

MULTI-SPECIFIC ANTIBODIES BINDING TO BCMA

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority benefit of the filing date of US Provisional Patent Application Serial No. 63/046,477, filed on June 30, 2020, the disclosure of which is incorporated by reference herein in its entirety.

FIELD OF THE INVENTION

[0002] The present invention concerns multi-specific, human heavy chain antibodies (e.g., UniAbs™) binding to BCMA. The invention further concerns methods of making such antibodies, compositions, including pharmaceutical compositions, comprising such antibodies, and their use to treat disorders that are characterized by the expression of BCMA.

BACKGROUND OF THE INVENTION

B-Cell Maturation Antigen (BCMA)

[0003] BCMA, also known as tumor necrosis factor superfamily member 17 (TNFRSF17) (UniProt Q02223), is a cell surface receptor exclusively expressed on plasma cells and plasmablasts. BCMA is a receptor for two ligands in the tumor necrosis factor (TNF) superfamily: APRIL (a proliferation-inducing ligand, also known as TNFSF13; TALL-2 and TRDL-1; the high affinity ligand for BCMA) and B cell activation factor (BAFF) (also known as BLyS; TALL-1; THANK; zTNF4; TNFSF20; and D8Ertd387e; the low affinity ligand for BCMA). APRIL and BAFF are growth factors that bind BCMA and promote survival of plasma cells. BCMA is also highly expressed on malignant plasma cells in human multiple myeloma (MM). Antibodies binding to BCMA are described, for example, in Gras et al., 1995, Int. Immunol. 7:1093-1106, WO200124811 and WO200124812. Anti-BCMA antibodies that cross-react with TACI are described in WO2002/066516. Bispecific antibodies against BCMA and CD3 are described, for example, in US 2013/0156769 A1 and US 2015/0376287 A1. An anti-BCMA antibody-MMAE or -MMAF conjugate has been reported to selectively induce killing of multiple myeloma cells (Tai et al., Blood 2014, 123(20): 3128-38). Ali et al., Blood 2016, 128(13):1688-700, have reported that in a clinical trial (#NCT02215967) chimeric antigen receptor (CAR) T cells targeting BCMA resulted in remission of multiple myeloma in human patients.

Heavy Chain Antibodies

- [0004] In a conventional IgG antibody, the association of the heavy chain and light chain is due in part to a hydrophobic interaction between the light chain constant region and the CH1 constant domain of the heavy chain. There are additional residues in the heavy chain framework 2 (FR2) and framework 4 (FR4) regions that also contribute to this hydrophobic interaction between the heavy and light chains.
- [0005] It is known, however, that sera of camelids (sub-order Tylopoda which includes camels, dromedaries and llamas) contain a major type of antibodies composed solely of paired H-chains (heavy-chain only antibodies or UniAbs™). The UniAbs™ of *Camelidae* (*Camelus dromedarius*, *Camelus bactrianus*, *Lama glama*, *Lama guanaco*, *Lama alpaca* and *Lama vicugna*) have a unique structure consisting of a single variable domain (VHH), a hinge region and two constant domains (CH2 and CH3), which are highly homologous to the CH2 and CH3 domains of classical antibodies. These UniAbs™ lack the first domain of the constant region (CH1) which is present in the genome, but is spliced out during mRNA processing. The absence of the CH1 domain explains the absence of the light chain in the UniAbs™, since this domain is the anchoring place for the constant domain of the light chain. Such UniAbs™ naturally evolved to confer antigen-binding specificity and high affinity by three CDRs from conventional antibodies or fragments thereof (Muyldermans, 2001; *J Biotechnol* 74:277-302; Revets et al., 2005; *Expert Opin Biol Ther* 5:111-124). Cartilaginous fish, such as sharks, have also evolved a distinctive type of immunoglobulin, designated as IgNAR, which lacks the light polypeptide chains and is composed entirely by heavy chains. IgNAR molecules can be manipulated by molecular engineering to produce the variable domain of a single heavy chain polypeptide (vNARs) (Nuttall et al. *Eur. J. Biochem.* 270, 3543-3554 (2003); Nuttall et al. *Function and Bioinformatics* 55, 187-197 (2004); Dooley et al., *Molecular Immunology* 40, 25-33 (2003)).
- [0006] The ability of heavy chain-only antibodies devoid of light chain to bind antigen was established in the 1960s (Jaton et al. (1968) *Biochemistry*, 7, 4185-4195). Heavy chain immunoglobulin physically separated from light chain retained 80% of antigen-binding activity relative to the tetrameric antibody. Sitia et al. (1990) *Cell*, 60, 781-790 demonstrated that removal of the CH1 domain from a rearranged mouse μ gene results in the production of a heavy chain-only antibody, devoid of light chain, in mammalian cell culture. The antibodies produced retained VH binding specificity and effector functions.
- [0007] Heavy chain antibodies with a high specificity and affinity can be generated against a variety of antigens through immunization (van der Linden, R. H., et al. *Biochim. Biophys. Acta.* 1431, 37-46 (1999)) and the VHH portion can be readily cloned and expressed in yeast (Frenken, L. G. J., et al. *J. Biotechnol.* 78, 11-21 (2000)). Their levels of expression, solubility and stability are significantly higher than those of classical F(ab) or Fv fragments (Ghahroudi, M. A. et al. *FEBS Lett.* 414, 521-526 (1997)).

[0008] Mice in which the λ (lambda) light (L) chain locus and/or the λ and κ (kappa) L chain loci have been functionally silenced and antibodies produced by such mice are described in U.S. Patent Nos. 7,541,513 and 8,367,888. Recombinant production of heavy chain-only antibodies in mice and rats has been reported, for example, in WO2006008548; U.S. Application Publication No. 20100122358; Nguyen et al., 2003, *Immunology*, 109(1), 93-101; Brüggemann et al., *Crit. Rev. Immunol.*; 2006, 26(5):377-90; and Zou et al., 2007, *J Exp Med*; 204(13): 3271-3283. The production of knockout rats via embryo microinjections of zinc-finger nucleases is described in Geurts et al., 2009, *Science*, 325(5939):433. Soluble heavy chain-only antibodies and transgenic rodents comprising a heterologous heavy chain locus producing such antibodies are described in U. S. Patent Nos. 8,883,150 and 9,365,655. CAR-T structures comprising single-domain antibodies as binding (targeting) domain are described, for example, in Iri-Sofla et al., 2011, *Experimental Cell Research* 317:2630-2641 and Jamnani et al., 2014, *Biochim Biophys Acta*, 1840:378-386.

SUMMARY OF THE INVENTION

[0009] Aspects of the invention relate to antibodies, such as heavy chain antibodies, including, but not limited to, UniAbs™, that bind to BCMA. Further aspects of the invention relate to methods of making such antibodies, compositions comprising such antibodies, and their use in the treatment of B-cell disorders that are characterized by the expression of BCMA.

[0010] Aspects of the invention include multi-specific antibodies that bind to BCMA, comprising a first binding unit comprising a variable region comprising a CDR3 sequence having at least 85% sequence identity to SEQ ID NO: 3.

[0011] Aspects of the invention include multi-specific antibodies that bind to BCMA, comprising a first binding unit comprising a variable region comprising a CDR1 sequence, a CDR2 sequence, and a CDR3 sequence, wherein the CDR1, CDR2 and CDR3 sequences combined have at least 85% sequence identity to SEQ ID NOs: 1-3 combined.

[0012] Aspects of the invention include multi-specific antibodies that bind to BCMA, comprising a first binding unit comprising a variable region comprising: a CDR1 sequence comprising the sequence of SEQ ID NO: 1; a CDR2 sequence comprising the sequence of SEQ ID NO: 2; and a CDR3 sequence comprising the sequence of SEQ ID NO: 3.

[0013] In some embodiments, the CDR1, CDR2 and CDR3 sequences are present in a human VH framework. In some embodiments, the variable region is a heavy chain-only variable region. In some embodiments, the variable region comprises a sequence having at least 95% sequence identity to SEQ ID NO: 12. In some embodiments, the variable region comprises a sequence comprising SEQ ID NO: 12. In some embodiments, the variable region is in a monovalent or bivalent configuration.

- [0014] In some embodiments, a multi-specific antibody further comprises a heavy chain constant region sequence, in the absence of a CH1 sequence. In some embodiments, a heavy chain constant region sequence comprises a CH2 domain and a CH3 domain, but no CH1 domain. In some embodiments, the CH2 domain comprises the sequence of a wild type human IgG4 CH2 domain (SEQ ID NO: 36).
- [0015] In some embodiments, the CH2 domain comprises a variant human IgG4 CH2 domain comprising an F234A mutation, an L235A mutation, or both an F234A mutation and an L235A mutation. In some embodiments, the CH3 domain comprises the sequence of a wild type human IgG4 CH3 domain (SEQ ID NO: 38). In some embodiments, the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366W mutation (SEQ ID NO: 39). In some embodiments, the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation (SEQ ID NO: 40).
- [0016] In some embodiments, a multi-specific antibody further comprises a hinge region sequence positioned between the heavy chain-only variable region and the CH2 domain. In some embodiments, the hinge region sequence comprises the sequence of a wild type human IgG4 hinge region (SEQ ID NO: 32). In some embodiments, the hinge region sequence comprises a variant human IgG4 hinge region sequence comprising an S228P mutation (SEQ ID NO: 33).
- [0017] In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH2 domain comprising an F234A mutation and an L235A mutation. In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH3 domain comprising a T366W mutation. In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation.
- [0018] In some embodiments, a multi-specific antibody further comprises a second binding unit that binds to La protein. In some embodiments, the second binding unit comprises: (a) a heavy chain variable region comprising: (i) a CDR1 sequence comprising any one of the sequences of SEQ ID NOs: 4 or 7; and (ii) a CDR2 sequence comprising the sequence of SEQ ID NO: 5; and (iii) a CDR3 sequence comprising any one of the sequences of SEQ ID NOs: 6 or 8; and (b) a light chain variable region comprising: (i) a CDR1 sequence comprising the sequence of SEQ ID NO: 9; and (ii) a CDR2 sequence comprising the sequence of SEQ ID NO: 10; and (iii) a CDR3 sequence comprising the sequence of SEQ ID NO: 11.
- [0019] In some embodiments, the heavy chain variable region of the second binding unit comprises: (i) a CDR1 sequence comprising the sequence of SEQ ID NO: 4, a CDR2 sequence comprising the sequence of SEQ ID NO: 5, and a CDR3 sequence comprising the sequence of SEQ ID NO: 6; or (i) a CDR1 sequence comprising the sequence of SEQ ID NO: 7, a CDR2 sequence comprising the sequence of SEQ ID NO: 5, and a CDR3 sequence comprising the sequence of SEQ ID NO: 8.

[0020] In some embodiments, the CDR1, CDR2 and CDR3 sequences of the heavy chain variable region of the second binding unit are present in a human VH framework. In some embodiments, the CDR1, CDR2 and CDR3 sequences of the light chain variable region of the second binding unit are present in a human VL framework. In some embodiments, the human VL framework is a human Vkappa framework. In some embodiments, the human VL framework is a human Vlambdab framework.

[0021] In some embodiments, the second binding unit comprises a heavy chain variable region comprising a sequence having at least 95% sequence identity to SEQ ID NO: 14 and a light chain variable region comprising a sequence having at least 95% sequence identity to SEQ ID NO: 16. In some embodiments, the second binding unit comprises a heavy chain variable region comprising the sequence of SEQ ID NO: 14 and a light chain variable region comprising the sequence of SEQ ID NO: 16. In some embodiments, the second binding unit comprises a heavy chain variable region comprising a sequence having at least 95% sequence identity to SEQ ID NO: 15 and a light chain variable region comprising a sequence having at least 95% sequence identity to SEQ ID NO: 17. In some embodiments, the second binding unit comprises a heavy chain variable region comprising the sequence of SEQ ID NO: 15 and a light chain variable region comprising the sequence of SEQ ID NO: 17.

[0022] In some embodiments, the heavy chain variable region and the light chain variable region of the second binding unit are on a common polypeptide subunit of the multi-specific antibody, and are connected by a linker sequence. In some embodiments, the heavy chain variable region and the light chain variable region of the second binding unit are on different polypeptide subunits of the multi-specific antibody.

[0023] In some embodiments, the second binding unit further comprises a heavy chain constant region. In some embodiments, the heavy chain constant region comprises a CH1 domain, a hinge region, a CH2 domain, and a CH3 domain. In some embodiments, the CH2 domain comprises the sequence of a wild type human IgG4 CH2 domain (SEQ ID NO: 36). In some embodiments, the CH2 domain comprises a variant human IgG4 CH2 domain comprising an F234A mutation, an L235A mutation, or both an F234A mutation and an L235A mutation. In some embodiments, the CH3 domain comprises the sequence of a wild type human IgG4 CH3 domain (SEQ ID NO: 38). In some embodiments, the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366W mutation (SEQ ID NO: 39). In some embodiments, the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation (SEQ ID NO: 40). In some embodiments, the hinge region comprises the sequence of a wild type human IgG4 hinge region (SEQ ID NO: 32). In some embodiments, the hinge region comprises a variant human IgG4 hinge region sequence comprising an S228P mutation (SEQ ID NO: 33).

[0024] In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH2 domain comprising an F234A mutation and an L235A mutation. In some embodiments, a multi-specific

antibody further comprises a variant human IgG4 CH3 domain comprising a T366W mutation. In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation.

[0025] In some embodiments, the second binding unit further comprises a light chain constant region. In some embodiments, the light chain constant region comprises a human Vkappa constant region sequence. In some embodiments, the light chain constant region comprises a human Vlambd constant region sequence. In some embodiments, the multi-specific antibody is bispecific.

[0026] Aspects of the invention include multi-specific antibodies comprising: (a) a first binding unit that binds to La protein, comprising: (i) a heavy chain variable region comprising: a CDR1 sequence of SEQ ID NOs: 4 or 7, a CDR2 sequence of SEQ ID NO: 5, and a CDR3 sequence of SEQ ID NO: 6 or 8, in a human VH framework; and (ii) a light chain variable region comprising: a CDR1 sequence of SEQ ID NO: 9, a CDR2 sequence of SEQ ID NO: 10, and a CDR3 sequence of SEQ ID NO: 11, in a human VL framework; and (b) a second binding unit that binds to BCMA, comprising: (i) an antigen-binding domain of an anti-BCMA heavy chain-only antibody, comprising a CDR1 sequence of SEQ ID NO: 1, a CDR2 sequence of SEQ ID NO: 2, and a CDR3 sequence of SEQ ID NO: 3, in a human VH framework, wherein the antigen-binding domain of the anti-BCMA heavy chain-only antibody is in a monovalent or bivalent configuration.

[0027] In some embodiments, the heavy chain variable region and the light chain variable region of the first binding unit are on a common polypeptide subunit of the multi-specific antibody, and are connected by a linker sequence. In some embodiments, the heavy chain variable region and the light chain variable region of the first binding unit are on different polypeptide subunits of the multi-specific antibody.

[0028] In some embodiments, the heavy chain variable region of the first binding unit comprises: (i) a CDR1 sequence of SEQ ID NO: 4, a CDR2 sequence of SEQ ID NO: 5, and a CDR3 sequence of SEQ ID NO: 6; or (ii) a CDR1 sequence of SEQ ID NO: 7, a CDR2 sequence of SEQ ID NO: 5, and a CDR3 sequence of SEQ ID NO: 8. In some embodiments, the human VL framework is a human Vkappa framework. In some embodiments, the human VL framework is a human Vlambd framework. In some embodiments, the heavy chain variable region of the first binding unit comprises a sequence having at least 95% identity to SEQ ID NO: 14 or SEQ ID NO: 15. In some embodiments, the heavy chain variable region of the first binding unit comprises the sequence of SEQ ID NO: 14 or SEQ ID NO: 15. In some embodiments, the light chain variable region of the first binding unit comprises a sequence having at least 95% identity to SEQ ID NO: 16 or SEQ ID NO: 17. In some embodiments, the light chain variable region of the first binding unit comprises the sequence of SEQ ID NO: 16 or SEQ ID NO: 17. In some embodiments, the antigen-binding domain of the anti-BCMA heavy chain-only antibody comprises a variable region sequence having at least 95% identity to the sequence of SEQ ID

NO: 12. In some embodiments, the antigen-binding domain of the anti-BCMA heavy chain-only antibody comprises a variable region sequence comprising the sequence of SEQ ID NO: 12.

[0029] In some embodiments, the antigen-binding domain of the anti-BCMA heavy chain-only antibody is in a bivalent configuration, and comprises a linker sequence. In some embodiments, the linker sequence comprises a G4S linker sequence.

