

CLAIMS

1. ~~An interleukin 15 (IL15) construct fusion protein comprising at least one IL15 molecule, at least one IL15 receptor alpha (IL15Ra) domain, at least one protease activatable moiety and at least one IgG1 Fc region consisting of:~~

- ~~a) a first domain consisting of SEQ ID NO:2 (IL15 receptor alpha (IL15Ra));~~
- ~~b) a second domain consisting of SEQ ID NO:1 (IL15);~~
- ~~c) a third domain consisting of SEQ ID NO:3 (Interleukin-2 receptor beta (IL2Rb));~~
- ~~d) a fourth domain consisting of a first IgG1 Fc region; and~~
- ~~e) a fifth domain consisting of a second IgG1 Fc region;~~

~~wherein~~

- ~~_____ the first and second domains are linked via a first linker (L1),~~
- ~~_____ the second and third domains are linked via a second linker comprising a protease-activatable moiety (L2),~~
- ~~_____ the third and fourth domains are linked via a third linker (L3), and~~
- ~~_____ the fourth and fifth domains are linked via a disulfide bond.~~

2. The IL15 fusion protein of claim 1, wherein the first and second IgG1 Fc regions are assembled, at least in part, through a knob into hole interaction.

2. ~~The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a bivalent, homodimeric interleukin 15 (IL15) construct comprising from N terminus to C terminus:—~~

- ~~a) an IL15 receptor alpha (IL15Ra) domain linked to;—~~
- ~~b) a first linker (L1) linked to;—~~
- ~~e) an IL15 domain, linked to;—~~
- ~~d) a second linker (L2) containing a protease activatable moiety linked to;—~~
- ~~e) an Interleukin 2 receptor beta (IL2Rb) domain; and~~
- ~~f) an IgG1 Fc region, wherein the bivalent IL15 construct comprises:~~
 - ~~—— (i) a homodimer set forth in SEQ ID NO:4 (M123);~~
 - ~~—— (ii) a homodimer set forth in SEQ ID NO:5 (M135);~~
 - ~~—— (iii) a homodimer set forth in SEQ ID NO:6 (M140);~~
 - ~~—— (iv) a homodimer set forth in SEQ ID NO:7 (M145);~~
 - ~~—— (v) a homodimer set forth in SEQ ID NO:8 (M175);~~
 - ~~—— (vi) a homodimer set forth in SEQ ID NO:9 (M176);~~
 - ~~—— (vii) a homodimer set forth in SEQ ID NO:10 (M177);~~
 - ~~—— (viii) a homodimer set forth in SEQ ID NO:11 (M178);~~
 - ~~—— (ix) a homodimer set forth in SEQ ID NO:12 (M207);~~
 - ~~—— (x) a homodimer set forth in SEQ ID NO:13 (M231);~~
 - ~~—— (xi) a homodimer set forth in SEQ ID NO:14 (M233);~~

- (xii) a homodimer set forth in SEQ ID NO:15 (M234);
- (xiii) a homodimer set forth in SEQ ID NO:16 (M238);
- (xiv) a homodimer set forth in SEQ ID NO:17 (M239);
- (xv) a homodimer set forth in SEQ ID NO:18 (M240);
- (xvi) a homodimer set forth in SEQ ID NO:19 (M241);
- (xvii) a homodimer set forth in SEQ ID NO:20 (M243);
- (xviii) a homodimer set forth in SEQ ID NO:21 (M244);
- (xix) a homodimer set forth in SEQ ID NO:22 (M245);
- (xx) a homodimer set forth in SEQ ID NO:23 (M246);
- (xxi) a homodimer set forth in SEQ ID NO:24 (M247);
- (xxii) a homodimer set forth in SEQ ID NO:25 (M248);
- (xxiii) a homodimer set forth in SEQ ID NO:26 (M249);
- (xxiv) a homodimer set forth in SEQ ID NO:27 (M327);
- (xxv) a homodimer set forth in SEQ ID NO:28 (M328);
- (xxvi) a homodimer set forth in SEQ ID NO:29 (M329);
- (xxvii) a homodimer set forth in SEQ ID NO:30 (M330);
- (xxviii) a homodimer set forth in SEQ ID NO:31 (M331); or
- (xxix) a homodimer set forth in SEQ ID NO:32 (M332).

3. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a bivalent, heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

- a) an Interleukin 2 receptor beta (IL2Rb) domain linked to;—
- b) a first linker (L1) containing a protease activatable moiety, linked to;
- e) an IL15 domain,—

comprising a first molecule and a second molecule comprising from N-terminus to C-terminus:—

- x) an IL15 receptor alpha (IL15Ra) domain; and
- y) an IgG1 Fc region, wherein the heterodimeric IL15 construct comprises:
 - (i) a first molecule set forth in SEQ ID NO:33 (M43) and a second molecule set forth in SEQ ID NO:34 (M24);
 - (ii) a first molecule set forth in SEQ ID NO:35 (M61) and a second molecule set forth in SEQ ID NO:36 (M60); or
 - (iii) a first molecule set forth in SEQ ID NO:37 (M62) and a second molecule set forth in SEQ ID NO:38 (M60).

4. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a bivalent, homodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

- a) an IgG1 Fc region, linked to;
- b) a first linker (L1) linked to;—
- e) an Interleukin 2 receptor beta (IL2Rb) domain linked to;—
- d) a second linker (L2) containing a protease activatable moiety linked to;—

e) an IL15 receptor alpha (IL15Ra) domain linked to;
 f) a third linker (L3) linked to;—
 g) an IL15 domain; and
 wherein the bivalent IL15 construct comprises:
 — (i) a homodimer set forth in SEQ ID NO:42 (M232)
 — (ii) a homodimer set forth in SEQ ID NO:43 (M1001);—
 — (iii) a homodimer set forth in SEQ ID NO:44 (M1002);
 — (vi) a homodimer set forth in SEQ ID NO:45 (M1003);—
 — (v) a homodimer set forth in SEQ ID NO:46 (M1004);
 — (vi) a homodimer set forth in SEQ ID NO:47 (M1005); or
 — (vii) a homodimer set forth in SEQ ID NO:48 (M1006).

5. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent, heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—
 a) an IL15 receptor alpha (IL15Ra) domain linked to;—
 b) a first linker (L1) linked to;—
 c) an IL15 domain linked to;
 d) a second linker (L2) containing a protease activatable moiety linked to;—
 e) an Interleukin 2 receptor beta (IL2Rb) domain; and
 f) a first IgG1 Fc region,—
 as a first molecule and a second molecule comprising a second IgG1 Fc region, wherein the heterodimeric IL15 construct comprises a first molecule set forth in SEQ ID NO:49 (MK107) and a second molecule set forth in SEQ ID NO:50 (MH2).

6. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent, heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—
 a) a first IgG1 Fc region linked to;
 b) a first linker (L1) linked to;
 c) an Interleukin 2 receptor beta (IL2Rb) domain linked to;—
 d) a second linker (L2) containing a protease activatable moiety to;—
 e) an IL15 receptor alpha (IL15Ra) domain linked to;—
 f) a third linker (L3) linked to;
 g) an IL15 domain;
 as a first molecule and a second molecule comprising a second IgG1 Fc region, wherein the monovalent, heterodimeric IL15 construct comprises:
 — (i) a first molecule set forth in SEQ ID NO:63 (M111) and a second molecule set forth in SEQ ID NO:64 (MH2);
 — (ii) a first molecule set forth in SEQ ID NO:65 (M2001) and a second molecule set forth in SEQ ID NO:52 (MH7); or

—(iii) a first molecule set forth in SEQ ID NO:66 (M2002) and a second molecule set forth in SEQ ID NO:52 (MH7);

37. The interleukin-15-IL15) construct fusion protein of claim 1, wherein the construct comprises a monovalent, heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus: a) an IL15 receptor alpha (IL15Ra) domain linked to;—

b) a first linker (L1) linked to;—

c) an IL15 domain linked to;

d) a second linker (L2) containing a protease activatable moiety linked to;—

e) an Interleukin 2 receptor beta (IL2Rb) domain linked to;—

f) a third linker (L3) linked to;

g) a first IgG1 Fc region;—

as a first molecule and a second molecule comprising a second IgG1 Fc region, wherein the heterodimeric IL15 construct comprises:

—(i) a first molecule set forth in SEQ ID NO:51 (MK114) and a second molecule set forth in SEQ ID NO:52 (MH7);—

