein of any one of claims 1 to 3922.

3563. A vector comprising the nucleic acid molecule of claim 6234.

3664. A host cell comprising the vector of claim 6335 and producing the fusion protein of any one of claims 1 to 3922.

3765. Use of the nanocage of any one of claims 4023 to 6032 or the composition of claim 6133 in the manufacture of a medicament~~A method~~ for treating and/or preventing cancer, ~~the method comprising administering the nanocage of any one of claims 40 to 60 or the composition of claim 61.~~

~~66. The method of claim 65, wherein the cancer is selected from the group consisting of breast cancer, colon cancer, lymphoma, or lung cancer.~~

3867. ~~Use of~~ The nanocage of any one of claims 4023 to 6032 or the composition of claim 6133 for ~~treating and/or preventing cancer~~ use in therapy.

~~68. The nanocage of any one of claims 40 to 60 or the composition of claim 61 for use in treating and/or preventing cancer.~~

39. The use according to claim 37, wherein the cancer is selected from the group consisting of breast cancer, colon cancer, lymphoma, and lung cancer.