[0030] In some embodiments, the first binding unit further comprises a heavy chain constant region. In some embodiments, the heavy chain constant region comprises a CH1 domain, a hinge region, a CH2 domain, and a CH3 domain. In some embodiments, the CH2 domain comprises the sequence of a wild type human IgG4 CH2 domain (SEQ ID NO: 36). In some embodiments, the CH2 domain comprises a variant human IgG4 CH2 domain comprising an F234A mutation, an L235A mutation, or both an F234A mutation and an L235A mutation. In some embodiments, the CH3 domain comprises the sequence of a wild type human IgG4 CH3 domain (SEQ ID NO: 38). In some embodiments, the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366W mutation (SEQ ID NO: 39). In some embodiments, the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation (SEQ ID NO: 40). In some embodiments, the hinge region comprises the sequence of a wild type human IgG4 hinge region (SEQ ID NO: 32). In some embodiments, the hinge region comprises a variant human IgG4 hinge region sequence comprising an S228P mutation (SEQ ID NO: 33).

[0031] In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH2 domain comprising an F234A mutation and an L235A mutation. In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH3 domain comprising a T366W mutation. In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation.

[0032] In some embodiments, the first binding unit further comprises a light chain constant region. In some embodiments, the light chain constant region comprises a human Vkappa constant region sequence. In some embodiments, the light chain constant region comprises a human Vlambda constant region sequence.

[0033] In some embodiments, the second binding unit further comprises a heavy chain constant region sequence, in the absence of a CH1 sequence. In some embodiments, the heavy chain constant region sequence comprises a CH2 domain and a CH3 domain, but no CH1 domain. In some embodiments, the CH2 domain comprises the sequence of a wild type human IgG4 CH2 domain (SEQ ID NO: 36). In some embodiments, the CH2 domain comprises a variant human IgG4 CH2 domain comprising an F234A mutation, an L235A mutation, or both an F234A mutation and an L235A mutation. In some embodiments, the CH3 domain comprises the sequence of a wild type human IgG4 CH3 domain (SEQ ID NO: 38). In some embodiments, the CH3 domain comprises a variant human IgG4 CH3 domain

comprising a T366W mutation (SEQ ID NO: 39). In some embodiments, the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation (SEQ ID NO: 40). In some embodiments, the hinge region comprises the sequence of a wild type human IgG4 hinge region (SEQ ID NO: 32). In some embodiments, the hinge region comprises a variant human IgG4 hinge region sequence comprising an S228P mutation (SEQ ID NO: 33).

[0034] In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH2 domain comprising an F234A mutation and an L235A mutation. In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH3 domain comprising a T366W mutation. In some embodiments, a multi-specific antibody further comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation. In some embodiments, the multi-specific antibody is bispecific.

[0035] Aspects of the invention include bispecific three-chain antibody-like molecules that bind to BCMA and La protein, comprising: (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 18; (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 24; and (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 20.

[0036] Aspects of the invention include bispecific three-chain antibody-like molecules that bind to BCMA and La protein, comprising: (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 18; (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 26; and (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 20.

[0037] Aspects of the invention include bispecific three-chain antibody-like molecules that bind to BCMA and La protein, comprising: (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 21; (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 24; and (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 23.

[0038] Aspects of the invention include bispecific three-chain antibody-like molecules that bind to BCMA and La protein, comprising: (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 21; (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 26; and (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 23.

[0039] Aspects of the invention include bispecific three-chain antibody-like molecules that bind to BCMA and La protein, comprising: (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 19; (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 25; and (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 20.

[0040] Aspects of the invention include bispecific three-chain antibody-like molecules that bind to BCMA and La protein, comprising: (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 19; (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 27; and (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 20.

- [0041] Aspects of the invention include bispecific three-chain antibody-like molecules that bind to BCMA and La protein, comprising: (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 22; (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 25; and (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 23.
- [0042] Aspects of the invention include bispecific three-chain antibody-like molecules that bind to BCMA and La protein, comprising: (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 22; (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 27; and (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 23.
- [0043] Aspects of the invention include pharmaceutical compositions comprising an antibody as described herein.
- [0044] Aspects of the invention include methods for the treatment of a B-cell disorder characterized by expression of BCMA, comprising administering to a subject with said disorder an antibody or a pharmaceutical composition as described herein.
- [0045] Aspects of the invention include use of an antibody as described herein in the preparation of a medicament for the treatment of a B-cell disorder characterized by expression of BCMA.
- [0046] Aspects of the invention include an antibody as described herein for use in the treatment of a B-cell disorder characterized by expression of BCMA.
- [0047] In some embodiments, the disorder is multiple myeloma (MM). In some embodiments, the disorder is an autoimmune disorder. In some embodiments, the autoimmune disorder is systemic lupus erythematosus (SLE). In some embodiments, the autoimmune disorder is rheumatoid arthritis (RA). In some embodiments, the autoimmune disorder is multiple sclerosis (MS).
- [0048] Aspects of the invention include a polynucleotide encoding an antibody as described herein. A vector comprising a polynucleotide as described herein, and a cell comprising a vector as described herein.
- [0049] Aspects of the invention include methods of producing an antibody as described herein, comprising growing a cell as described herein under conditions permissive for expression of the antibody, and isolating the antibody from the cell.
- [0050] Aspects of the invention include methods of making an antibody as described herein, comprising immunizing a UniRat animal with BCMA and identifying BCMA-binding heavy chain sequences.
- [0051] Aspects of the invention include methods of treatment, comprising administering to an individual in need an effective dose of an antibody or pharmaceutical composition as described herein.
- [0052] These and further aspects will be further explained in the rest of the disclosure, including the Examples.

BRIEF DESCRIPTION OF THE DRAWINGS

- [0053] FIGS. 1-6 are tables providing the amino acid sequences of the indicated proteins.
- [0054] FIG. 7, Panel A, is a schematic illustration of a three-chain antibody-like molecule comprising three polypeptide subunits. A first binding unit is formed from the first light chain polypeptide subunit and the first heavy chain polypeptide subunit. The second heavy chain polypeptide comprises a heavy chain-only variable region domain in a bivalent configuration, which provides a second binding unit.
- [0055] FIG. 7, Panel B, is a schematic illustration of a three-chain antibody-like molecule comprising three polypeptide subunits. A first binding unit is formed from the first light chain polypeptide subunit and the first heavy chain polypeptide subunit. The second heavy chain polypeptide comprises a heavy chain-only variable region domain in a monovalent configuration, which provides a second binding unit.
- [0056] FIG. 8, Panel A is a schematic diagram of a CAR-T structure comprising an anti-BCMA extracellular binding domain, as described herein.
- [0057] FIG. 8, Panel B is a graph showing antigen-specific binding and activation of CAR constructs comprising an anti-BCMA extracellular binding domain, as described herein, tested in the indicated cell lines.
- [0058] FIG. 9, Panel A is a graph showing mean fluorescence intensity (MFI) for test antibodies binding to MM1.S cells.
- [0059] FIG. 9, Panel B is a graph showing mean fluorescence intensity (MFI) for test antibodies binding to H929 cells.
- [0060] FIG. 10, Panel A is a graph showing A450-A570 values indicating AntiX**BCMA_F7E binding to BCMA in the presence of BCMA agonist, APRIL.
- [0061] FIG. 10, Panel B is a graph showing A450-A570 values indicating AntiX**BCMA_F7E_F7E binding to BCMA in the presence of BCMA agonist, APRIL.
- [0062] FIG. 10, Panel C is a graph showing A450-A570 values indicating AntiX**GP120_F8A binding to BCMA in the presence of BCMA agonist, APRIL.

DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENTS

- [0063] The practice of the present invention will employ, unless otherwise indicated, conventional techniques of molecular biology (including recombinant techniques), microbiology, cell biology, biochemistry, and immunology, which are within the skill of the art. Such techniques are explained fully in the literature, such as, "Molecular Cloning: A Laboratory Manual", second edition (Sambrook et al., 1989); "Oligonucleotide Synthesis" (M. J. Gait, ed., 1984); "Animal Cell Culture" (R. I. Freshney, ed., 1987); "Methods in Enzymology" (Academic Press, Inc.); "Current Protocols in Molecular Biology" (F. M. Ausubel et al., eds., 1987, and periodic updates); "PCR: The Polymerase Chain Reaction", (Mullis et al., ed., 1994); "A Practical Guide to Molecular Cloning" (Perbal Bernard V., 1988); "Phage

Display: A Laboratory Manual” (Barbas et al., 2001); Harlow, Lane and Harlow, Using Antibodies: A Laboratory Manual: Portable Protocol No. I, Cold Spring Harbor Laboratory (1998); and Harlow and Lane, Antibodies: A Laboratory Manual, Cold Spring Harbor Laboratory; (1988).

[0064] Where a range of values is provided, it is understood that each intervening value, to the tenth of the unit of the lower limit unless the context clearly dictates otherwise, between the upper and lower limit of that range and any other stated or intervening value in that stated range is encompassed within the invention. The upper and lower limits of these smaller ranges may independently be included in the smaller ranges is also encompassed within the invention, subject to any specifically excluded limit in the stated range. Where the stated range includes one or both of the limits, ranges excluding either or both of those included limits are also included in the invention.

[0065] Unless indicated otherwise, antibody residues herein are numbered according to the Kabat numbering system (*e.g.*, Kabat *et al.*, Sequences of Immunological Interest. 5th Ed. Public Health Service, National Institutes of Health, Bethesda, Md. (1991)).

[0066] In the following description, numerous specific details are set forth to provide a more thorough understanding of the present invention. However, it will be apparent to one of skill in the art that the present invention may be practiced without one or more of these specific details. In other instances, well-known features and procedures well known to those skilled in the art have not been described in order to avoid obscuring the invention.

[0067] All references cited throughout the disclosure, including patent applications and publications, are incorporated by reference herein in their entirety.

I. Definitions

[0068] By “comprising” it is meant that the recited elements are required in the composition/method/kit, but other elements may be included to form the composition/method/kit etc. within the scope of the claim.

[0069] By “consisting essentially of”, it is meant a limitation of the scope of composition or method described to the specified materials or steps that do not materially affect the basic and novel characteristic(s) of the subject invention.

[0070] By “consisting of”, it is meant the exclusion from the composition, method, or kit of any element, step, or ingredient not specified in the claim.

[0071] Antibody residues herein are numbered according to the Kabat numbering system and the EU numbering system. The Kabat numbering system is generally used when referring to a residue in the variable domain (approximately residues 1-113 of the heavy chain) (*e.g.*, Kabat *et al.*, Sequences of Immunological Interest. 5th Ed. Public Health Service, National Institutes of Health, Bethesda, Md. (1991)). The “EU numbering system” or “EU index” is generally used when referring to a residue in an immunoglobulin heavy chain constant region (*e.g.*, the EU index reported in Kabat et al., *supra*). The

“EU index as in Kabat” refers to the residue numbering of the human IgG1 EU antibody. Unless stated otherwise herein, references to residue numbers in the variable domain of antibodies mean residue numbering by the Kabat numbering system. Unless stated otherwise herein, references to residue numbers in the constant domain of antibodies mean residue numbering by the EU numbering system.

[0072] Antibodies, also referred to as immunoglobulins, conventionally comprise at least one heavy chain and one light chain, where the amino terminal domain of the heavy and light chains is variable in sequence, hence is commonly referred to as a variable region domain, or a variable heavy (VH) or variable light (VL) domain. The two domains conventionally associate to form a specific binding region, although as will be discussed here, specific binding can also be obtained with heavy chain-only variable sequences, and a variety of non-natural configurations of antibodies are known and used in the art.

[0073] A “functional” or “biologically active” antibody or antigen-binding molecule (including heavy chain-only antibodies and multi-specific (e.g., bispecific) three-chain antibody-like molecules (TCAs), described herein) is one capable of exerting one or more of its natural activities in structural, regulatory, biochemical or biophysical events. For example, a functional antibody or other binding molecule, e.g., a TCA, may have the ability to specifically bind an antigen and the binding may in turn elicit or alter a cellular or molecular event such as signal transduction or enzymatic activity. A functional antibody or other binding molecule, e.g., a TCA, may also block ligand activation of a receptor or act as an agonist or antagonist. The capability of an antibody or other binding molecule, e.g., a TCA, to exert one or more of its natural activities depends on several factors, including proper folding and assembly of the polypeptide chains.

[0074] The term “antibody” herein is used in the broadest sense and specifically covers monoclonal antibodies, polyclonal antibodies, monomers, dimers, multimers, multi-specific antibodies (e.g., bispecific antibodies), heavy chain-only antibodies, three chain antibodies, three-chain antibody-like molecules (TCAs), single chain Fv (scFv), nanobodies, etc., and also includes antibody fragments, so long as they exhibit the desired biological activity (Miller et al (2003) *Jour. of Immunology* 170:4854-4861). Antibodies may be murine, human, humanized, chimeric, or derived from other species.

[0075] The term antibody may reference a full-length heavy chain, a full length light chain, an intact immunoglobulin molecule, or an immunologically active portion of any of these polypeptide subunits, i.e., a polypeptide that comprises an antigen binding site that immunospecifically binds an antigen of a target of interest or part thereof, such targets including, but not limited to, a cancer cell, or cells that produce autoimmune antibodies associated with an autoimmune disease. The immunoglobulin disclosed herein can be of any type (e.g., IgG, IgE, IgM, IgD, and IgA), class (e.g., IgG1, IgG2, IgG3, IgG4, IgA1 and IgA2) or subclass of immunoglobulin molecule, including engineered subclasses with altered Fc portions that provide for reduced or enhanced effector cell activity. Light chains of the subject

antibodies can be kappa light chains (Vkappa) or lambda light chains (Vlambda). The immunoglobulins can be derived from any species. In one aspect, the immunoglobulin is of largely human origin.

[0076] The term “monoclonal antibody” as used herein refers to an antibody obtained from a population of substantially homogeneous antibodies, i.e., the individual antibodies comprising the population are identical except for possible naturally occurring mutations that may be present in minor amounts. Monoclonal antibodies are highly specific, being directed against a single antigenic site. Furthermore, in contrast to conventional (polyclonal) antibody preparations which typically include different antibodies directed against different determinants (epitopes), each monoclonal antibody is directed against a single determinant on the antigen. Monoclonal antibodies in accordance with the present invention can be made by the hybridoma method first described by Kohler et al. (1975) *Nature* 256:495, and can also be made via recombinant protein production methods (see, e.g., U.S. Patent No. 4,816,567), for example.

[0077] The term “variable”, as used in connection with antibodies, refers to the fact that certain portions of the antibody variable domains differ extensively in sequence among antibodies and are used in the binding and specificity of each particular antibody for its particular antigen. However, the variability is not evenly distributed throughout the variable domains of antibodies. It is concentrated in three segments called hypervariable regions both in the light chain and the heavy chain variable domains. The more highly conserved portions of variable domains are called the framework regions (FRs). The variable domains of native heavy and light chains each comprise four FRs, largely adopting a β -sheet configuration, connected by three hypervariable regions, which form loops connecting, and in some cases forming part of, the β -sheet structure. The hypervariable regions in each chain are held together in close proximity by the FRs and, with the hypervariable regions from the other chain, contribute to the formation of the antigen-binding site of antibodies (see Kabat et al., *Sequences of Proteins of Immunological Interest*, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, MD. (1991)). The constant domains are not involved directly in binding an antibody to an antigen, but exhibit various effector functions, such as participation of the antibody in antibody dependent cellular cytotoxicity (ADCC).

[0078] The term “hypervariable region” when used herein refers to the amino acid residues of an antibody which are responsible for antigen-binding. The hypervariable region generally comprises amino acid residues from a “complementarity determining region” or “CDR” (e.g., residues 31-35 (H1), 50-65 (H2) and 95-102 (H3) in the heavy chain variable domain; Kabat et al., *Sequences of Proteins of Immunological Interest*, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, MD. (1991)) and/or those residues from a “hypervariable loop” residues 26-32 (H1), 53-55 (H2) and 96-101 (H3) in the heavy chain variable domain; Chothia and Lesk *J. Mol. Biol.* 196:901-917 (1987)).

[0079] The terms “CDR”, and its plural “CDRs”, refer to a complementarity determining region (CDR) of which three make up the binding character of a light chain variable region (CDRL1, CDRL2 and CDRL3) and three make up the binding character of a heavy chain variable region (CDRH1, CDRH2 and CDRH3). CDRs contribute to the functional activity of an antibody molecule and are separated by amino acid sequences that comprise scaffolding or framework regions. The exact definitional CDR boundaries and lengths are subject to different classification and numbering systems.

