

Claims

1. ~~An~~1. A bispecific antibody construct comprising at least three domains in an amino to carboxyl order, wherein:

- a first domain binds to a target cell surface antigen,
- a second domain binds to an extracellular epitope of the human and/or the *Macaca* CD3ε chain; and
- a third domain comprises two polypeptide monomers, each comprising a hinge, a CH2 and a CH3 domain, wherein said two polypeptide monomers are fused to each other via a peptide linker, wherein said third domain comprises in an amino to carboxyl order:

hinge-CH2-CH3-linker-hinge-CH2-CH3.

2. The antibody construct of claim 1, wherein the antibody construct is a single chain antibody construct.

3. The antibody construct of ~~claim 1 or 2~~, wherein ~~said third domain comprises in an amino to carboxyl order:~~

~~hinge-CH2-CH3-linker-hinge-CH2-CH3.~~

4. ~~The antibody construct of any one of claims 1 to 3~~or 2, wherein each of said polypeptide monomers has an amino acid sequence that is at least 90% identical to a sequence selected from the group from the group consisting of: SEQ ID NO: 17-24.

~~54.~~ The antibody construct of claim ~~43~~, wherein each of said polypeptide monomers has an amino acid sequence selected from SEQ ID NO: 17-24.

~~65.~~ The antibody construct of any one of claims 1 to ~~54~~, wherein the CH2 domain comprises an intra domain cysteine disulfide bridge.

~~76.~~ The antibody construct of any one of claims 1 to ~~65~~, wherein

- (i) the first domain comprises two antibody variable domains and the second domain comprises two antibody variable domains;
- (ii) the first domain comprises one antibody variable domain and the second domain comprises two antibody variable domains;

- (iii) the first domain comprises two antibody variable domains and the second domain comprises one antibody variable domain; or
- (iv) the first domain comprises one antibody variable domain and the second domain comprises one antibody variable domain.

~~87~~. The antibody construct of any one of claims 1 to ~~76~~, wherein the first and second domain are fused to the third domain via a peptide linker.

~~98~~. The antibody construct according to claims 1 to ~~87~~, wherein the antibody construct comprises in an amino to carboxyl order:

- (a) the first domain;
- (b) a peptide linker having an amino acid sequence selected from the group consisting of SEQ ID NOs: 1-3;
- (c) the second domain;
- (d) a peptide linker having an amino acid sequence selected from the group consisting of SEQ ID NOs: 1, 2, 3, 9, 10, 11 and 12;
- (e) the first polypeptide monomer of the third domain;
- (f) a peptide linker having an amino acid sequence selected from the group consisting of SEQ ID NOs: 5, 6, 7 and 8; and
- (g) the second polypeptide monomer of the third domain.

~~109~~. The antibody construct according to any one of the preceding claims, wherein the target cell surface antigen is a tumor antigen, an antigen specific for an immunological disorder or a viral antigen.

~~110~~. The antibody construct according to claim ~~109~~, wherein the tumor antigen is selected from the group consisting of CDH19, MSLN, DLL3, FLT3, EGFRvIII, CD33, CD19, CD20, and CD70.

~~111~~. The antibody construct according to any one of the preceding claims, wherein the antibody construct comprises in an amino to carboxyl order:

- (a) the first domain having an amino acid sequence selected from the group consisting of SEQ ID NOs: 52, 70, 58, 76, 88, 106, 124, 94, 112, 130, 142, 160, 178, 148, 166, 184, 196, 214, 232, 202, 220, 238, 250, 266, 282, 298, 255, 271, 287, 303, 322, 338, 354, 370, 386, 402, 418, 434, 450, 466, 482, 498, 514, 530, 546, 327, 343, 359, 375, 391,

407, 423, 439, 455, 471, 487, 503, 519, 353, 551, 592, 608, 624, 640, 656, 672, 688, 704, 720, 736, 752, 768, 784, 800, 816, 832, 848, 864, 880, 896, 912, 928, 944, 960, 976, 992, 1008, 1024, 1040, 1056, 1072, 1088, 1104, 1120, 1136, 1152, 1168, 1184, 597, 613, 629, 645, 661, 677, 693, 709, 725, 741, 757, 773, 789, 805, 821, 837, 853, 869, 885, 901, 917, 933, 949, 965, 981, 997, 1013, 1029, 1045, 1061, 1077, 1093, 1109, 1125, 1141, 1157, 1173, 1189, 1277, 1289, 1301, 1313, 1325, 1337, 1349, 1361, 1373, 1385, 1397, 1409, 1421, 1433, 1445;

- (b) a peptide linker having an amino acid sequence selected from the group consisting of SEQ ID NOs: 1-3;
- (c) the second domain having an amino acid sequence selected from the group consisting of SEQ ID NOs: SEQ ID NOs: 23, 25, 41, 43, 59, 61, 77, 79, 95, 97, 113, 115, 131, 133, 149, 151, 167, 169, 185 or 187 of WO 2008/119567 or of SEQ ID NO: 15;
- (d) a peptide linker having an amino acid sequence selected from the group consisting of SEQ ID NOs: 1, 2, 3, 9, 10, 11 and 12;
- (e) the first polypeptide monomer of the third domain having a polypeptide sequence selected from the group consisting of SEQ ID NOs:17-24;
- (f) a peptide linker having an amino acid sequence selected from the group consisting of SEQ ID NOs: 5, 6, 7 and 8; and
- (g) the second polypeptide monomer of the third domain having a polypeptide sequence selected from the group consisting of SEQ ID NOs:17-24.

