

embodiment has a higher affinity towards human prostate specific membrane antigen than that of a binding protein which has the sequence set forth in SEQ ID NO: 4. In some cases, the prostate specific membrane antigen binding protein of the above embodiment further has a higher affinity towards cynomolgus prostate specific membrane antigen than that of a binding protein which has the sequence set forth in SEQ ID NO: 4.

[0012] One embodiment provides a method for the treatment or amelioration of prostate cancer, the method comprising administration of the PSMA binding protein comprising complementarity determining regions CDR1, CDR2, and CDR3, wherein (a) the amino acid sequence of CDR1 is as set forth in RFMISX₁YX₂MH (SEQ ID NO: 1); (b) the amino acid sequence of CDR2 is as set forth in X₃INPAX₄X₅TDYAEX₆VKG (SEQ ID NO: 2); and (c) the amino acid sequence of CDR3 is as set forth in DX₇YGY (SEQ ID NO: 3), to a subject in need thereof.

[0013] In some embodiments the PSMA binding protein is a single domain antibody. In some embodiments, said single domain antibody is part of a trispecific antibody.

[0013a] Another embodiment of the invention provides a prostate specific membrane antigen binding protein, comprising complementarity determining regions CDR1, CDR2, and CDR3, wherein (a) the amino acid sequence of CDR1 is selected from the group consisting of SEQ ID Nos. 5-7, and 16; (b) the amino acid sequence of CDR2 is selected from the group consisting of SEQ ID Nos. 8-14, and 17; and (c) the amino acid sequence of CDR3 is selected from the group consisting of SEQ ID Nos. 15 and 18.

[0013b] Another embodiment of the invention provides the PSMA binding protein as disclosed herein for use in therapy.

[0013c] Another embodiment of the invention provides a use of the PSMA binding protein as disclosed herein in the manufacture of a medicament for the treatment or amelioration of a proliferative disease, a tumorous disease, an inflammatory disease, an immunological disorder, an autoimmune disease, an infectious disease, a viral disease, an allergic reaction, a parasitic reaction, a graft-versus-host disease or a host-versus-graft disease, wherein the medicament is to be administered to a subject in need thereof

BRIEF DESCRIPTION OF THE DRAWINGS

[0014] The novel features of the invention are set forth with particularity in the appended claims. A better understanding of the features and advantages of the present invention will be obtained by reference to the following detailed description that sets forth illustrative embodiments, in which the principles of the invention are utilized, and the accompanying drawings of which:

[0015] **Figure 1** is schematic representation of an exemplary PSMA targeting trispecific antigen-binding protein where the protein has an constant core element comprising an anti-CD3 ϵ single chain variable fragment (scFv) and an anti-HSA variable heavy chain region; and a PSMA binding domain that can be a VH, scFv, a non-Ig binder, or ligand.

[0016] **Figures 2A-B** compare the ability of exemplary PSMA targeting trispecific proteins (PSMA targeting TriTAC molecules) with different affinities for CD3 to induce T cells to kill human prostate cancer cells. **Figure 2A** shows killing by different PSMA targeting TriTAC molecules in prostate cancer model LNCaP. **Figure 2B** shows killing by different PSMA targeting TriTAC molecules in prostate cancer model 22Rv1. **Figure 2C** shows EC50 values for PSMA targeting TriTAC in LNCaP and 22Rv1 prostate cancer models.

[0017] **Figure 3** shows the serum concentration of PSMA targeting TriTAC C236 in Cynomolgus monkeys after i.v. administration (100 μ g/kg) over three weeks.

CLAIMS

WHAT IS CLAIMED IS:

1. A prostate specific membrane antigen binding protein, comprising complementarity determining regions CDR1, CDR2, and CDR3, wherein
 - (a) the amino acid sequence of CDR1 is selected from the group consisting of SEQ ID Nos. 5-7, and 16;
 - (b) the amino acid sequence of CDR2 is selected from the group consisting of SEQ ID Nos. 8-14, and 17; and
 - (c) the amino acid sequence of CDR3 is selected from the group consisting of SEQ ID Nos. 15 and 18.
2. The prostate specific membrane antigen binding protein of claim 1, wherein said protein comprises the following formula:
$$f_1-r_1-f_2-r_2-f_3-r_3-f_4$$
wherein, r₁ is the CDR1; r₂ is the CDR2; and r₃ is the CDR3; and wherein f₁, f₂, f₃ and f₄ are framework residues selected so that said protein is at least eighty percent identical to the amino acid sequence set forth in SEQ ID NO: 4.
3. The prostate specific membrane antigen binding protein of claim 1 or 2, wherein the CDR1 comprises SEQ ID NO: 5, SEQ ID NO: 6, or SEQ ID NO: 7.
4. The prostate specific membrane antigen binding protein of any one of claims 1-3, wherein r₂ comprises SEQ ID NO: 8, SEQ ID NO: 9, SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, SEQ ID NO: 13, or SEQ ID NO: 14.
5. The prostate specific membrane antigen binding protein of any one of claims 1-4, wherein r₃ comprises SEQ ID NO: 15.
6. The prostate specific membrane antigen binding protein of any one of claims 1-5, wherein the protein comprises a sequence selected from the group consisting of SEQ ID Nos. 21-32.
7. A prostate specific membrane antigen binding protein comprising CDR1, CDR2, and CDR3, comprising the sequence set forth as SEQ ID NO: 4 wherein one or more amino acid residues selected from amino acid positions 31, 33, 50, 55, 56, 62, and 97 are substituted.
8. The prostate specific membrane antigen binding protein of claim 7, comprising one or more additional substitutions at amino acid positions other than positions 31, 33, 50, 55, 56, 62, and 97.

9. The prostate specific membrane antigen binding protein of any one of claims 1-8, wherein said binding protein binds to one or both of human prostate specific membrane antigen and cynomolgus prostate specific membrane antigen.
10. The prostate specific membrane antigen binding protein of any one of claims 1-9, wherein said binding protein binds to human prostate specific membrane antigen and cynomolgus prostate specific membrane antigen with comparable binding affinities.
11. The prostate specific membrane antigen binding protein of any one of claims 1-10, wherein said binding protein binds to human prostate specific membrane antigen with a higher binding affinity than cynomolgus prostate specific membrane antigen.
12. A polynucleotide encoding a PSMA binding protein according to any one of claims 1-11.
13. A vector comprising the polynucleotide of claim 12.
14. A host cell transformed with the vector according to claim 13.
15. A pharmaceutical composition comprising (i) a PSMA binding protein according to any one of claims 1-13, the polynucleotide according to claim 12, the vector according to claim 13 or the host cell according to claim 14, and (ii) a pharmaceutically acceptable carrier.
16. Use of the PSMA binding protein according to any one of claims 1-11 in the manufacture of a medicament for the treatment or amelioration of a proliferative disease, a tumorous disease, an inflammatory disease, an immunological disorder, an autoimmune disease, an infectious disease, a viral disease, an allergic reaction, a parasitic reaction, a graft-versus-host disease or a host-versus-graft disease, wherein the medicament is to be administered to a subject in need thereof.
17. The use of claim 16, wherein the subject is human.
18. The use of claim 17, wherein an agent is to be further administered in combination with the medicament.
19. A multispecific binding protein comprising the PSMA binding protein according to any one of claim 1, or claim 7.
20. The PSMA binding protein according to any one of claims 1-11 for use in therapy.