[0080] Exemplary CDR designations are shown herein, however, one of skill in the art will understand that a number of definitions of the CDRs are commonly in use, including the Kabat definition (see “Zhao et al. A germline knowledge based computational approach for determining antibody complementarity determining regions.” *Mol Immunol.* 2010;47:694–700), which is based on sequence variability and is the most commonly used. The Chothia definition is based on the location of the structural loop regions (Chothia et al. “Conformations of immunoglobulin hypervariable regions.” *Nature.* 1989; 342:877–883). Alternative CDR definitions of interest include, without limitation, those disclosed by Honegger, “Yet another numbering scheme for immunoglobulin variable domains: an automatic modeling and analysis tool.” *J Mol Biol.* 2001;309:657–670; Ofran et al. “Automated identification of complementarity determining regions (CDRs) reveals peculiar characteristics of CDRs and B cell epitopes.” *J Immunol.* 2008;181:6230–6235; Almagro “Identification of differences in the specificity-determining residues of antibodies that recognize antigens of different size: implications for the rational design of antibody repertoires.” *J Mol Recognit.* 2004;17:132–143; and Padlan et al. “Identification of specificity-determining residues in antibodies.” *Faseb J.* 1995;9:133–139., each of which is herein specifically incorporated by reference.

[0081] In one particular embodiment, “CDR” means a complementarity determining region of an antibody as defined in Lefranc, MP et al., IMGT, the international ImMunoGeneTics database, *Nucleic Acids Res.*, 27:209-212 (1999).

[0082] “Framework Region” or “FR” residues are those variable domain residues other than the hypervariable region/CDR residues as herein defined.

[0083] The terms “heavy chain-only antibody,” and “heavy-chain antibody” are used interchangeably herein and refer, in the broadest sense, to antibodies lacking the light chain of a conventional antibody. The terms specifically include, without limitation, homodimeric antibodies comprising the VH antigen-binding domain and the CH2 and CH3 constant domains, in the absence of the CH1 domain; functional (antigen-binding) variants of such antibodies, soluble VH variants, Ig-NAR comprising a homodimer of one variable domain (V-NAR) and five C-like constant domains (C-NAR) and functional fragments thereof; and soluble single domain antibodies (sUniDabs™). In one embodiment, a heavy chain-only antibody is composed of the variable region antigen-binding domain composed of framework 1, CDR1, framework 2, CDR2, framework 3, CDR3, and framework 4. In another embodiment, the heavy chain-

only antibody is composed of an antigen-binding domain, at least part of a hinge region and CH2 and CH3 domains, in the absence of a CH1 domain. In another embodiment, the heavy chain-only antibody is composed of an antigen-binding domain, at least part of a hinge region, and a CH2 domain. In a further embodiment, the heavy chain-only antibody is composed of an antigen-binding domain, at least part of a hinge region, and a CH3 domain. Heavy chain-only antibodies in which the CH2 and/or CH3 domain is truncated are also included herein. In a further embodiment, the heavy chain-only antibody is composed of an antigen binding domain, and at least one CH (CH1, CH2, CH3, or CH4) domain, but no hinge region. In a further embodiment, the heavy chain-only antibody is composed of an antigen binding domain, at least one CH (CH1, CH2, CH3, or CH4) domain, and at least a portion of a hinge region. The heavy chain-only antibody can be in the form of a dimer, in which two heavy chains are disulfide bonded or otherwise, covalently or non-covalently, attached with each other. The heavy chain-only antibody may belong to the IgG subclass, but antibodies belonging to other subclasses, such as IgM, IgA, IgD and IgE subclass, are also included herein. In a particular embodiment, the heavy-chain antibody is of the IgG1, IgG2, IgG3, or IgG4 subtype, in particular the IgG1 or IgG4 subtype. In one embodiment, the heavy-chain antibody is of the IgG4 subtype, wherein one or more of the CH domains are modified to alter an effector function of the antibody. In one embodiment, the heavy-chain antibody is of the IgG1 subtype, wherein one or more of the CH domains are modified to alter an effector function of the antibody. Modifications of CH domains that alter effector function are further described herein. Non-limiting examples of heavy-chain antibodies are described, for example, in WO2018/039180, the disclosure of which is incorporated herein by reference in its entirety.

[0084] In some embodiments, the antibodies herein (e.g., heavy-chain only antibodies) are used as a binding (targeting) domain of a chimeric antigen receptor (CAR). The definition specifically includes human heavy chain-only antibodies produced by human immunoglobulin transgenic rats (UniRat™), called UniAbs™. The variable regions (VH) of UniAbs™ are called UniDabs™, and are versatile building blocks that can be linked to Fc regions or serum albumin for the development of novel therapeutics with multi-specificity, increased potency and extended half-life. Since the homodimeric UniAbs™ lack a light chain and thus a VL domain, the antigen is recognized by one single domain, i.e., the variable domain (antigen-binding domain) of the heavy chain of a heavy-chain antibody (VH or VHH). In some embodiments, the antibodies herein are multi-specific (e.g., bispecific), comprising a first binding unit with binding affinity for a first antigen of interest (e.g., an antigen on a target cell, e.g., BCMA) and a second binding unit with binding affinity for a second antigen of interest (e.g., an antigen on a universal chimeric antigen receptor (CAR) complex). As such, in some embodiments, the antibodies described herein can functionalize a universal CAR complex by providing binding affinity to a particular antigenic target (e.g., BCMA).

[0085] An “intact antibody chain” as used herein is one comprising a full length variable region and a full length constant region (Fc). An intact “conventional” antibody comprises an intact light chain and an intact heavy chain, as well as a light chain constant domain (CL) and heavy chain constant domains, CH1, hinge, CH2 and CH3 for secreted IgG. Other isotypes, such as IgM or IgA may have different CH domains. The constant domains may be native sequence constant domains (e.g., human native sequence constant domains) or amino acid sequence variants thereof. The intact antibody may have one or more “effector functions” which refer to those biological activities attributable to the Fc constant region (a native sequence Fc region or amino acid sequence variant Fc region) of an antibody. Examples of antibody effector functions include C1q binding; complement dependent cytotoxicity; Fc receptor binding; antibody-dependent cell-mediated cytotoxicity (ADCC); phagocytosis; and down regulation of cell surface receptors. Constant region variants include those that alter the effector profile, binding to Fc receptors, and the like.

[0086] Depending on the amino acid sequence of the Fc (constant domain) of their heavy chains, antibodies and various antigen-binding proteins can be provided as different classes. There are five major classes of heavy chain Fc regions: IgA, IgD, IgE, IgG, and IgM, and several of these may be further divided into “subclasses” (isotypes), e.g., IgG1, IgG2, IgG3, IgG4, IgA, and IgA2. The Fc constant domains that correspond to the different classes of antibodies may be referenced as α , δ , ϵ , γ , and μ , respectively. The subunit structures and three-dimensional configurations of different classes of immunoglobulins are well known. Ig forms include hinge-modifications or hingeless forms (Roux et al (1998) J. Immunol. 161:4083-4090; Lund et al (2000) Eur. J. Biochem. 267:7246-7256; US 2005/0048572; US 2004/0229310). The light chains of antibodies from any vertebrate species can be assigned to one of two types, called κ (kappa) and λ (lambda), based on the amino acid sequences of their constant domains. Antibodies in accordance with embodiments of the invention can comprise kappa light chain sequences or lambda light chain sequences.

[0087] A “functional Fc region” possesses an “effector function” of a native-sequence Fc region. Non-limiting examples of effector functions include C1q binding; CDC; Fc-receptor binding; ADCC; ADCP; down-regulation of cell-surface receptors (e.g., B-cell receptor), etc. Such effector functions generally require the Fc region to interact with a receptor, e.g., the Fc γ RI; Fc γ RIIA; Fc γ RIIB1; Fc γ RIIB2; Fc γ RIIIA; Fc γ RIIIB receptors, and the low affinity FcRn receptor; and can be assessed using various assays known in the art. A “dead” or “silenced” Fc is one that has been mutated to retain activity with respect to, for example, prolonging serum half-life, but which does not activate a high affinity Fc receptor, or which has a reduced affinity to an Fc receptor.

[0088] A “native-sequence Fc region” comprises an amino acid sequence identical to the amino acid sequence of an Fc region found in nature. Native-sequence human Fc regions include, for example, a native-sequence human IgG1 Fc region (non-A and A allotypes); native-sequence human IgG2 Fc

region; native-sequence human IgG3 Fc region; and native-sequence human IgG4 Fc region, as well as naturally occurring variants thereof.

[0089] A “variant Fc region” comprises an amino acid sequence that differs from that of a native-sequence Fc region by virtue of at least one amino acid modification, preferably one or more amino acid substitution(s). Preferably, the variant Fc region has at least one amino acid substitution compared to a native-sequence Fc region or to the Fc region of a parent polypeptide, e.g., from about one to about ten amino acid substitutions, and preferably from about one to about five amino acid substitutions in a native-sequence Fc region or in the Fc region of the parent polypeptide. The variant Fc region herein will preferably possess at least about 80% homology with a native-sequence Fc region and/or with an Fc region of a parent polypeptide, and most preferably at least about 90% homology therewith, more preferably at least about 95% homology therewith.

[0090] Variant Fc sequences may include three amino acid substitutions in the CH2 region to reduce FcγRI binding at EU index positions 234, 235, and 237 (see Duncan et al., (1988) *Nature* 332:563). Two amino acid substitutions in the complement C1q binding site at EU index positions 330 and 331 reduce complement fixation (see Tao et al., *J. Exp. Med.* 178:661 (1993) and Canfield and Morrison, *J. Exp. Med.* 173:1483 (1991)). Substitution into human IgG1 or IgG2 residues at positions 233-236 and IgG4 residues at positions 327, 330 and 331 greatly reduces ADCC and CDC (see, for example, Armour KL, *et al.*, 1999 *Eur J Immunol.* 29(8):2613-24; and Shields RL, *et al.*, 2001. *J Biol Chem.* 276(9):6591-604). The human IgG4 Fc amino acid sequence (UniProtKB No. P01861) is provided herein as SEQ ID NO: 28. Silenced IgG1 is described, for example, in Boesch, A.W., et al., “Highly parallel characterization of IgG Fc binding interactions.” *MAbs*, 2014. 6(4): p. 915-27, the disclosure of which is incorporated herein by reference in its entirety.

[0091] Other Fc variants are possible, including, without limitation, one in which a region capable of forming a disulfide bond is deleted, or in which certain amino acid residues are eliminated at the N-terminal end of a native Fc, or a methionine residue is added thereto. Thus, in some embodiments, one or more Fc portions of a binding compound can comprise one or more mutations in the hinge region to eliminate disulfide bonding. In yet another embodiment, the hinge region of an Fc can be removed entirely. In still another embodiment, a binding compound can comprise an Fc variant.

[0092] Further, an Fc variant can be constructed to remove or substantially reduce effector functions by substituting (mutating), deleting or adding amino acid residues to effect complement binding or Fc receptor binding. For example, and not limitation, a deletion may occur in a complement-binding site, such as a C1q-binding site. Techniques for preparing such sequence derivatives of the immunoglobulin Fc fragment are disclosed in International Patent Publication Nos. WO 97/34631 and WO 96/32478. In addition, the Fc domain may be modified by phosphorylation, sulfation, acylation, glycosylation, methylation, farnesylation, acetylation, amidation, and the like.

- [0093] In some embodiments, an antibody comprises a hinge region sequence of a wild type human IgG4 (SEQ ID NO: 32). In some embodiments, an antibody comprises a variant human IgG4 hinge region sequence comprising the mutation S228P (SEQ ID NO: 33).
- [0094] In some embodiments, an antibody comprises a wild type human IgG4 CH2 domain sequence (SEQ ID NO: 36). In some embodiments, an antibody comprises a variant human IgG4 CH2 domain sequence comprising an F234A mutation, an L235A mutation, or both an F234A mutation and an L235A mutation (SEQ ID NO: 37).
- [0095] In some embodiments, an antibody comprises a wild type human IgG4 CH3 domain sequence (SEQ ID NO: 38). In some embodiments, an antibody comprises a variant human IgG4 CH3 domain sequence comprising a T366W mutation (SEQ ID NO: 39), which can optionally be referred to herein as an IgG4 CH3 knob sequence. In some embodiments, an antibody comprises a variant human IgG4 CH3 domain sequence comprising a T366S mutation, an L368A mutation, and a Y407V mutation (SEQ ID NO: 40), which can optionally be referred to herein as an IgG4 CH3 hole sequence. The IgG4 CH3 mutations described herein can be utilized in any suitable manner so as to place a “knob” on a first heavy chain constant region of a first monomer in an antibody dimer, and a “hole” on a second heavy chain constant region of a second monomer in an antibody dimer, thereby facilitating proper pairing (heterodimerization) of the desired pair of heavy chain polypeptide subunits in the antibody.
- [0096] The above-identified hinge region, CH2 domain, and CH3 domain mutations can be incorporated into the antibodies of the invention in any combination. In some embodiments, an antibody comprises a heavy chain polypeptide subunit comprising a variant human IgG4 Fc region comprising an S228P mutation, an F234A mutation, an L235A mutation, and a T366W mutation (knob). In some embodiments, an antibody comprises a heavy chain polypeptide subunit comprising a variant human IgG4 Fc region comprising an S228P mutation, an F234A mutation, an L235A mutation, a T366S mutation, an L368A mutation, and a Y407V mutation (hole).
- [0097] The term “Fc-region-comprising antibody” refers to an antibody that comprises an Fc region. The C-terminal lysine (residue 447 according to the EU numbering system) of the Fc region may be removed, for example, during purification of the antibody or by recombinant engineering of the nucleic acid encoding the antibody. Accordingly, an antibody having an Fc region according to this invention can comprise an antibody with or without K447.
- [0098] Aspects of the invention include binding compounds having multi-specific configurations, which include, without limitation, bispecific, trispecific, etc. A large variety of methods and protein configurations are known and used in bispecific monoclonal antibodies (BsMAB), tri-specific antibodies, etc.
- [0099] Aspects of the invention include antibodies comprising a heavy chain-only variable region in a monovalent or bivalent configuration. As used herein, the term “monovalent configuration” as used in

reference to a heavy chain-only variable region domain means that only one heavy chain-only variable region domain is present, having a single binding site (see FIG. 7, Panel B). In contrast, the term “bivalent configuration” as used in reference to a heavy chain-only variable region domain means that two heavy chain-only variable region domains are present (each having a single binding site), and are connected by a linker sequence (see FIG. 7, Panel A). Non-limiting examples of linker sequences are discussed further herein, and include, without limitation, GS linker sequences of various lengths. When a heavy chain-only variable region is in a bivalent configuration, each of the two heavy chain-only variable region domains can have binding affinity to the same antigen, or to different antigens (e.g., to different epitopes on the same protein; to two different proteins, etc.). However, unless specifically noted otherwise, a heavy chain-only variable region denoted as being in a “bivalent configuration” is understood to contain two identical heavy chain-only variable region domains, connected by a linker sequence, wherein each of the two identical heavy chain-only variable region domains has binding affinity to the same target epitope.

[0100] Various methods for the production of multivalent artificial antibodies have been developed by recombinantly fusing variable domains of two or more antibodies. In some embodiments, a first and a second antigen-binding domain on a polypeptide are connected by a polypeptide linker. One non-limiting example of such a polypeptide linker is a GS linker, having an amino acid sequence of four glycine residues, followed by one serine residue, and wherein the sequence is repeated n times, where n is an integer ranging from 1 to about 10, such as 2, 3, 4, 5, 6, 7, 8, or 9. Non-limiting examples of such linkers include GGGGS (SEQ ID NO: 34) ($n=1$) and GGGGSGGGGS (SEQ ID NO: 35) ($n=2$). In some embodiments, a first heavy chain-only variable region domain and a second heavy chain-only variable region domain are connected to one another on the same heavy chain polypeptide subunit of an antibody to form a bivalent configuration of heavy chain-only variable region domains. In some embodiments, a heavy chain variable region and a light chain variable region are connected to one another on the same heavy chain polypeptide subunit of an antibody (i.e., are disposed on a common polypeptide subunit of the antibody) to form an scFv configuration of the heavy chain and light chain variable region domains. In some embodiments, a heavy chain variable region and a light chain variable region reside on different polypeptide subunits of an antibody to form a binding unit having a traditional antibody configuration of the heavy chain variable region and the light chain variable region. Other suitable linkers can also be used, and are described, for example, in Chen et al., *Adv Drug Deliv Rev.* 2013 October 15; 65(10): 1357-69, the disclosure of which is incorporated herein by reference in its entirety.