12.The antibody construct according to claim 11, having an amino acid sequence selected from the group consisting of:

- (a) SEQ ID NOs: 54, 55, 60, and 61;
- (b) SEQ ID NOs: 72, 73, 78, and 79;
- (c) SEQ ID NOs: 90, 91, 96, 97, 108, 109, 114, and 115;
- (d) SEQ ID NOs: 144, 145, 150, 151, 162, 163, 168, 169, 180, 181, 186, and 187;
- (e) SEQ ID NOs: 198, 199, 204, 205, 216, 217, 222, 223, 234, 235, 240, and 241;
- (f) SEQ ID NOs: 252, 306, 257, 307, 268, 308, 273, 309, 284, 310, 289, 311, 300, 312, 305, and 313;
- (g) SEQ ID NOs: 324, 554, 329, 555, 340, 556, 345, 557, 356, 558, 361, 559, 372, 560, 377, 561, 388, 562, 393, 563, 404, 564, 409, 565, 420, 566, 425, 567, 436, 568, 441, 569, 452, 570, 457, 571, 468, 572, 473, 573, 484, 574, 489, 575, 500, 576, 505, 577, 516, 578, 521, 579, 532, 580, 537, 581, 548, 582, 553, and 583;

- (h) SEQ ID NOs: 594, 610, 626, 642, 658, 674, 690, 706, 722, 738, 754, 77, 786, 802, 818, 834, 850, 866, 882, 898, 914, 930, 946, 962, 978, 994, 1010, 1026, 1042, 1058, 1074, 1090, 1106, 1122, 1138, 1154, 1170, 1186, 599, 615, 631, 647, 663, 679, 695, 711, 727, 743, 759, 775, 791, 807, 823, 839, 855, 871, 887, 903, 919, 935, 951, 967, 983, 999, 1015, 1031, 1047, 1063, 1079, 1095, 1111, 1127, 1143, 1159, 1175, 1191, and 1192-1267;
- (i) SEQ ID NO: 43; and
- (j) SEQ ID Nos: 1279, 1280, 1291, 1292, 1303, 1304, 1315, 1316, 1327, 1328, 1339, 1340, 1351, 1352, 1363, 1364, 1375, 1376, 1387, 1388, 1399, 1400, 1411, 1412, 1423, 1424, 1435, 1436, 1447, 1448.

~~14~~13. A polynucleotide encoding an antibody construct as defined in any one of claims 1 to ~~13~~12.

~~15~~14. A vector comprising a polynucleotide as defined in claim ~~14~~13.

~~16~~15. A host cell transformed or transfected with the polynucleotide as defined in claim ~~14~~13 or with the vector as defined in claim ~~15~~14.

~~17~~16. A process for the production of an antibody construct according to any one of claims 1 to ~~13~~12, said process comprising culturing a host cell as defined in claim ~~14~~13 under conditions allowing the expression of the antibody construct as defined in any one of claims 1 to ~~13~~12 and recovering the produced antibody construct from the culture.

~~18~~17. A pharmaceutical composition comprising an antibody construct according to any one of claims 1 to ~~13~~12, or produced according to the process of claim ~~17~~16.

~~19~~18. The pharmaceutical composition of claims ~~18~~17, which is stable for at least four weeks at about -20°C.

~~20~~19. The antibody construct according to any one of claims 1 to ~~13~~, ~~or produced according to the process of claim 17, 12~~, for use in the prevention, treatment or amelioration of a disease selected from a proliferative disease, a tumorous disease, a viral disease or an immunological disorder.

~~2120.~~ A method for the treatment or amelioration of a proliferative disease, a tumorous disease, a viral disease or an immunological disorder, comprising the step of administering to a subject in need thereof Use of the antibody construct according to any one of claims 1 to 13, or produced according to the process of claim 17 in the manufacture of a medicament for the treatment or amelioration of a proliferative disease, a tumorous disease, a viral disease or an immunological disorder.

~~2221.~~ A kit comprising an antibody construct according to any one of claims 1 to 13, ~~or produced according to the process of claim 17~~12, a polynucleotide as defined in claim ~~14~~13, a vector as defined in claim ~~15~~14, and/or a host cell as defined in claim ~~16~~15.