—(ii) a first molecule set forth in SEQ ID NO:53 (MK115) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(iii) a first molecule set forth in SEQ ID NO:54 (MK117) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(iv) a first molecule set forth in SEQ ID NO:55 (MK118) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(v) a first molecule set forth in SEQ ID NO:56 (MK119) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(vi) a first molecule set forth in SEQ ID NO:57 (MK120) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(vii) a first molecule set forth in SEQ ID NO:58 (MK121) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(viii) a first molecule set forth in SEQ ID NO:59 (MK123) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(ix) a first molecule set forth in SEQ ID NO:60 (MK124) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(x) a first molecule set forth in SEQ ID NO:61 (MK125) and a second molecule set forth in SEQ ID NO:52 (MH7);—

—(xi) a first molecule set forth in SEQ ID NO:62 (MK126) and a second molecule set forth in SEQ ID NO:52 (MH7);—

—(xii) a first molecule set forth in SEQ ID NO:67 (MK136) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(xiii) a first molecule set forth in SEQ ID NO:68 (MK137) and a second molecule set forth in SEQ ID NO:52 (MH7);—

—(xiv) a first molecule set forth in SEQ ID NO:69 (MK138) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(xv) a first molecule set forth in SEQ ID NO:70 (MK139) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(xvi) a first molecule set forth in SEQ ID NO:71 (MK140) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(xvii) a first molecule set forth in SEQ ID NO:72 (MK141) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(xviii) a first molecule set forth in SEQ ID NO:73 (MK146) and a second molecule set forth in SEQ ID NO:52 (MH7);

—(xix) a first molecule set forth in SEQ ID NO:74 (MK145) and a second molecule set forth in SEQ ID NO:75 (MH8);

—(xx) a first molecule set forth in SEQ ID NO:76 (MK149) and a second molecule set forth in SEQ ID NO:75 (MH8);

—(xxi) comprising a first molecule set forth in SEQ ID NO:77 (MK150) and a second molecule set forth in SEQ ID NO:75 (MH8);

—(xxii) a first molecule set forth in SEQ ID NO:78 (MK151) and a second molecule set forth in SEQ ID NO:75 (MH8);—

—(xxiii) a first molecule set forth in SEQ ID NO:79 (MK152) and a second molecule set forth in SEQ ID NO:75 (MH8);—

(xxiv) a first molecule set forth in SEQ ID NO:80 (MK153) and a second molecule set forth in SEQ ID NO:75 (MH8);—

—(xxv) a first molecule set forth in SEQ ID NO:81 (MK154) and a second molecule set forth in SEQ ID NO:75 (MH8);—

(xxvi) a first molecule set forth in SEQ ID NO:82 (MK155) and a second molecule set forth in SEQ ID NO:52 (MH7); or—

—(xxvii) a first molecule set forth in SEQ ID NO:172 (MK157) and a second molecule set forth in SEQ ID NO:75 (MH8).

8. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent, heterodimeric interleukin 15 (IL15) construct comprising from N terminus to C terminus:—

- a) a first IgG1 Fc region linked to;
 - b) a first linker (L1) containing a protease activatable moiety linked to;
 - c) an IL15 receptor alpha (IL15Ra) domain linked to;—
 - d) a second linker (L2) linked to;—
 - e) an IL15 domain;
- as a first molecule and a second molecule comprising:
- x) a second IgG1 Fc region;
 - y) a linker (L3) linked to;—

z) an Interleukin 2 receptor beta (IL2Rb) domain;—

wherein the monovalent, heterodimeric IL15 construct comprises:

—— (i) a heterodimer set forth in SEQ ID NO:83 (M109) and set forth in SEQ ID NO:84 (MH110);

—— (ii) a heterodimer set forth in SEQ ID NO:85 (M2003) and set forth in SEQ ID NO:86 (MH2004); or

—— (iii) a heterodimer set forth in SEQ ID NO:87 (M2003) and set forth in SEQ ID NO:88 (MH2005).

9. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent, heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

a) an Interleukin 2 receptor beta (IL2Rb) domain linked to;—

b) a first linker (L1) containing a protease activatable moiety linked to;

c) an IL15 domain and;—

d) a first IgG1 Fc region;

as a first molecule and a second molecule comprising:

x) an IL15 receptor alpha (IL15Ra) domain linked to;—

y) a linker (L3) linked to;—

z) a second IgG1 Fc region;—

wherein the monovalent, heterodimeric IL15 construct comprises a first molecule set forth in SEQ ID NO:95 (M108) and a second molecule set forth in SEQ ID NO:96 (MH4).—

10. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent, heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

a) a first IgG1 Fc region linked to;

b) a first linker (L1) linked to;

c) an IL15 domain linked to;

d) a second linker (L2) containing a protease activatable moiety linked to;

e) an Interleukin 2 receptor beta (IL2Rb) domain;—

as a first molecule and a second molecule comprising:

x) a second IgG1 Fc region linked to;—

y) a linker (L3) linked to;—

z) an IL15 receptor alpha (IL15Ra) domain;—

wherein the monovalent, heterodimeric IL15 construct comprises:

—— a first molecule set forth in SEQ ID NO:97 (M112) and a second molecule set forth in SEQ ID NO:98 (MK113).—

11. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a bivalent homodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

a) a tumor associated antigen (TAA) binding antibody with a first IgG1 Fc region linked to;

b) a first linker (L1) linked to;

- e) an Interleukin 2 receptor beta (IL2Rb) domain linked to;—
- d) a second linker (L2) containing a protease activatable moiety linked to;—
- e) an IL15 receptor alpha (IL15Ra) domain linked a third linker (L3) to;—
- f) an IL15 domain;

wherein the bivalent, homodimeric IL15 construct comprises the sequence set forth in SEQ ID NO:99 (M001) and the sequence set forth in SEQ ID NO:100 (MH333LC).—

12. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

- a) a tumor associated antigen (TAA) binding antibody with a first IgG1 Fc region linked to;
- b) a first linker (L1) linked to;
- e) an Interleukin 2 receptor beta (IL2Rb) domain linked to;—
- d) a second linker (L2) containing a protease activatable moiety linked to;—
- e) an IL15 receptor alpha (IL15Ra) domain linked to third linker (L3) linked to;
- f) an IL15 domain;

wherein the monovalent, heterodimeric IL15 construct comprises the sequence set forth in SEQ ID NO:101 (M002), the sequence set forth in SEQ ID NO:102 (MH2) and the sequence set forth in SEQ ID NO:100 (MH333LC).—

13. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

- a) a tumor associated antigen (TAA) binding antibody with a first IgG1 Fc region linked to;
- b) a first linker (L1) containing a protease activatable moiety linked to;
- e) an IL15 receptor alpha (IL15Ra) domain linked to;
- d) a second linker (L2) linked to;—
- e) an IL15 domain;—

wherein the monovalent, heterodimeric IL15 construct comprises the sequence set forth in SEQ ID NO:103 (MK3), and—

- x) a tumor associated antigen (TAA) binding antibody comprising a second IgG1 Fc region linked to;
- y) a first linker (L3) linked to;—
- z) an Interleukin 2 receptor beta (IL2Rb) domain,—

wherein the sequence is set forth in SEQ ID NO:104 (MH3) and is set forth in SEQ ID NO:100 (MH333LC).—

14. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

- a) a first tumor associated antigen (TAA) binding antibody with a first IgG1 Fc region linked to;
- b) a first linker (L1) containing a protease activatable moiety linked to;
- e) an IL15 domain linked to;—

d) a second linker (L2) linked to;—
e) an IL15 receptor alpha (IL15Ra) domain;
wherein the monovalent, heterodimeric IL15 construct comprises the sequence set forth in SEQ ID NO:105 (MK4), and—
x) a first tumor associated antigen (TAA) binding antibody with a second IgG1 Fc region linked to;
y) a first linker (L3) set forth in;
z) an Interleukin 2 receptor beta (IL2Rb) domain,—
wherein the sequence is set forth in SEQ ID NO:106 (MH3) and is set forth in SEQ ID NO:100 (MH333LC).—

15. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent, heterodimeric interleukin 15 (IL15) construct comprising from N terminus to C terminus:—
a) an Interleukin 2 receptor beta (IL2Rb) domain linked to;
b) a first linker (L1) containing a protease activatable moiety linked to;—
c) an IL15 domain;
as a first molecule; and
a second molecule comprising—
x) an IL15 receptor alpha (IL15Ra) domain; and
y) a first IgG1 Fc region,—
and a third molecule comprising a second IgG1 Fc region, wherein the heterodimeric IL15 construct comprises:—
—— a first molecule set forth in SEQ ID NO:107 (MK143), a second molecule set forth in SEQ ID NO:108 (MK144) and a third molecule set forth in SEQ ID NO:52 (H7).

16. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent, heterodimeric interleukin 15 (IL15) construct comprising from N terminus to C terminus:—
a) a first IgG1 Fc region linked to;
b) a first linker (L1) linked to;
c) an IL15 receptor alpha (IL15Ra) domain linked to;
d) a second linker (L2) linked to;—
e) an IL15 domain
f) a third linker (L3) containing a protease activatable moiety linked to;—
g) an Interleukin 2 receptor beta (IL2Rb) domain;
as a first molecule and a second molecule comprising a second IgG1 Fc region, wherein the monovalent, heterodimeric IL15 construct comprises:
(i) —— a first molecule set forth in SEQ ID NO:109 (MK142) and a second molecule set forth in SEQ ID NO:52 (MH7);—
(ii) —— a first molecule set forth in SEQ ID NO:173 (MK156) and a second molecule set forth in SEQ ID NO:75 (MH8); or

(iii) a first molecule set forth in SEQ ID NO:174 (MK165) and a second molecule set forth in SEQ ID NO:75 (MH8).

17. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent, heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

- a) an IL15 receptor alpha (IL15Ra) domain as a first molecule that is linked via a disulfide bond to;—
- b) an IL15 domain linked to;
- e) a first linker (L1) containing a protease activatable moiety linked to;—
- d) an Interleukin 2 receptor beta (IL2Rb) domain linked to;—
- e) second linker (L2), linked to
- f) a first IgG1 Fc region;—

and a second molecule comprising a second IgG1 Fc region, wherein the heterodimeric IL15 construct comprises: a first molecule set forth in SEQ ID NO:110 (MK147) and a second molecule set forth in SEQ ID NO:111 (MK148) and a third molecule set forth in SEQ ID NO:52 (MH7).—

18. The interleukin 15 (IL15) construct of claim 1, wherein the construct comprises a monovalent heterodimeric interleukin 15 (IL15) construct comprising from N-terminus to C-terminus:—

- a) a tumor associated antigen (TAA) binding antibody with a first IgG1 Fc region linked to;
- b) a first linker (L1) linked to;
- e) an IL15 receptor alpha (IL15Ra) domain linked to;
- d) a second linker (L2) linked to;
- e) an IL15 domain linked to;
- f) a third linker (L3) containing a protease activatable moiety linked to;—
- g) an Interleukin 2 receptor beta (IL2Rb) domain; and

wherein the monovalent, heterodimeric IL15 construct comprises the sequence set forth in SEQ ID NO:175 (MK14), the sequence set forth in SEQ ID NO:102 (MH2) and the sequence set forth in SEQ ID NO:100 (MH333LC).—

419. A pharmaceutical composition comprising the IL15 ~~construct~~ fusion protein of any one of claims 1 to 483 in combination with at least one additional IL15 construct.

520. Use of the IL15 fusion protein of any one of claims 1 to 3 in the manufacture of a medicament for the therapeutic treatment of ~~A method of treating cancer comprising administering to a patient in need an effective amount of the IL15 construct of any one of claims 1 to 16.~~

621. _____ The use ~~method~~ of claim 520, wherein the cancer is gastric cancer, colon cancer, pancreatic cancer, breast cancer, head and neck cancer, kidney cancer, liver cancer, small cell lung cancer, non-small cell lung cancer, ovarian cancer, skin cancer, mesothelioma, lymphoma, leukemia, myeloma and sarcoma.

722.— _____ Use of the IL15 fusion protein of any one of claims 1 to 3 in the manufacture of a medicament for the therapeutic treatment of cancer, ~~The method of claim 20, wherein the IL15~~

~~construct is~~ wherein the medicament is to be administered in combination with another therapeutic agent.

~~823.~~— The ~~method~~use of claim 227, wherein the therapeutic agent is an immune checkpoint agent.

~~924.~~— The ~~method~~use of claim 238, wherein the immune checkpoint agent is a PD-1, PD-L1, PD-L2, TIM3, LAG-3, OX40 or TIGIT antibody.

10. The use of claim 7, wherein the cancer is gastric cancer, colon cancer, pancreatic cancer, breast cancer, head and neck cancer, kidney cancer, liver cancer, small cell lung cancer, non-small cell lung cancer, ovarian cancer, skin cancer, mesothelioma, lymphoma, leukemia, myeloma and sarcoma.

~~25. A method of increasing the survival of an immune cell, comprising administering an IL15-construct of any one of claims 1 to 18 prior to, during or after administration of an effective amount of immune cells to a patient.—~~

~~26. The method of claim 25 wherein the immune cell expresses a chimeric antigen receptor (CAR).—~~

~~27. The method of claim 26 wherein the immune cell is an NK cell.—~~

~~28. The method of claim 26 wherein the immune cell is a T cell.—~~