[0101] The term “three-chain antibody-like molecule” or “TCA” is used herein to refer to antibody-like molecules comprising, consisting essentially of, or consisting of three polypeptide subunits, two of which comprise, consist essentially of, or consist of one heavy and one light chain of a monoclonal

antibody, or functional antigen-binding fragments of such antibody chains, comprising an antigen-binding region and at least one CH domain. This heavy chain/light chain pair has binding specificity for a first antigen. The third polypeptide subunit comprises, consists essentially of, or consists of a heavy-chain only antibody comprising an Fc portion comprising CH2 and/or CH3 and/or CH4 domains, in the absence of a CH1 domain, and one or more antigen binding domains (e.g., two antigen binding domains) that binds an epitope of a second antigen or a different epitope of the first antigen, where such binding domain is derived from or has sequence identity with the variable region of an antibody heavy or light chain. Parts of such variable region may be encoded by V_H and/or V_L gene segments, D and J_H gene segments, or J_L gene segments. The variable region may be encoded by rearranged V_HDJ_H, V_LDJ_L, V_HJ_L, or V_LJ_L gene segments. A TCA protein makes use of a heavy chain-only antibody as hereinabove defined.

[0102] A TCA binding compound makes use of a “heavy chain-only antibody” or “heavy chain-antibody” or “heavy chain polypeptide” which, as used herein, mean a single chain antibody comprising heavy chain constant regions CH2 and/or CH3 and/or CH4, but no CH1 domain. In one embodiment, the heavy chain antibody is composed of an antigen-binding domain, at least part of a hinge region and CH2 and CH3 domains. In another embodiment, the heavy chain antibody is composed of an antigen-binding domain, at least part of a hinge region and a CH2 domain. In a further embodiment, the heavy chain antibody is composed of an antigen-binding domain, at least part of a hinge region and a CH3 domain. Heavy chain antibodies in which the CH2 and/or CH3 domain is truncated are also included herein. In a further embodiment, the heavy chain is composed of an antigen binding domain, and at least one CH (CH1, CH2, CH3, or CH4) domain but no hinge region. The heavy chain only antibody can be in the form of a dimer, in which two heavy chains are disulfide bonded or otherwise covalently or non-covalently attached with each other, and can optionally include an asymmetric interface between two or more of the CH domains to facilitate proper pairing between polypeptide chains. The heavy-chain antibody may belong to the IgG subclass, but antibodies belonging to other subclasses, such as IgM, IgA, IgD and IgE subclass, are also included herein. In a particular embodiment, the heavy chain antibody is of the IgG1, IgG2, IgG3, or IgG4 subtype, in particular the IgG1 subtype or the IgG4 subtype. Non-limiting examples of a TCA binding compound are described in, for example, WO2017/223111 and WO2018/052503, the disclosures of which are incorporated herein by reference in their entirety.

[0103] Heavy-chain antibodies constitute about one fourth of the IgG antibodies produced by the camelids, e.g., camels and llamas (Hamers-Casterman C., et al. *Nature*. 363, 446-448 (1993)). These antibodies are formed by two heavy chains but are devoid of light chains. As a consequence, the variable antigen binding part is referred to as the VHH domain and it represents the smallest naturally occurring, intact, antigen-binding site, being only around 120 amino acids in length (Desmyter, A., et al. *J. Biol. Chem.* 276, 26285-26290 (2001)). Heavy chain antibodies with a high specificity and affinity can be

generated against a variety of antigens through immunization (van der Linden, R. H., et al. *Biochim. Biophys. Acta.* 1431, 37-46 (1999)) and the VHH portion can be readily cloned and expressed in yeast (Frenken, L. G. J., et al. *J. Biotechnol.* 78, 11-21 (2000)). Their levels of expression, solubility and stability are significantly higher than those of classical F(ab) or Fv fragments (Ghahroudi, M. A. et al. *FEBS Lett.* 414, 521-526 (1997)). Sharks have also been shown to have a single VH-like domain in their antibodies, termed VNAR. (Nuttall et al. *Eur. J. Biochem.* 270, 3543-3554 (2003); Nuttall et al. *Function and Bioinformatics* 55, 187-197 (2004); Dooley et al., *Molecular Immunology* 40, 25-33 (2003)).

[0104] The term “BCMA” as used herein relates to human B cell maturation antigen, also known as BCMA, CD269, and TNFRSF17 (UniProt Q02223), which is a member of the tumor necrosis receptor superfamily that is preferentially expressed in differentiated plasma cells. The extracellular domain of human BCMA consists, according to UniProt of amino acids 1-54 (or 5-51). The term “BCMA” includes a BCMA protein of any human or non-human animal species, and specifically includes human BCMA as well as BCMA of non-human animals.

[0105] The term “anti-BCMA heavy chain-only antibody,” and “BCMA heavy chain-only antibody” are used herein to refer to a heavy chain-only antibody as hereinabove defined, immunospecifically binding to BCMA. The term “human BCMA” as used herein includes any variants, isoforms and species homologs of human BCMA (UniProt Q02223), regardless of its source or mode of preparation. Thus, “human BCMA” includes human BCMA naturally expressed by cells and BCMA expressed on cells transfected with the human BCMA gene.

[0106] The terms “anti-BCMA heavy chain-only antibody,” “BCMA heavy chain-only antibody,” “anti-BCMA heavy chain antibody” and “BCMA heavy chain antibody” are used herein interchangeably to refer to a heavy chain-only antibody as hereinabove defined, immunospecifically binding to BCMA, including human BCMA, as hereinabove defined. The definition includes, without limitation, human heavy chain antibodies produced by transgenic animals, such as transgenic rats or transgenic mice expressing human immunoglobulin, including UniRats™ producing human anti-BCMA UniAb™ antibodies, as hereinabove defined.

[0107] “Percent (%) amino acid sequence identity” with respect to a reference polypeptide sequence is defined as the percentage of amino acid residues in a candidate sequence that are identical with the amino acid residues in the reference polypeptide sequence, after aligning the sequences and introducing gaps, if necessary, to achieve the maximum percent sequence identity, and not considering any conservative substitutions as part of the sequence identity. Alignment for purposes of determining percent amino acid sequence identity can be achieved in various ways that are within the skill in the art, for instance, using publicly available computer software such as BLAST, BLAST-2, ALIGN or Megalign (DNASTAR) software. Those skilled in the art can determine appropriate parameters for

aligning sequences, including any algorithms needed to achieve maximal alignment over the full length of the sequences being compared. For purposes herein, however, % amino acid sequence identity values are generated using the sequence comparison computer program ALIGN-2.

[0108] An “isolated” antibody is one which has been identified and separated and/or recovered from a component of its natural environment. Contaminant components of its natural environment are materials which would interfere with diagnostic or therapeutic uses for the antibody, and may include enzymes, hormones, and other proteinaceous or nonproteinaceous solutes. In preferred embodiments, the antibody will be purified (1) to greater than 95% by weight of antibody as determined by the Lowry method, and most preferably more than 99% by weight, (2) to a degree sufficient to obtain at least 15 residues of N-terminal or internal amino acid sequence by use of a spinning cup sequenator, or (3) to homogeneity by SDS-PAGE under reducing or nonreducing conditions using Coomassie blue or, preferably, silver stain. Isolated antibody includes the antibody *in situ* within recombinant cells since at least one component of the antibody's natural environment will not be present. Ordinarily, however, isolated antibody will be prepared by at least one purification step.

[0109] Antibodies of the invention include multi-specific antibodies. Multi-specific antibodies have more than one binding specificity. The term “multi-specific” specifically includes “bispecific” and “trisppecific,” as well as higher-order independent specific binding affinities, such as higher-order polypepitopic specificity, as well as tetravalent antibodies and antibody fragments. The terms “multi-specific antibody,” “multi-specific heavy chain-only antibody,” “multi-specific heavy chain antibody,” “multi-specific UniAb™”, and “multi-specific binding compound” are used herein in the broadest sense and cover all antibodies with more than one binding specificity. The multi-specific anti-BCMA antibodies of the present invention specifically include antibodies immunospecifically binding to one single epitope on a BCMA protein, such as a human BCMA, and to an epitope on a different protein, such as, for example, a CD3 protein. The multi-specific anti-BCMA antibodies of the present invention specifically include antibodies immunospecifically binding to two or more non-overlapping epitopes on a BCMA protein, such as a human BCMA. The multi-specific anti-BCMA antibodies of the present invention also specifically include antibodies immunospecifically binding to an epitope on a BCMA protein, such as human BCMA, and to an epitope on a different protein, such as, for example, human La protein. The multi-specific anti-BCMA antibodies of the present invention also specifically include antibodies immunospecifically binding to two or more non-overlapping or partially overlapping epitopes on a BCMA protein, such as a human BCMA protein, and to an epitope on a different protein, such as, for example, a human La protein.

[0110] Antibodies of the invention include monospecific antibodies, having one binding specificity. Monospecific antibodies specifically include antibodies comprising a single binding specificity, as well as antibodies comprising more than one binding unit having the same binding specificity. The terms

“monospecific antibody,” “monospecific heavy chain-only antibody,” “monospecific heavy chain antibody,” and “monospecific UniAb™” are used herein in the broadest sense and cover all antibodies with one binding specificity. The monospecific heavy chain anti-BCMA antibodies of the present invention specifically include antibodies immunospecifically binding to one epitope on a BCMA protein, such as a human BCMA (monovalent and monospecific). The monospecific heavy chain anti-BCMA antibodies of the present invention also specifically include antibodies having more than one binding unit (e.g., multivalent antibodies) immunospecifically binding to an epitope on a BCMA protein, such as human BCMA. For example, a monospecific antibody in accordance with embodiments of the invention can include a heavy chain-only variable region comprising two heavy chain-only antigen-binding domains (i.e., an anti-BCMA heavy chain-only variable region in a tandem configuration), wherein each of the two antigen-binding domains binds to the same epitope on a BCMA protein (i.e., bivalent and monospecific).

[0111] An “epitope” is the site on the surface of an antigen molecule to which a single antibody molecule binds. Generally, an antigen has several or many different epitopes and reacts with many different antibodies. The term specifically includes linear epitopes and conformational epitopes.

[0112] “Epitope mapping” is the process of identifying the binding sites, or epitopes, of antibodies on their target antigens. Antibody epitopes may be linear epitopes or conformational epitopes. Linear epitopes are formed by a continuous sequence of amino acids in a protein. Conformational epitopes are formed of amino acids that are discontinuous in the protein sequence, but which are brought together upon folding of the protein into its three-dimensional structure.

[0113] “Polyepitopic specificity” refers to the ability to specifically bind to two or more different epitopes on the same or different target(s). As noted above, the present invention specifically includes anti-BCMA heavy chain antibodies with polyepitopic specificities, i.e., anti-BCMA heavy chain antibodies binding to one or more non-overlapping epitopes on a BCMA protein, such as a human BCMA; and anti-BCMA heavy chain antibodies binding to one or more epitopes on a BCMA protein and to an epitope on a different protein, such as, for example, human La protein. The term “non-overlapping epitope(s)” or “non-competitive epitope(s)” of an antigen is defined herein to mean epitope(s) that are recognized by one member of a pair of antigen-specific antibodies but not the other member. Pairs of antibodies, or antigen-binding regions targeting the same antigen on a multi-specific antibody, recognizing non-overlapping epitopes, do not compete for binding to that antigen and are able to bind that antigen simultaneously.

[0114] An antibody binds “essentially the same epitope” as a reference antibody, when the two antibodies recognize identical or sterically overlapping epitopes. The most widely used and rapid methods for determining whether two epitopes bind to identical or sterically overlapping epitopes are competition assays, which can be configured in all number of different formats, using either labeled

antigen or labeled antibody. Usually, the antigen is immobilized on a 96-well plate, and the ability of unlabeled antibodies to block the binding of labeled antibodies is measured using radioactive or enzyme labels.

[0115] The term “valent” as used herein refers to a specified number of binding sites in an antibody molecule.

[0116] A “monovalent” antibody has one binding site. Thus a monovalent antibody is also monospecific.

[0117] A “multi-valent” antibody has two or more binding sites. Thus, the terms “bivalent”, “trivalent”, and “tetravalent” refer to the presence of two binding sites, three binding sites, and four binding sites, respectively. Thus, a bispecific antibody according to the invention is at least bivalent and may be trivalent, tetravalent, or otherwise multi-valent. A bivalent antibody in accordance with embodiments of the invention may have two binding sites to the same epitope (i.e., bivalent, monoparatopic), or to two different epitopes (i.e., bivalent, biparatopic).

[0118] A large variety of methods and protein configurations are known and used for the preparation of bispecific monoclonal antibodies (BsMAB), tri-specific antibodies, and the like.

[0119] The term “chimeric antigen receptor” or “CAR” is used herein in the broadest sense to refer to an engineered receptor, which grafts a desired binding specificity (e.g., the antigen-binding region of a monoclonal antibody or other ligand) to membrane-spanning and intracellular-signaling domains. Typically, the receptor is used to graft the specificity of a monoclonal antibody onto a T-cell to create a chimeric antigen receptor (CAR). (*J Natl Cancer Inst*, 2015; 108(7):dvj439; and Jackson et al., *Nature Reviews Clinical Oncology*, 2016; 13:370–383). CAR-T cells are T-cells that have been genetically engineered to produce an artificial T-cell receptor for use in immunotherapy. In one embodiment, “CAR-T-cell” means a therapeutic T-cell expressing a transgene encoding one or more chimeric antigen receptors comprised minimally of an extracellular domain, a transmembrane domain, and at least one cytosolic domain.

[0120] The term “human antibody” is used herein to include antibodies having variable and constant regions derived from human germline immunoglobulin sequences. The human antibodies herein may include amino acid residues not encoded by human germline immunoglobulin sequences, e.g., mutations introduced by random or site-specific mutagenesis *in vitro* or by somatic mutation *in vivo*. The term “human antibody” specifically includes heavy chain-only antibodies having human heavy chain variable region sequences, produced by transgenic animals, such as transgenic rats or mice, in particular UniAbs™ produced by UniRats™, as defined above.

[0121] By a “chimeric antibody” or a “chimeric immunoglobulin” is meant an immunoglobulin molecule comprising amino acid sequences from at least two different Ig loci, e.g., a transgenic antibody comprising a portion encoded by a human Ig locus and a portion encoded by a rat Ig locus. Chimeric

antibodies include transgenic antibodies with non-human Fc-regions or artificial Fc-regions, and human idiotypes. Such immunoglobulins can be isolated from animals of the invention that have been engineered to produce such chimeric antibodies.

[0122] As used herein, the term “effector cell” refers to an immune cell which is involved in the effector phase of an immune response, as opposed to the cognitive and activation phases of an immune response. Some effector cells express specific Fc receptors and carry out specific immune functions. In some embodiments, an effector cell such as a natural killer cell is capable of inducing antibody-dependent cellular cytotoxicity (ADCC). For example, monocytes and macrophages, which express FcR, are involved in specific killing of target cells and presenting antigens to other components of the immune system, or binding to cells that present antigens. In some embodiments, an effector cell may phagocytose a target antigen or target cell.

[0123] “Human effector cells” are leukocytes which express receptors such as T cell receptors or FcRs and perform effector functions. Preferably, the cells express at least FcγRIII and perform ADCC effector function. Examples of human leukocytes which mediate ADCC include natural killer (NK) cells, monocytes, cytotoxic T cells and neutrophils, with NK cells being preferred. The effector cells may be isolated from a native source thereof, e.g., from blood or PBMCs as described herein.

[0124] The term “immune cell” is used herein in the broadest sense, including, without limitation, cells of myeloid or lymphoid origin, for instance lymphocytes (such as B cells and T cells including cytolytic T cells (CTLs)), killer cells, natural killer (NK) cells, macrophages, monocytes, eosinophils, polymorphonuclear cells, such as neutrophils, granulocytes, mast cells, and basophils.

[0125] Antibody “effector functions” refer to those biological activities attributable to the Fc region (a native sequence Fc region or amino acid sequence variant Fc region) of an antibody. Examples of antibody effector functions include C1q binding; complement dependent cytotoxicity (CDC); Fc receptor binding; antibody-dependent cell-mediated cytotoxicity (ADCC); phagocytosis; down regulation of cell surface receptors (e.g., B cell receptor; BCR), etc.

[0126] “Antibody-dependent cell-mediated cytotoxicity” and “ADCC” refer to a cell-mediated reaction in which nonspecific cytotoxic cells that express Fc receptors (FcRs) (e.g., Natural Killer (NK) cells, neutrophils, and macrophages) recognize bound antibody on a target cell and subsequently cause lysis of the target cell. The primary cells for mediating ADCC, NK cells, express FcγRIII only, whereas monocytes express FcγRI, FcγRII and FcγRIII. FcR expression on hematopoietic cells is summarized in Table 3 on page 464 of Ravetch and Kinet, *Annu. Rev. Immunol* 9:457-92 (1991). To assess ADCC activity of a molecule of interest, an *in vitro* ADCC assay, such as that described in US Patent No. 5,500,362 or 5,821,337 may be performed. Useful effector cells for such assays include peripheral blood mononuclear cells (PBMC) and Natural Killer (NK) cells. Alternatively, or additionally, ADCC activity

of the molecule of interest may be assessed *in vivo*, e.g., in an animal model such as that disclosed in Clynes *et al. PNAS (USA)* 95:652-656 (1998).

- [0127] “Complement dependent cytotoxicity” or “CDC” refers to the ability of a molecule to lyse a target in the presence of complement. The complement activation pathway is initiated by the binding of the first component of the complement system (C1q) to a molecule (e.g. an antibody) complexed with a cognate antigen. To assess complement activation, a CDC assay, e.g., as described in Gazzano-Santoro *et al., J. Immunol. Methods* 202:163 (1996), may be performed.
- [0128] “Binding affinity” refers to the strength of the sum total of noncovalent interactions between a single binding site of a molecule (e.g., an antibody) and its binding partner (e.g., an antigen). Unless indicated otherwise, as used herein, “binding affinity” refers to intrinsic binding affinity which reflects a 1:1 interaction between members of a binding pair (e.g., antibody and antigen). The affinity of a molecule X for its partner Y can generally be represented by the dissociation constant (Kd). Affinity can be measured by common methods known in the art. Low-affinity antibodies generally bind antigen slowly and tend to dissociate readily, whereas high-affinity antibodies generally bind antigen faster and tend to remain bound.
- [0129] As used herein, the “Kd” or “Kd value” refers to a dissociation constant determined by BioLayer Interferometry, using an Octet QK384 instrument (Fortebio Inc., Menlo Park, CA) in kinetics mode. For example, anti-mouse Fc sensors are loaded with mouse-Fc fused antigen and then dipped into antibody-containing wells to measure concentration dependent association rates (kon). Antibody dissociation rates (koff) are measured in the final step, where the sensors are dipped into wells containing buffer only. The Kd is the ratio of koff/kon. (For further details see, Concepcion, J, et al., *Comb Chem High Throughput Screen*, 12(8), 791-800, 2009).
- [0130] As used herein, the terms “specifically interacting”, “specifically binding” or “specifically bind(s)” mean that a binding domain exhibits appreciable affinity for a particular target protein or antigen and, generally, does not exhibit significant reactivity with non-target proteins or antigens. “Appreciable affinity” includes binding with an affinity of about 10^{-6} M (KD) or stronger. Preferably, binding is considered specific when binding affinity is about 10^{-12} to 10^{-8} M, 10^{-12} to 10^{-9} M, 10^{-12} to 10^{-10} M, 10^{-11} to 10^{-8} M, preferably of about 10^{-11} to 10^{-9} M. Whether a binding domain specifically reacts with or binds to a target protein or antigen can be tested readily by, inter alia, comparing the reaction of said binding domain with a target protein or antigen with the reaction of said binding domain with non-target proteins or antigens. Preferably, a binding domain of the invention does not essentially bind or is not capable of binding to non-target proteins or antigens.
- [0131] The term “does not essentially bind”, or “is not capable of binding” means that a binding domain of the present invention does not bind to a non-target protein or antigen, i.e., does not show reactivity of more than 30%, preferably not more than 20%, more preferably not more than 10%,

particularly preferably not more than 9%, 8%, 7%, 6% or 5% with non-target proteins or antigens, whereby binding to a target protein or antigen, respectively, is set to be 100%.

[0132] The terms “treatment”, “treating” and the like are used herein to generally mean obtaining a desired pharmacologic and/or physiologic effect. The effect may be prophylactic in terms of completely or partially preventing a disease or symptom thereof and/or may be therapeutic in terms of a partial or complete cure for a disease and/or adverse effect attributable to the disease. “Treatment” as used herein covers any treatment of a disease in a mammal, and includes: (a) preventing the disease from occurring in a subject which may be predisposed to the disease but has not yet been diagnosed as having it; (b) inhibiting the disease, i.e., arresting its development; or (c) relieving the disease, i.e., causing regression of the disease. The therapeutic agent may be administered before, during or after the onset of disease or injury. The treatment of ongoing disease, where the treatment stabilizes or reduces the undesirable clinical symptoms of the patient, is of particular interest. Such treatment is desirably performed prior to complete loss of function in the affected tissues. The subject therapy may be administered during the symptomatic stage of the disease, and in some cases after the symptomatic stage of the disease.

[0133] A “therapeutically effective amount” is intended for an amount of active agent which is necessary to impart therapeutic benefit to a subject. For example, a “therapeutically effective amount” is an amount which induces, ameliorates or otherwise causes an improvement in the pathological symptoms, disease progression or physiological conditions associated with a disease or which improves resistance to a disorder.

[0134] The terms “B-cell neoplasms” or “mature B-cell neoplasms” in the context of the present invention include, but are not limited to, all lymphoid leukemias and lymphomas, chronic lymphocytic leukemia, acute lymphoblastic leukemia, prolymphocytic leukemia, precursor B-lymphoblastic leukemia, hair cell leukemia, small lymphocytic lymphoma, B-cell prolymphocytic lymphoma, B-cell chronic lymphocytic leukemia, mantle cell lymphoma, Burkitt's lymphoma, follicular lymphoma, diffuse large B-cell lymphoma (DLBCL), multiple myeloma, lymphoplasmacytic lymphoma, splenic marginal zone lymphoma, plasma cell neoplasms, such as plasma cell myeloma, plasmacytoma, monoclonal immunoglobulin deposition disease, heavy chain disease, MALT lymphoma, nodal marginal B cell lymphoma, intravascular large B cell lymphoma, primary effusion lymphoma, lymphomatoid granulomatosis, non-Hodgkins lymphoma, Hodgkins lymphoma, hairy cell leukemia, primary effusion lymphoma and AIDS-related non-Hodgkins lymphoma.

[0135] The term “characterized by expression of BCMA” broadly refers to any disease or disorder in which BCMA expression is associated with or involved with one or more pathological processes that are characteristic of the disease or disorder. Such disorders include, but are not limited to, B-cell neoplasms.

[0136] The terms “subject,” “individual,” and “patient” are used interchangeably herein to refer to a mammal being assessed for treatment and/or being treated. In an embodiment, the mammal is a human. The terms “subject,” “individual,” and “patient” encompass, without limitation, individuals having cancer, individuals with autoimmune diseases, with pathogen infections, and the like. Subjects may be human, but also include other mammals, particularly those mammals useful as laboratory models for human disease, e.g., mouse, rat, etc.

[0137] The term “pharmaceutical formulation” refers to a preparation which is in such form as to permit the biological activity of the active ingredient to be effective, and which contains no additional components which are unacceptably toxic to a subject to which the formulation would be administered. Such formulations are sterile. “Pharmaceutically acceptable” excipients (vehicles, additives) are those which can reasonably be administered to a subject mammal to provide an effective dose of the active ingredient employed.

[0138] A “sterile” formulation is aseptic or free or essentially free from all living microorganisms and their spores. A “frozen” formulation is one at a temperature below 0 °C.

[0139] A “stable” formulation is one in which the protein therein essentially retains its physical stability and/or chemical stability and/or biological activity upon storage. Preferably, the formulation essentially retains its physical and chemical stability, as well as its biological activity upon storage. The storage period is generally selected based on the intended shelf-life of the formulation. Various analytical techniques for measuring protein stability are available in the art and are reviewed in Peptide and Protein Drug Delivery, 247-301. Vincent Lee Ed., Marcel Dekker, Inc., New York, N.Y., Pubs. (1991) and Jones. A. Adv. Drug Delivery Rev. 10: 29-90) (1993), for example. Stability can be measured at a selected temperature for a selected time period. Stability can be evaluated qualitatively and/or quantitatively in a variety of different ways, including evaluation of aggregate formation (for example using size exclusion chromatography, by measuring turbidity, and/or by visual inspection); by assessing charge heterogeneity using cation exchange chromatography, image capillary isoelectric focusing (icIEF) or capillary zone electrophoresis; amino-terminal or carboxy-terminal sequence analysis; mass spectrometric analysis; SDS-PAGE analysis to compare reduced and intact antibody; peptide map (for example tryptic or LYS-C) analysis; evaluating biological activity or antigen binding function of the antibody; etc. Instability may involve any one or more of: aggregation, deamidation (e.g., Asn deamidation), oxidation (e.g., Met oxidation), isomerization (e.g., Asp isomerization), clipping/hydrolysis/fragmentation (e.g., hinge region fragmentation), succinimide formation, unpaired cysteine(s), N-terminal extension, C-terminal processing, glycosylation differences, etc.

II. Detailed Description

Anti-BCMA Antibodies

- [0140] The present invention provides antibodies, including, but not limited to, heavy chain-only antibodies (UniAbs) that bind to human BCMA. The anti-BCMA UniAbs of the invention comprise a set of CDR sequences as defined herein and shown in FIG. 1, and are exemplified by the provided heavy chain variable region (VH) sequence of SEQ ID NO: 12, set forth in FIG. 2. These antibodies provide a number of benefits that contribute to utility as clinically therapeutic agent(s). The antibodies include members with a range of binding affinities, allowing the selection of a specific sequence with a desired binding affinity.
- [0141] The anti-BCMA antibodies provided herein are not cross-reactive with the BCMA protein of *Cynomolgus macaque*, but can be engineered to provide cross-reactivity with the BCMA protein of *Cynomolgus macaque*, or with the BCMA of any other animal species, if desired.
- [0142] In some embodiments, the anti-BCMA UniAb antibodies herein comprise a VH domain, comprising CDR1, CDR2 and CDR3 sequences in a human VH framework. The CDR sequences may be situated, as an example, in the region of around amino acid residues 26-35; 53-59; and 98-117 for CDR1, CDR2 and CDR3, respectively, of the provided exemplary variable region sequence set forth in SEQ ID NO: 12. It will be understood by one of skill in the art that the CDR sequences may be in different positions if a different framework sequence is selected, although generally the order of the sequences will remain the same.
- [0143] In some embodiments, an anti-BCMA antibody comprises a variable region comprising a CDR1 sequence comprising two or fewer amino acid substitutions in the sequence of SEQ ID NO: 1, and/or a CDR2 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 2, and/or a CDR3 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 3.
- [0144] In some embodiments, an anti-BCMA antibody comprises a variable region comprising a CDR1 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 1, and a CDR2 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 2, and a CDR3 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 3.
- [0145] In some embodiments, an anti-BCMA antibody comprises a variable region comprising a CDR1 sequence of SEQ ID NO: 1, a CDR2 sequence of SEQ ID NO: 2 and a CDR3 sequence of SEQ ID NO: 3.
- [0146] In some embodiments, an anti-BCMA antibody comprises a heavy chain-only variable region comprising the CDR1, CDR2 and CDR3 sequences of SEQ ID NOs 1, 2, and 3, respectively. In some embodiments, an anti-BCMA antibody comprises a heavy chain-only variable region in a monovalent or bivalent configuration.
- [0147] In further embodiments, an anti-BCMA antibody of the present invention comprises a heavy chain variable region amino acid sequence of SEQ ID NO: 12, in a monovalent or bivalent configuration (FIG. 2).

- [0148] In some embodiments, an anti-BCMA antibody preferably comprises a CDR sequence comprising two or fewer amino acid substitutions relative to a CDR1, CDR2 and/or CDR3 sequence in any one of SEQ ID NOs: 1-3 (FIG. 1), and binds to BCMA. In some embodiments, an anti-BCMA antibody preferably comprises a heavy chain variable region sequence with at least 80% identity, at least 85% identity, at least 90% identity, at least 95% identity, at least 98% identity, or at least 99% identity to the heavy chain variable region sequence shown in FIG. 2 (SEQ ID NO: 12), and binds to BCMA.
- [0149] In some embodiments, an anti-BCMA antibody preferably comprises a heavy chain variable domain (VH) in which the CDR3 sequence has greater than or equal to 80%, such as at least 85%, at least 90%, at least 95%, or at least 99% sequence identity at the amino acid level to SEQ ID NO: 3, and binds to BCMA.
- [0150] In some embodiments, an anti-BCMA antibody preferably comprises a heavy chain variable domain (VH) in which the full set of CDRs 1, 2, and 3 (combined) has greater than or equal to eighty-five percent (85%) sequence identity at the amino acid level to the CDRs 1, 2, and 3 (combined) of SEQ ID NOs: 1-3, and binds to BCMA.
- [0151] The present invention provides multi-specific antibodies that bind to human BCMA as well as human La protein. The multi-specific antibodies of the invention can comprise a first binding unit that binds to BCMA, and a second binding unit that binds to human La protein. In some embodiments, a binding unit that binds to human La protein comprises a set of CDR sequences as defined herein and shown in FIG. 1, and are exemplified by the provided heavy chain variable region (VH) sequences of SEQ ID NOs: 14-15 and light chain variable region (VL) sequences of SEQ ID NOs: 16 and 17, set forth in FIG. 3. In some embodiments, an antibody that binds to human La protein binds to an epitope comprising the amino acid sequence KPLPEVTDEY (SEQ ID NO: 42). In some embodiments, an antibody that binds to human La protein binds to an epitope comprising the amino acid sequence VEKEALKKIHEDQQESLNKW (SEQ ID NO: 43). Antibodies that bind to human La protein are described, for example, in WO2016030414, US20200181228, US20200131262, and US20170240612, the disclosures of which are herein incorporated by reference in their entireties.
- [0152] The antibodies described herein provide a number of benefits that contribute to utility as clinically therapeutic agent(s). The antibodies include members with a range of binding affinities, allowing the selection of a specific sequence with a desired binding affinity.
- [0153] A suitable antibody may be selected from those provided herein for development and therapeutic or other use, including, without limitation, use as a bispecific or tri-specific antibody, or part of a CAR-T structure.
- [0154] Determination of affinity for a candidate protein can be performed using methods known in the art, such as Biacore measurements. The subject antibodies may have an affinity for human La protein

with a K_d of from about 10^{-6} to around about 10^{-11} , including without limitation: from about 10^{-6} to around about 10^{-10} ; from about 10^{-6} to around about 10^{-9} ; from about 10^{-6} to around about 10^{-8} ; from about 10^{-8} to around about 10^{-11} ; from about 10^{-8} to around about 10^{-10} ; from about 10^{-8} to around about 10^{-9} ; from about 10^{-9} to around about 10^{-11} ; from about 10^{-9} to around about 10^{-10} ; or any value within these ranges. The affinity selection may be confirmed with a biological assessment for modulating, e.g., increasing, a desired activity (e.g., a desired binding affinity between a universal CAR structure and a binding unit of a subject multi-specific antibody), including in vitro assays, pre-clinical models, and clinical trials, as well as assessment of potential toxicity.

[0155] In some embodiments, the anti-human La protein binding unit of a subject antibody comprises a VH domain, comprising CDR1, CDR2 and CDR3 sequences in a human VH framework, and a VL domain, comprising CDR1, CDR2 and CDR3 sequences in a human VL framework, e.g., a human V κ framework or a human V λ framework. The CDR sequences may be situated, as an example, in the region of around amino acid residues 26-35; 53-59; and 98-117 for CDR1, CDR2 and CDR3, respectively, of the provided exemplary variable region sequences set forth in SEQ ID NOs: 14-17. It will be understood by one of skill in the art that the CDR sequences may be in different positions if a different framework sequence is selected, although generally the order of the sequences will remain the same.

[0156] In some embodiments, the anti-human La protein binding unit of a subject antibody comprises a heavy chain variable region comprising a CDR1 sequence comprising two or fewer substitutions in any one of the sequences of SEQ ID NOs: 4 or 7, and/or a CDR2 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 5, and/or a CDR3 sequence comprising two or fewer substitutions in any one of the sequences of SEQ ID NOs: 6 or 8, and a light chain variable region comprising a CDR1 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 9, and/or a CDR2 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 10, and/or a CDR3 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 11.

[0157] In some embodiments, the anti-human La protein binding unit of a subject antibody comprises a heavy chain variable region comprising a CDR1 sequence comprising two or fewer substitutions in any one of the sequences of SEQ ID NOs: 4 or 7, and a CDR2 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 5, and a CDR3 sequence comprising two or fewer substitutions in any one of the sequences of SEQ ID NOs: 6 or 8, and a light chain variable region comprising a CDR1 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 9, and a CDR2 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 10, and a CDR3 sequence comprising two or fewer substitutions in the sequence of SEQ ID NO: 11.

[0158] In one embodiment, the anti-human La protein binding unit of an antibody of the present invention comprises a heavy chain comprising the CDR1 sequence of SEQ ID NO: 4; the CDR2

sequence of SEQ ID NO: 5 and a CDR3 sequence of SEQ ID NO: 6 and a light chain comprising the CDR1 sequence of SEQ ID NO: 9, the CDR2 sequence of SEQ ID NO: 10 and the CDR3 sequence of SEQ ID NO: 11. In one embodiment, the anti-human La protein binding unit of an antibody of the present invention comprises a heavy chain comprising the CDR1 sequence of SEQ ID NO: 7; the CDR2 sequence of SEQ ID NO: 5 and a CDR3 sequence of SEQ ID NO: 8 and a light chain comprising the CDR1 sequence of SEQ ID NO: 9, the CDR2 sequence of SEQ ID NO: 10 and the CDR3 sequence of SEQ ID NO: 11.

[0159] In further embodiments, the anti-human La protein binding unit of an antibody of the present invention comprises the heavy chain variable region amino acid sequence of SEQ ID NO: 14 and a light chain variable region sequence of SEQ ID NO: 16. In some embodiments, the anti-human La protein binding unit of an antibody of the present invention comprises the heavy chain variable region amino acid sequence of SEQ ID NO: 15 and a light chain variable region sequence of SEQ ID NO: 17.

[0160] In some embodiments, a heavy chain CDR sequence in the anti-human La protein binding unit of the antibodies of the present invention comprises two or fewer amino acid substitutions relative to a CDR1, CDR2 and/or CDR3 sequence in any one of SEQ ID NOs: 4-8 (FIG. 1). In some embodiments, the anti-human La protein binding unit of the antibodies of the present invention will comprise a heavy chain variable region sequence with at least 85% identity, at least 90% identity, at least 95% identity, at least 98% identity, or at least 99% identity to the heavy chain variable region sequences shown in FIG. 3 (SEQ ID NOs: 14 and 15).

[0161] In some embodiments, a light chain CDR sequence in the anti-human La protein binding unit of the antibodies of the present invention comprises two or fewer amino acid substitutions relative to a CDR1, CDR2 and/or CDR3 sequence in any one of SEQ ID NOs: 9-11 (FIG. 1). In some embodiments, the anti-human La protein binding unit of the antibodies of the present invention will comprise a light chain variable region sequence with at least 85% identity, at least 90% identity, at least 95% identity, at least 98% identity, or at least 99% identity to the light chain variable region sequences shown in FIG. 3 (SEQ ID NOs: 16 and 17).

[0162] In some embodiments, multi-specific (e.g., bispecific) antibodies are provided, which may have any of the configurations discussed herein, including, without limitation, a three chain bispecific antibody, or a three chain bispecific antibody-like molecule (a bispecific TCA). Bispecific antibodies comprise at least the heavy chain variable region of an antibody specific for a protein other than BCMA.

[0163] Where a protein of the invention is a bispecific antibody, one binding unit is specific for human BCMA, while another binding unit may be specific for target cells (e.g., effector cells, e.g., T-cells), tumor associated antigens, targeting antigens, e.g., integrins, etc., pathogen antigens, checkpoint proteins, a protein on a universal CAR structure, and the like. Target cells specifically include cancer cells, such as hematologic tumors, e.g., B-cell tumors, as discussed below. In some embodiments, a

bispecific antibody comprises a first binding unit that binds to BCMA and a second binding unit that binds to human La protein.

[0164] Various formats of multi-specific antibodies are within the ambit of the invention, including, without limitation, single chain polypeptides, two chain polypeptides, three chain polypeptides, four chain polypeptides, and multiples thereof. The multi-specific (e.g., bispecific) antibodies herein specifically include T-cell bispecific antibodies binding to BCMA, which is selectively expressed on plasma cells (PCs) and multiple myeloma (MM) cells, and CD3 (anti-BCMA × anti-CD3 antibodies). The multi-specific (e.g., bispecific) antibodies herein also specifically include universal CAR bispecific antibodies binding to BCMA, which is selectively expressed on plasma cells (PCs) and multiple myeloma (MM) cells, and an antigen on a universal CAR structure (anti-BCMA × anti-La protein antibodies). Such antibodies induce potent T-cell or CAR T-cell mediated killing of cells expressing BCMA, and can be used to treat tumors, in particular hematologic tumors, such as B-cell tumors, as discussed below, as well as autoimmune disorders characterized by the presence of self-reactive plasma cells, which are also described further herein.

[0165] Bispecific antibodies against CD3 and BCMA are described, for example, in WO2007117600, WO2009132058, WO2012066058, WO2012143498, WO2013072406, WO2013072415, and WO2014122144, and in US 20170051068. Universal chimeric antigen receptors comprising human La protein are described, for example, in WO2016030414, the disclosure of which application is incorporated by reference herein in its entirety.

Preparation of Antibodies

[0166] The multi-specific antibodies of the present invention can be prepared by methods known in the art. In a preferred embodiment, the antibodies herein are produced by transgenic animals, including transgenic mice and rats, preferably rats, in which the endogenous immunoglobulin genes are knocked out or disabled. In a preferred embodiment, the antibodies herein are produced in a UniRat™. UniRats™ have their endogenous immunoglobulin genes silenced and use a human immunoglobulin heavy-chain translocus to express a diverse, naturally optimized repertoire of fully human HCAs. While endogenous immunoglobulin loci in rats can be knocked out or silenced using a variety of technologies, in UniRat™ the zinc-finger (endo)nuclease (ZNF) technology was used to inactivate the endogenous rat heavy chain J-locus, light chain Cκ locus and light chain Cλ locus. ZNF constructs for microinjection into oocytes can produce IgH and IgL knock out (KO) lines. For details see, e.g., Geurts et al., 2009, Science 325:433. Characterization of Ig heavy chain knockout rats has been reported by Menoret et al., 2010, Eur. J. Immunol. 40:2932-2941. Advantages of the ZNF technology are that non-homologous end joining to silence a gene or locus via deletions up to several kb can also provide a target site for homologous integration (Cui et al., 2011, Nat Biotechnol 29:64-67). Human heavy chain

antibodies produced in UniRat™ are called UniAbs™ and can bind epitopes that cannot be attacked with conventional antibodies. Their high specificity, affinity, and small size make them ideal for mono- and poly-specific applications.

[0167] In addition to UniAbs™, specifically included herein are heavy chain-only antibodies lacking the camelid VHH framework and mutations, and their functional VH regions. Such heavy chain-only antibodies can, for example, be produced in transgenic rats or mice which comprise fully human heavy chain-only gene loci as described, e.g., in WO2006/008548, but other transgenic mammals, such as rabbit, guinea pig, rat can also be used, rats and mice being preferred. Heavy chain-only antibodies, including their VHH or VH functional fragments, can also be produced by recombinant DNA technology, by expression of the encoding nucleic acid in a suitable eukaryotic or prokaryotic host, including, for example, mammalian cells (e.g., CHO cells), E. coli or yeast.

[0168] Domains of heavy chain-only antibodies combine advantages of antibodies and small molecule drugs: can be mono- or multi-valent; have low toxicity; and are cost-effective to manufacture. Due to their small size, these domains are easy to administer, including oral or topical administration, are characterized by high stability, including gastrointestinal stability; and their half-life can be tailored to the desired use or indication. In addition, VH and VHH domains of HCAs can be manufactured in a cost effective manner.

[0169] In a particular embodiment, the heavy chain antibodies of the present invention, including UniAbs™, have the native amino acid residue at the first position of the FR4 region (amino acid position 101 according to the Kabat numbering system), substituted by another amino acid residue, which is capable of disrupting a surface-exposed hydrophobic patch comprising or associated with the native amino acid residue at that position. Such hydrophobic patches are normally buried in the interface with the antibody light chain constant region but become surface exposed in HCAs and are, at least partially, for the unwanted aggregation and light chain association of HCAs. The substituted amino acid residue preferably is charged, and more preferably is positively charged, such as lysine (Lys, K), arginine (Arg, R) or histidine (His, H), preferably arginine (R). In a preferred embodiment the heavy chain-only antibodies derived from the transgenic animals contain a Trp to Arg mutation at position 101. The resultant HCAs preferably have high antigen-binding affinity and solubility under physiological conditions in the absence of aggregation.

[0170] As part of the present invention, human anti-BCMA heavy chain antibodies with unique sequences from UniRat™ animals (UniAb™) were identified that bind human BCMA in ELISA protein and cell-binding assays. The identified heavy chain variable region (VH) sequences (see, e.g., FIG. 2) are positive for human BCMA protein binding and/or for binding to BCMA+ cells, and are all negative for binding to cells that do not express BCMA.

[0171] Heavy chain antibodies binding to non-overlapping epitopes on a BCMA protein, e.g., UniAbs™ can be identified by competition binding assays, such as enzyme-linked immunoassays (ELISA assays) or flow cytometric competitive binding assays. For example, one can use competition between known antibodies binding to the target antigen and the antibody of interest. By using this approach, one can divide a set of antibodies into those that compete with the reference antibody and those that do not. The non-competing antibodies are identified as binding to a distinct epitope that does not overlap with the epitope bound by the reference antibody. Often, one antibody is immobilized, the antigen is bound, and a second, labeled (e.g., biotinylated) antibody is tested in an ELISA assay for ability to bind the captured antigen. This can be performed also by using surface plasmon resonance (SPR) platforms, including ProteOn XPR36 (BioRad, Inc), Biacore 2000 and Biacore T200 (GE Healthcare Life Sciences), and MX96 SPR imager (Ibis technologies B.V.), as well as on biolayer interferometry platforms, such as Octet Red384 and Octet HTX (ForteBio, Pall Inc). For further details see the examples herein.

[0172] Typically, an antibody “competes” with a reference antibody if it causes about 15-100% reduction in the binding of the reference antibody to the target antigen, as determined by standard techniques, such as by the competition binding assays described above. In various embodiments, the relative inhibition is at least about 15%, at least about 20%, at least about 25%, at least about 30%, at least about 35%, at least about 40%, at least about 45%, at least about 50%, at least about 55%, at least about 60%, at least about 65%, at least about 70%, at least about 75%, at least about 80%, at least about 85%, at least about 90%, at least about 95% or higher.

Pharmaceutical Compositions

[0173] It is another aspect of the present invention to provide pharmaceutical compositions comprising one or more antibodies of the present invention in admixture with a suitable pharmaceutically acceptable carrier. Pharmaceutically acceptable carriers as used herein are exemplified, but not limited to, adjuvants, solid carriers, water, buffers, or other carriers used in the art to hold therapeutic components, or combinations thereof.

[0174] Pharmaceutical compositions of the antibodies used in accordance with the present invention are prepared for storage by mixing proteins having the desired degree of purity with optional pharmaceutically acceptable carriers, excipients or stabilizers (see, e.g., Remington's Pharmaceutical Sciences 16th edition, Osol, A. Ed. (1980)), such as in the form of lyophilized formulations or aqueous solutions. Acceptable carriers, excipients, or stabilizers are nontoxic to recipients at the dosages and concentrations employed, and include buffers such as phosphate, citrate, and other organic acids; antioxidants including ascorbic acid and methionine; preservatives (such as octadecyldimethylbenzyl ammonium chloride; hexamethonium chloride; benzalkonium chloride, benzethonium chloride; phenol,

butyl or benzyl alcohol; alkyl parabens such as methyl or propyl paraben; catechol; resorcinol; cyclohexanol; 3-pentanol; and m-cresol); low molecular weight (less than about 10 residues) polypeptides; proteins, such as serum albumin, gelatin, or immunoglobulins; hydrophilic polymers such as polyvinylpyrrolidone; amino acids such as glycine, glutamine, asparagine, histidine, arginine, or lysine; monosaccharides, disaccharides, and other carbohydrates including glucose, mannose, or dextrins; chelating agents such as EDTA; sugars such as sucrose, mannitol, trehalose or sorbitol; salt-forming counter-ions such as sodium; metal complexes (e.g., Zn-protein complexes); and/or non-ionic surfactants such as TWEEN™, PLURONICS™ or polyethylene glycol (PEG).

[0175] Pharmaceutical compositions for parenteral administration are preferably sterile and substantially isotonic and manufactured under Good Manufacturing Practice (GMP) conditions. Pharmaceutical compositions can be provided in unit dosage form (i.e., the dosage for a single administration). The formulation depends on the route of administration chosen. The antibodies herein can be administered by intravenous injection or infusion or subcutaneously. For injection administration, the antibodies herein can be formulated in aqueous solutions, preferably in physiologically-compatible buffers to reduce discomfort at the site of injection. The solution can contain carriers, excipients, or stabilizers as discussed above. Alternatively, antibodies can be in lyophilized form for constitution with a suitable vehicle, e.g., sterile pyrogen-free water, before use.

[0176] Anti-BCMA antibody formulations are disclosed, for example, in U.S. Patent No. 9,034,324. Similar formulations can be used for the proteins of the present invention. Subcutaneous antibody formulations are described, for example, in US 20160355591 and US 20160166689.

Methods of Use

[0177] The antibodies and pharmaceutical compositions herein can be used for the treatment of B-cell related disorders, including B-cell and plasma cell malignancies and autoimmune disorders characterized by the expression or overexpression of BCMA.

[0178] Such B-cell related disorders include B-cell and plasma cell malignancies and autoimmune disorders, including, without limitation, plasmacytoma, Hodgkins' lymphoma, follicular lymphomas, small non-cleaved cell lymphomas, endemic Burkitt's lymphoma, sporadic Burkitt's lymphoma, marginal zone lymphoma, extranodal mucosa-associated lymphoid tissue lymphoma, nodal monocytoid B cell lymphoma, splenic lymphoma, mantle cell lymphoma, large cell lymphoma, diffuse mixed cell lymphoma, immunoblastic lymphoma, primary mediastinal B cell lymphoma, pulmonary B cell angiocentric lymphoma, small lymphocytic lymphoma, B cell proliferations of uncertain malignant potential, lymphomatoid granulomatosis, post-transplant lymphoproliferative disorder, an immunoregulatory disorder, rheumatoid arthritis, myasthenia gravis, idiopathic thrombocytopenia purpura, anti-phospholipid syndrome, Chagas' disease, Grave's disease, Wegener's granulomatosis,

poly-arteritis nodosa, Sjogren's syndrome, pemphigus vulgaris, scleroderma, multiple sclerosis, anti-phospholipid syndrome, ANCA associated vasculitis, Goodpasture's disease, Kawasaki disease, autoimmune hemolytic anemia, and rapidly progressive glomerulonephritis, heavy-chain disease, primary or immunocyte-associated amyloidosis, or monoclonal gammopathy.

[0179] The plasma cell disorders characterized by the expression of BCMA include Multiple Myeloma (MM). MM is a B-cell malignancy characterized by a monoclonal expansion and accumulation of abnormal plasma cells in the bone marrow compartment. Current therapies for MM often cause remissions, but nearly all patients eventually relapse and die. There is substantial evidence of an immune-mediated elimination of myeloma cells in the setting of allogeneic hematopoietic stem cell transplantation; however, the toxicity of this approach is high, and few patients are cured. Although some monoclonal antibodies have shown promise for treating MM in preclinical studies and early clinical trials, consistent clinical efficacy of any monoclonal antibody therapy for MM has not been conclusively demonstrated. There is therefore a great need for new therapies, including immunotherapies, for MM (see, e.g. Carpenter et al., Clin Cancer Res 2013, 19(8):2048-2060).

[0180] Overexpression or activation of BCMA by its proliferation-inducing ligand, APRIL is known to promote human Multiple Myeloma (MM) progression in vivo. BCMA has also been shown to promote in vivo growth of xenografted MM cells harboring p53 mutation in mice. Since activity of the APRIL/BCMA pathway plays a central role in MM pathogenesis and drug resistance via bidirectional interactions between tumor cells and their supporting bone marrow microenvironment, BCMA has been identified as a target for the treatment of MM. For further details see, e.g., Yu-Tsu Tai et al., Blood 2016; 127(25):3225-3236.

[0181] Another B-cell disorder involving plasma cells expressing BCMA is systemic lupus erythematosus (SLE), also known as lupus. SLE is a systemic, autoimmune disease that can affect any part of the body and is represented with the immune system attacking the body's own cells and tissue, resulting in chronic inflammation and tissue damage. It is a Type III hypersensitivity reaction in which antibody-immune complexes precipitate and cause a further immune response (Inaki & Lee, Nat Rev Rheumatol 2010; 6: 326-337).

[0182] The anti-BCMA heavy chain-only antibodies (UniAbs) of the present invention can be used to develop therapeutic agents for the treatment of MM, SLE, and other B-cell disorders or plasma cell disorders characterized by the expression of BCMA, such as those listed above. In particular, the anti-BCMA heavy chain-only antibodies (UniAbs) of the present invention are candidates for the treatment of MM, alone or in combination with other MM treatments.

[0183] In one embodiment, the antibodies herein can be in the form of heavy chain-only anti-BCMA antibody-CAR structures, i.e., heavy chain-only anti-BCMA antibody-CAR-transduced T-cell

structures. CARs having antigen specificity for BCMA, and their methods of use, are described, for example, in WO2019/006072, the disclosure of which is incorporated by reference herein in its entirety.

[0184] In some embodiments, the antibodies herein are multi-specific (e.g., bispecific), comprising a first binding unit with binding affinity to BCMA, and a second binding unit with binding affinity for La protein, which is present on a universal chimeric antigen receptor (CAR) complex on an effector cell (e.g., a T-cell). As such, the multi-specific antibodies of the invention can be used to functionalize a universal CAR complex by binding to the universal CAR complex via a first binding unit, and providing binding affinity to BCMA via a second binding unit. The methods can further involve treating a subject in need for a disease or disorder characterized by the expression of BCMA by administering an effective amount of a cell therapy comprising a plurality of cells comprising a universal CAR complex that has been functionalized to bind to BCMA, using the multi-specific antibodies of the invention, and thereby treating the disease or disorder in the subject.

[0185] Effective doses of the compositions of the present invention for the treatment of disease vary depending upon many different factors, including means of administration, target site, physiological state of the patient, whether the patient is human or an animal, other medications administered, and whether treatment is prophylactic or therapeutic. Usually, the patient is a human, but nonhuman mammals may also be treated, e.g., companion animals such as dogs, cats, horses, etc., laboratory mammals such as rabbits, mice, rats, etc., and the like. Treatment dosages can be titrated to optimize safety and efficacy.

[0186] Dosage levels can be readily determined by the ordinarily skilled clinician, and can be modified as required, e.g., as required to modify a subject's response to therapy. The amount of active ingredient that can be combined with the carrier materials to produce a single dosage form varies depending upon the host treated and the particular mode of administration. Dosage unit forms generally contain between from about 1 mg to about 500 mg of an active ingredient.

[0187] In some embodiments, the therapeutic dosage of the agent may range from about 0.0001 to 100 mg/kg, and more usually 0.01 to 5 mg/kg, of the host body weight. For example, dosages can be 1 mg/kg body weight or 10 mg/kg body weight or within the range of 1-10 mg/kg. An exemplary treatment regimen entails administration once every two weeks or once a month or once every 3 to 6 months. Therapeutic entities of the present invention are usually administered on multiple occasions. Intervals between single dosages can be weekly, monthly or yearly. Intervals can also be irregular as indicated by measuring blood levels of the therapeutic entity in the patient. Alternatively, therapeutic entities of the present invention can be administered as a sustained release formulation, in which case less frequent administration is required. Dosage and frequency vary depending on the half-life of the polypeptide in the patient.

[0188] Typically, compositions are prepared as injectables, either as liquid solutions or suspensions; solid forms suitable for solution in, or suspension in, liquid vehicles prior to injection can also be prepared. The pharmaceutical compositions herein are suitable for intravenous or subcutaneous administration, directly or after reconstitution of solid (e.g., lyophilized) compositions. The preparation also can be emulsified or encapsulated in liposomes or micro particles such as polylactide, polyglycolide, or copolymer for enhanced adjuvant effect, as discussed above. Langer, *Science* 249: 1527, 1990 and Hanes, *Advanced Drug Delivery Reviews* 28: 97-119, 1997. The agents of this invention can be administered in the form of a depot injection or implant preparation which can be formulated in such a manner as to permit a sustained or pulsatile release of the active ingredient. The pharmaceutical compositions are generally formulated as sterile, substantially isotonic and in full compliance with all Good Manufacturing Practice (GMP) regulations of the U.S. Food and Drug Administration.

[0189] Toxicity of the antibodies and antibody structures described herein can be determined by standard pharmaceutical procedures in cell cultures or experimental animals, e.g., by determining the LD₅₀ (the dose lethal to 50% of the population) or the LD₁₀₀ (the dose lethal to 100% of the population). The dose ratio between toxic and therapeutic effect is the therapeutic index. The data obtained from these cell culture assays and animal studies can be used in formulating a dosage range that is not toxic for use in humans. The dosage of the antibodies described herein lies preferably within a range of circulating concentrations that include the effective dose with little or no toxicity. The dosage can vary within this range depending upon the dosage form employed and the route of administration utilized. The exact formulation, route of administration and dosage can be chosen by the individual physician in view of the patient's condition.

[0190] The compositions for administration will commonly comprise an antibody or other ablative agent dissolved in a pharmaceutically acceptable carrier, preferably an aqueous carrier. A variety of aqueous carriers can be used, e.g., buffered saline and the like. These solutions are sterile and generally free of undesirable matter. These compositions may be sterilized by conventional, well known sterilization techniques. The compositions may contain pharmaceutically acceptable auxiliary substances as required to approximate physiological conditions such as pH adjusting and buffering agents, toxicity adjusting agents and the like, e.g., sodium acetate, sodium chloride, potassium chloride, calcium chloride, sodium lactate and the like. The concentration of active agent in these formulations can vary widely, and will be selected primarily based on fluid volumes, viscosities, body weight and the like in accordance with the particular mode of administration selected and the patient's needs (e.g., Remington's *Pharmaceutical Science* (15th ed., 1980) and Goodman & Gillman, *The Pharmacological Basis of Therapeutics* (Hardman et al., eds., 1996)).

[0191] Also within the scope of the invention are kits comprising the active agents of the invention, and formulations thereof, and instructions for use. The kits can further contain a least one additional reagent, e.g., a chemotherapeutic drug, etc. Kits typically include a label indicating the intended use of the contents of the kit. The term label includes any writing, or recorded material supplied on or with the kit, or which otherwise accompanies the kit.

[0192] The invention now being fully described, it will be apparent to one of ordinary skill in the art that various changes and modifications can be made without departing from the spirit or scope of the invention.

EXAMPLES

[0193] The following examples are provided to illustrate certain embodiments and are not to be construed as limiting the scope of this disclosure in any way.

Example 1: CAR-T mediated T-cell activation by human tumor cell lines

[0194] CAR-T cell activity was measured by transfecting Jurkat T lymphocyte cells with an anti-BCMA CAR and a 6x NFAT TK nano luciferase reporter. Transfected Jurkat cells were co-cultured for 24 hours with BCMA-positive NCI-H929, U266, and Daudi, or BCMA-negative K562 cells. Luciferase activity was measured using the Promega Nano-Glo Luciferase Assay System (catalog # N1110) and data was normalized to co-culture containing the CAR transfected Jurkat and BCMA-negative K562 cell lines. Statistical significance was determined using an unpaired, two-tailed t-test.

[0195] The results are provided in FIG. 8, Panel B.

[0196] FIG. 8, Panel A, is a schematic illustration of a CAR-T structure comprising an anti-BCMA extracellular binding domain comprising the VH sequence of SEQ ID NO: 12 (Clone ID No: 316302). FIG. 8, Panel B is a graph depicting T cell activity of Jurkat cells transfected with an NFAT luciferase reporter of T-cell signaling and an anti-BCMA 316302 CAR with NCI-H929 (*p= 0.04), U266 (**p=0.0008), and Daudi (*p=0.03) cells. These results demonstrate that T-cell activation was specific to BCMA target binding, as co-culture of BCMA CAR Jurkat cells with the BCMA-negative K562 cell line did not result in appreciable luciferase reporter signal. Moreover, incubation of Jurkat cells, transfected with reporter but not the CAR, with BCMA positive NCI-H929, U266, and Daudi cells also did not result in appreciable luciferase reporter signal.

Example 2: Antibody binding to BCMA expressing cells

[0197] Antibody binding to BCMA-expressing MM1.S and H929 cells was assessed by flow cytometry. Test antibodies included bispecific constructs that bind to La protein (designated herein as “AntiX”) and BCMA (designated herein as “BCMA_F7E”). As described elsewhere herein, bispecific constructs in accordance with embodiments of the invention can include a BCMA binding domain in a bivalent format, as depicted in FIG. 7, Panel A, right side; also referred to herein as “BCMA_F7E_F7E”; or a monovalent format, as depicted in FIG. 7, Panel B, right side; also referred to herein as “BCMA_F7E”.

[0198] Binding experiments were conducted with the following constructs: AntiX**BCMA_F7E, a bispecific antibody with a monovalent arm targeting BCMA (comprising SEQ ID NO: 12) and an La protein binding arm; AntiX**BCMA_F7E_F7E, a bispecific antibody with a bivalent arm targeting BCMA (comprising SEQ ID NO: 12 in a bivalent configuration) and an La protein binding arm; AntiX**GP120_F8A, a bispecific negative control antibody comprising an La protein binding arm and an arm that binds to GP120. Briefly, 300,000-500,000 cells were co-incubated with test antibody as listed in Table 1 and with commercial anti-BCMA antibody (Biolegend, 19F2) at 0.5 µl/test in 150 µl of FACS buffer (1X PBS, 2% FBS, 1 mM EDTA). Cells were subsequently washed twice and incubated with anti-IgG (PE) secondary antibody. After two additional washes, cells were processed on a Cytotflex instrument (Beckman Coulter) and analyzed using the FlowJo software package. Table 1, FIG. 9, Panel A, and FIG. 9, Panel B show mean fluorescence intensity (MFI) for anti-IgG4 secondary antibody in MM1.S and H929 cells.

Table 1. Mean Fluorescence intensity (MFI) of anti-IgG4 secondary antibody.

Cell Type	Test Antibody (ng/ml)	AntiX**	AntiX**	AntiX**
		BCMA_F7E	BCMA_F7E_F7E	GP120_F8A
H929	200.000	14,064	52,932	5,616
	100.000	7,562	20,874	5,403
	50.000	6,070	10,927	5,239
	25.000	5,479	6,614	5,067
	12.500	5,186	5,682	4,988
	6.250	5,241	5,220	4,983
	3.130	5,168	5,141	5,018
	1.560	5,096	5,057	4,946
	0.781	5,056	5,023	4,874
	0.391	5,027	5,036	4,918
	0.195	5,031	5,008	4,917
	0.098	5,061	5,067	4,897
	0.000	4,997		
MM1.S	200.000	3,183	7,339	1,994
	100.000	2,444	4,135	2,013
	50.000	2,176	2,928	2,010
	25.000	2,129	2,345	2,007
	12.500	2,120	2,176	2,003
	6.250	2,134	2,108	1,957
	3.130	2,107	2,073	1,998
	1.560	2,109	2,027	1,992
	0.781	2,064	2,010	1,997
	0.391	2,073	2,032	2,018
	0.195	2,072	2,009	1,976
	0.098	2,036	1,965	2,016
	0.000	2,991		

Example 3: Competitive ELISA with APRIL and test antibodies

- [0199] The avidity of bispecific BCMA-binding was tested in a competitive ELISA with BCMA's natural agonist, APRIL, also known as BAFF. In this assay, BCMA bound to the surface of plates was co-incubated with test antibody and APRIL, each at various concentrations. Clear Flat-Bottom Immuno Nonsterile 96-well plates (Corning, catalog no. 3455) were incubated overnight with 10 ng per well recombinant BCMA (R&D Systems, catalog no. 193-BC) in 100 μ L PBS. Plates were washed and incubated with 1X Blocker BSA (Thermo Scientific, catalog no. 37525).
- [0200] Test antibodies and recombinant APRIL (R&D Systems, catalog no. 560-AP) were incubated with BCMA-coated plates at the concentrations described in Table 2. Plates were washed and incubated with mouse anti-human IgG-HRP antibodies (Southern Biotech, catalog no. 9200-05). Plates were washed and developed with the Pierce TMB Substrate kit (Thermo Fisher, catalog no. 34021) according to the manufacturer's instructions. Absorbance was measured at 450 nm and 570 nm within 15 minutes. Table 2 shows A450 - A570 values for each sample. FIG. 10, Panels A-C are graphs showing A450-A570 values for bispecific antibody constructs AntiX**BCMA__F7E (pane A), AntiX**BCMA_F7E_F7E (Panel B), and AntiX**GP120_F8A (Panel C), respectively. More antibody binding is indicated by higher A450-A570 values. Even at higher APRIL concentration compared to bispecific antibody construct concentration, the AntiX**BCMA_F7E_F7E construct, which comprises a BCMA-binding VH sequence in a bivalent configuration, demonstrates significant binding. The AntiX**BCMA_F7E_F7E construct demonstrated stronger binding than the AntiX**BCMA_F7E bispecific antibody construct, which comprises a BCMA-binding VH sequence in a monovalent configuration.

Table 2. (A450-A570) values indicating antibody binding to BCMA in the presence of BCMA agonist, APRIL.

Sample	Antibody (ng/mL)	APRIL (ng/mL)							
		500.00	250.00	125.00	62.50	31.25	15.63	7.81	0.00
AntiX** BCMA _F7E	200.000	0.4818	0.9693	1.3717	2.0748	2.2860	2.4012	2.5108	2.4362
	20.000	0.1117	0.1479	0.2030	0.3589	0.4181	0.4825	0.4951	0.4656
	2.000	0.0711	0.0837	0.0736	0.0834	0.0875	0.0867	0.0895	0.0894
	0.200	0.0745	0.0615	0.0643	0.0631	0.0620	0.0661	0.0643	0.0708
	0.020	0.0835	0.0617	0.0640	0.0629	0.0622	0.0614	0.0631	0.0630
	0.002	0.0836	0.0647	0.0660	0.0670	0.0580	0.0607	0.0607	0.0616
AntiX** BCMA _F7E _F7E	200.000	1.4064	2.2066	2.2959	2.4405	2.4211	2.3723	2.4742	2.4734
	20.000	0.2533	0.4130	0.6371	0.5369	0.8281	0.7776	0.7889	0.6478
	2.000	0.0732	0.0921	0.1081	0.0980	0.1211	0.1066	0.1078	0.1071
	0.200	0.0603	0.0614	0.0651	0.0595	0.0601	0.0587	0.0577	0.0676
	0.020	0.0631	0.0618	0.0557	0.0577	0.0551	0.0584	0.0578	0.0570
	0.002	0.0617	0.0607	0.0607	0.0569	0.0528	0.0620	0.0602	0.0578
AntiX** GP120 _F8A	200.000	0.0838	0.0682	0.0779	0.0701	0.0700	0.0774	0.0734	0.0755
	20.000	0.0615	0.0603	0.0597	0.0586	0.0623	0.0630	0.0659	0.0687
	2.000	0.0615	0.0591	0.0575	0.0628	0.0575	0.0580	0.0644	0.0656
	0.200	0.0590	0.0615	0.0614	0.0596	0.0589	0.0623	0.0597	0.0679
	0.020	0.0563	0.0600	0.0582	0.0611	0.0591	0.0615	0.0639	0.0639
	0.002	0.0617	0.0644	0.0595	0.0634	0.0600	0.0757	0.0685	0.0669

CLAIMS:

1. A multi-specific antibody that binds to BCMA, comprising a first binding unit comprising a variable region comprising a CDR3 sequence having at least 85% sequence identity to SEQ ID NO: 3.
2. A multi-specific antibody that binds to BCMA, comprising a first binding unit comprising a variable region comprising a CDR1 sequence, a CDR2 sequence, and a CDR3 sequence, wherein the CDR1, CDR2 and CDR3 sequences combined have at least 85% sequence identity to SEQ ID NOs: 1-3 combined.
3. A multi-specific antibody that binds to BCMA, comprising a first binding unit comprising a variable region comprising:
 - a CDR1 sequence comprising the sequence of SEQ ID NO: 1;
 - a CDR2 sequence comprising the sequence of SEQ ID NO: 2; and
 - a CDR3 sequence comprising the sequence of SEQ ID NO: 3.
4. The multi-specific antibody of any one of claims 1-3, wherein the CDR1, CDR2 and CDR3 sequences are present in a human VH framework.
5. The multi-specific antibody of any one of claims 1-4, wherein the variable region is a heavy chain-only variable region.
6. The multi-specific antibody of any one of claims 1-5, wherein the variable region comprises a sequence having at least 95% sequence identity to SEQ ID NO: 12.
7. The multi-specific antibody of claim 6, wherein the variable region comprises a sequence comprising SEQ ID NO: 12.
8. The multi-specific antibody of any one of claims 1-7, wherein the variable region is in a monovalent or bivalent configuration.
9. The multi-specific antibody of any one of claims 1-8, further comprising a heavy chain constant region sequence, in the absence of a CH1 sequence.
10. The multi-specific antibody of claim 9, wherein the heavy chain constant region sequence comprises a CH2 domain and a CH3 domain, but no CH1 domain.

11. The multi-specific antibody of claim 10, wherein the CH2 domain comprises the sequence of a wild type human IgG4 CH2 domain (SEQ ID NO: 36).
12. The multi-specific antibody of claim 10, wherein the CH2 domain comprises a variant human IgG4 CH2 domain comprising an F234A mutation, an L235A mutation, or both an F234A mutation and an L235A mutation.
13. The multi-specific antibody of claim 10, wherein the CH3 domain comprises the sequence of a wild type human IgG4 CH3 domain (SEQ ID NO: 38).
14. The multi-specific antibody of claim 10, wherein the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366W mutation (SEQ ID NO: 39).
15. The multi-specific antibody of claim 10, wherein the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation (SEQ ID NO: 40).
16. The multi-specific antibody of claim 10, further comprising a hinge region sequence positioned between the heavy chain-only variable region and the CH2 domain.
17. The multi-specific antibody of claim 16, wherein the hinge region sequence comprises the sequence of a wild type human IgG4 hinge region (SEQ ID NO: 32).
18. The multi-specific antibody of claim 16, wherein the hinge region sequence comprises a variant human IgG4 hinge region sequence comprising an S228P mutation (SEQ ID NO: 33).
19. The multi-specific antibody of claim 18, further comprising a variant human IgG4 CH2 domain comprising an F234A mutation and an L235A mutation.
20. The multi-specific antibody of claim 19, further comprising a variant human IgG4 CH3 domain comprising a T366W mutation.
21. The multi-specific antibody of claim 19, further comprising a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation.

22. The multi-specific antibody of any one of claims 1-21, further comprising a second binding unit that binds to La protein.
23. The multi-specific antibody of claim 22, wherein the second binding unit comprises:
- (a) a heavy chain variable region comprising:
 - (i) a CDR1 sequence comprising any one of the sequences of SEQ ID NOs: 4 or 7;
 - and
 - (ii) a CDR2 sequence comprising the sequence of SEQ ID NO: 5; and
 - (iii) a CDR3 sequence comprising any one of the sequences of SEQ ID NOs: 6 or 8;
 - and
 - (b) a light chain variable region comprising:
 - (i) a CDR1 sequence comprising the sequence of SEQ ID NO: 9; and
 - (ii) a CDR2 sequence comprising the sequence of SEQ ID NO: 10; and
 - (iii) a CDR3 sequence comprising the sequence of SEQ ID NO: 11.
24. The multi-specific antibody of claim 23, wherein the heavy chain variable region of the second binding unit comprises:
- (i) a CDR1 sequence comprising the sequence of SEQ ID NO: 4, a CDR2 sequence comprising the sequence of SEQ ID NO: 5, and a CDR3 sequence comprising the sequence of SEQ ID NO: 6; or
 - (i) a CDR1 sequence comprising the sequence of SEQ ID NO: 7, a CDR2 sequence comprising the sequence of SEQ ID NO: 5, and a CDR3 sequence comprising the sequence of SEQ ID NO: 8.
25. The multi-specific antibody of any one of claims 23-24, wherein the CDR1, CDR2 and CDR3 sequences of the heavy chain variable region of the second binding unit are present in a human VH framework.
26. The multi-specific antibody of any one of claims 23-25, wherein the CDR1, CDR2 and CDR3 sequences of the light chain variable region of the second binding unit are present in a human VL framework.
27. The multi-specific antibody of claim 26, wherein the human VL framework is a human Vkappa framework.

28. The multi-specific antibody of claim 26, wherein the human VL framework is a human Vlambda framework.
29. The multi-specific antibody of any one of claims 22-28, wherein the second binding unit comprises a heavy chain variable region comprising a sequence having at least 95% sequence identity to SEQ ID NO: 14 and a light chain variable region comprising a sequence having at least 95% sequence identity to SEQ ID NO: 16.
30. The multi-specific antibody of claim 29, wherein the second binding unit comprises a heavy chain variable region comprising the sequence of SEQ ID NO: 14 and a light chain variable region comprising the sequence of SEQ ID NO: 16.
31. The multi-specific antibody of any one of claims 22-28, wherein the second binding unit comprises a heavy chain variable region comprising a sequence having at least 95% sequence identity to SEQ ID NO: 15 and a light chain variable region comprising a sequence having at least 95% sequence identity to SEQ ID NO: 17.
32. The multi-specific antibody of claim 31, wherein the second binding unit comprises a heavy chain variable region comprising the sequence of SEQ ID NO: 15 and a light chain variable region comprising the sequence of SEQ ID NO: 17.
33. The multi-specific antibody of any one of claims 23-32, wherein the heavy chain variable region and the light chain variable region of the second binding unit are on a common polypeptide subunit of the multi-specific antibody, and are connected by a linker sequence.
34. The multi-specific antibody of any one of claims 23-32, wherein the heavy chain variable region and the light chain variable region of the second binding unit are on different polypeptide subunits of the multi-specific antibody.
35. The multi-specific antibody of any one of claims 22-34, wherein the second binding unit further comprises a heavy chain constant region.
36. The multi-specific antibody of claim 35, wherein the heavy chain constant region comprises a CH1 domain, a hinge region, a CH2 domain, and a CH3 domain.

37. The multi-specific antibody of claim 36, wherein the CH2 domain comprises the sequence of a wild type human IgG4 CH2 domain (SEQ ID NO: 36).
38. The multi-specific antibody of claim 36, wherein the CH2 domain comprises a variant human IgG4 CH2 domain comprising an F234A mutation, an L235A mutation, or both an F234A mutation and an L235A mutation.
39. The multi-specific antibody of claim 36, wherein the CH3 domain comprises the sequence of a wild type human IgG4 CH3 domain (SEQ ID NO: 38).
40. The multi-specific antibody of claim 36, wherein the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366W mutation (SEQ ID NO: 39).
41. The multi-specific antibody of claim 36, wherein the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation (SEQ ID NO: 40).
42. The multi-specific antibody of claim 36, wherein the hinge region comprises the sequence of a wild type human IgG4 hinge region (SEQ ID NO: 32).
43. The multi-specific antibody of claim 36, wherein the hinge region comprises a variant human IgG4 hinge region sequence comprising an S228P mutation (SEQ ID NO: 33).
44. The multi-specific antibody of claim 43, further comprising a variant human IgG4 CH2 domain comprising an F234A mutation and an L235A mutation.
45. The multi-specific antibody of claim 44, further comprising a variant human IgG4 CH3 domain comprising a T366W mutation.
46. The multi-specific antibody of claim 44, further comprising a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation.
47. The multi-specific antibody of any one of claims 22-46, wherein the second binding unit further comprises a light chain constant region.

48. The multi-specific antibody of claim 47, wherein the light chain constant region comprises a human V κ appa constant region sequence.
49. The multi-specific antibody of claim 47, wherein the light chain constant region comprises a human V λ mbda constant region sequence.
50. The multi-specific antibody of any one of claims 1-49, which is bispecific.
51. A multi-specific antibody comprising:
- (a) a first binding unit that binds to La protein, comprising:
 - (i) a heavy chain variable region comprising: a CDR1 sequence of SEQ ID NOs: 4 or 7, a CDR2 sequence of SEQ ID NO: 5, and a CDR3 sequence of SEQ ID NO: 6 or 8, in a human VH framework; and
 - (ii) a light chain variable region comprising: a CDR1 sequence of SEQ ID NO: 9, a CDR2 sequence of SEQ ID NO: 10, and a CDR3 sequence of SEQ ID NO: 11, in a human VL framework; and
 - (b) a second binding unit that binds to BCMA, comprising:
 - (i) an antigen-binding domain of an anti-BCMA heavy chain-only antibody, comprising a CDR1 sequence of SEQ ID NO: 1, a CDR2 sequence of SEQ ID NO: 2, and a CDR3 sequence of SEQ ID NO: 3, in a human VH framework, wherein the antigen-binding domain of the anti-BCMA heavy chain-only antibody is in a monovalent or bivalent configuration.
52. The multi-specific antibody of claim 51, wherein the heavy chain variable region and the light chain variable region of the first binding unit are on a common polypeptide subunit of the multi-specific antibody, and are connected by a linker sequence.
53. The multi-specific antibody of claim 51, wherein the heavy chain variable region and the light chain variable region of the first binding unit are on different polypeptide subunits of the multi-specific antibody.

54. The multi-specific antibody of any one of claims 51-53, wherein the heavy chain variable region of the first binding unit comprises:
- (i) a CDR1 sequence of SEQ ID NO: 4, a CDR2 sequence of SEQ ID NO: 5, and a CDR3 sequence of SEQ ID NO: 6; or
 - (ii) a CDR1 sequence of SEQ ID NO: 7, a CDR2 sequence of SEQ ID NO: 5, and a CDR3 sequence of SEQ ID NO: 8.
55. The multi-specific antibody of any one of claims 51-54, wherein the human VL framework is a human V κ framework.
56. The multi-specific antibody of any one of claims 51-54, wherein the human VL framework is a human V λ framework.
57. The multi-specific antibody of any one of claims 51-56, wherein the heavy chain variable region of the first binding unit comprises a sequence having at least 95% identity to SEQ ID NO: 14 or SEQ ID NO: 15.
58. The multi-specific antibody of claim 57, wherein the heavy chain variable region of the first binding unit comprises the sequence of SEQ ID NO: 14 or SEQ ID NO: 15.
59. The multi-specific antibody of any one of claims 51-58, wherein the light chain variable region of the first binding unit comprises a sequence having at least 95% identity to SEQ ID NO: 16 or SEQ ID NO: 17.
60. The multi-specific antibody of claim 59, wherein the light chain variable region of the first binding unit comprises the sequence of SEQ ID NO: 16 or SEQ ID NO: 17.
61. The multi-specific antibody of any one of claims 51-60, wherein the antigen-binding domain of the anti-BCMA heavy chain-only antibody comprises a variable region sequence having at least 95% identity to the sequence of SEQ ID NO: 12.
62. The multi-specific antibody of claim 61, wherein the antigen-binding domain of the anti-BCMA heavy chain-only antibody comprises a variable region sequence comprising the sequence of SEQ ID NO: 12.

63. The multi-specific antibody of any one of claims 51-62, wherein the antigen-binding domain of the anti-BCMA heavy chain-only antibody is in a bivalent configuration, and comprises a linker sequence.
64. The multi-specific antibody of claim 63, wherein the linker sequence comprises a G4S linker sequence.
65. The multi-specific antibody of any one of claims 51-64, wherein the first binding unit further comprises a heavy chain constant region.
66. The multi-specific antibody of claim 65, wherein the heavy chain constant region comprises a CH1 domain, a hinge region, a CH2 domain, and a CH3 domain.
67. The multi-specific antibody of claim 66, wherein the CH2 domain comprises the sequence of a wild type human IgG4 CH2 domain (SEQ ID NO: 36).
68. The multi-specific antibody of claim 66, wherein the CH2 domain comprises a variant human IgG4 CH2 domain comprising an F234A mutation, an L235A mutation, or both an F234A mutation and an L235A mutation.
69. The multi-specific antibody of claim 66, wherein the CH3 domain comprises the sequence of a wild type human IgG4 CH3 domain (SEQ ID NO: 38).
70. The multi-specific antibody of claim 66, wherein the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366W mutation (SEQ ID NO: 39).
71. The multi-specific antibody of claim 66, wherein the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation (SEQ ID NO: 40).
72. The multi-specific antibody of claim 66, wherein the hinge region comprises the sequence of a wild type human IgG4 hinge region (SEQ ID NO: 32).
73. The multi-specific antibody of claim 66, wherein the hinge region comprises a variant human IgG4 hinge region sequence comprising an S228P mutation (SEQ ID NO: 33).

74. The multi-specific antibody of claim 73, further comprising a variant human IgG4 CH2 domain comprising an F234A mutation and an L235A mutation.
75. The multi-specific antibody of claim 74, further comprising a variant human IgG4 CH3 domain comprising a T366W mutation.
76. The multi-specific antibody of claim 74, further comprising a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation.
77. The multi-specific antibody of any one of claims 51-76, wherein the first binding unit further comprises a light chain constant region.
78. The multi-specific antibody of claim 77, wherein the light chain constant region comprises a human Vkappa constant region sequence.
79. The multi-specific antibody of claim 77, wherein the light chain constant region comprises a human Vlambd constant region sequence.
80. The multi-specific antibody of any one of claims 51-79, wherein the second binding unit further comprises a heavy chain constant region sequence, in the absence of a CH1 sequence.
81. The multi-specific antibody of claim 80, wherein the heavy chain constant region sequence comprises a CH2 domain and a CH3 domain, but no CH1 domain.
82. The multi-specific antibody of claim 81, wherein the CH2 domain comprises the sequence of a wild type human IgG4 CH2 domain (SEQ ID NO: 36).
83. The multi-specific antibody of claim 81, wherein the CH2 domain comprises a variant human IgG4 CH2 domain comprising an F234A mutation, an L235A mutation, or both an F234A mutation and an L235A mutation.
84. The multi-specific antibody of claim 81, wherein the CH3 domain comprises the sequence of a wild type human IgG4 CH3 domain (SEQ ID NO: 38).
85. The multi-specific antibody of claim 81, wherein the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366W mutation (SEQ ID NO: 39).

86. The multi-specific antibody of claim 81, wherein the CH3 domain comprises a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation (SEQ ID NO: 40).
87. The multi-specific antibody of claim 81, wherein the hinge region comprises the sequence of a wild type human IgG4 hinge region (SEQ ID NO: 32).
88. The multi-specific antibody of claim 81, wherein the hinge region comprises a variant human IgG4 hinge region sequence comprising an S228P mutation (SEQ ID NO: 33).
89. The multi-specific antibody of claim 88, further comprising a variant human IgG4 CH2 domain comprising an F234A mutation and an L235A mutation.
90. The multi-specific antibody of claim 89, further comprising a variant human IgG4 CH3 domain comprising a T366W mutation.
91. The multi-specific antibody of claim 89, further comprising a variant human IgG4 CH3 domain comprising a T366S mutation, an L368A mutation, and a Y407V mutation.
92. The multi-specific antibody of any one of claims 51-91, which is bispecific.
93. A bispecific three-chain antibody-like molecule that binds to BCMA and La protein, comprising:
(a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 18;
(b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 24; and
(c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 20.
94. A bispecific three-chain antibody-like molecule that binds to BCMA and La protein, comprising:
(a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 18;
(b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 26; and
(c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 20.

95. A bispecific three-chain antibody-like molecule that binds to BCMA and La protein, comprising:
- (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 21;
 - (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 24; and
 - (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 23.
96. A bispecific three-chain antibody-like molecule that binds to BCMA and La protein, comprising:
- (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 21;
 - (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 26; and
 - (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 23.
97. A bispecific three-chain antibody-like molecule that binds to BCMA and La protein, comprising:
- (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 19;
 - (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 25; and
 - (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 20.
98. A bispecific three-chain antibody-like molecule that binds to BCMA and La protein, comprising:
- (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 19;
 - (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 27; and
 - (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 20.
99. A bispecific three-chain antibody-like molecule that binds to BCMA and La protein, comprising:
- (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 22;
 - (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 25; and
 - (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 23.

100. A bispecific three-chain antibody-like molecule that binds to BCMA and La protein, comprising:
- (a) a first heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 22;
 - (b) a second heavy chain polypeptide subunit comprising the sequence of SEQ ID NO: 27; and
 - (c) a first light chain polypeptide subunit comprising the sequence of SEQ ID NO: 23.
101. A pharmaceutical composition comprising an antibody of any one of claims 1 to 100.
102. A method for the treatment of a B-cell disorder characterized by expression of BCMA, comprising administering to a subject with said disorder an antibody of any one of claims 1 to 100, or the pharmaceutical composition of claim 101.
103. Use of an antibody of any one of claims 1 to 100, in the preparation of a medicament for the treatment of a B-cell disorder characterized by expression of BCMA.
104. An antibody of any one of claims 1 to 100 for use in the treatment of a B-cell disorder characterized by expression of BCMA.
105. The method, use or antibody of any one of claims 102-104, wherein the disorder is multiple myeloma (MM).
106. The method, use or antibody of any one of claims 102-104, wherein the disorder is an autoimmune disorder.
107. The method, use or antibody of claim 106, wherein the autoimmune disorder is systemic lupus erythematosus (SLE).
108. The method, use or antibody of claim 106, wherein the autoimmune disorder is rheumatoid arthritis (RA).
109. The method, use or antibody of claim 106, wherein the autoimmune disorder is multiple sclerosis (MS).

110. A polynucleotide encoding an antibody of any of claims 1 to 100.
111. A vector comprising the polynucleotide of claim 110.
112. A cell comprising the vector of claim 111.
113. A method of producing an antibody of any one of claims 1 to 100, comprising growing a cell according to claim 112 under conditions permissive for expression of the antibody, and isolating the antibody from the cell.
114. A method of making an antibody of any one of claims 1 to 100, comprising immunizing a UniRat animal with BCMA and identifying BCMA-binding heavy chain sequences.
115. A method of treatment, comprising administering to an individual in need an effective dose of the antibody of any one of claims 1 to 100, or the pharmaceutical composition of claim 101.