HER-2 TARGETED BISPECIFIC COMPOSITIONS AND METHODS FOR MAKING AND USING THE SAME

SEQUENCE LISTING STATEMENT:

[0001] A computer readable form of the Sequence Listing is filed with this application by electronic submission and is incorporated into this application by reference in its entirety. The Sequence Listing is contained in the file created on June 14, 2021 having the file name "789-601_20-1832-WO_ST25_FINAL.txt" and is 1502 kb in size.

REFERENCE STATEMENT

[0002] This application claims priority to U.S. Provisional Patent Application No. 63/044,301 filed on June 25, 2020, entitled "BARCODED BISPECIFIC COMPOSITIONS AND METHODS FOR MAKING AND USING THE SAME"; U.S. Provisional Patent Application No. 63/077,503 filed on September 11, 2020, entitled "BARCODED BISPECIFIC COMPOSITIONS AND METHODS FOR MAKING AND USING THE SAME"; U.S. Provisional Patent Application No. 63/108,783 filed on November 2, 2020, entitled "BARCODED BISPECIFIC COMPOSITIONS AND METHODS FOR MAKING AND USING THE SAME"; U.S. Provisional Patent Application No. 63/166,857 filed on March 26, 2021, entitled "HER2 TARGETED BISPECIFIC COMPOSITIONS AND METHODS FOR MAKING AND USING THE SAME"; and U.S. Provisional Patent Application No. 63/196,408 filed on June 3, 2021, entitled "HER2 TARGETED BISPECIFIC COMPOSITIONS AND METHODS FOR MAKING AND USING THE SAME" all of which are incorporated herein in their entireties.

BACKGROUND

[0003] Bispecific T Cell Engagers (TCEs) present a highly potent modality for cancer therapy that redirects T-cell cytotoxicity against tumors that express a selected tumor-associated antigen, bypassing the requirements for T-cell recognition of tumor antigens. The activity of a TCE depends on its ability to activate T cells through effective stimulation of the T cell receptor (TCR). Their extreme potency derives from the minimal requirement for as few as 3 TCRs to become stimulated and coalesce to form an immune synapse between the T cell and target cell to initiate cytotoxicity. In addition to their induction of cytotoxicity, their potency also involves cytokine-driven actions downstream of T-cell activation that enhance and amplify the anti-tumor immune response. Thus, TCEs offer the promise of immunotherapy to patients whose tumors harbor insufficient

mutations or have escaped immune surveillance by other means. However, this modality is not without its own challenges and the use of TCEs in solid tumors has been limited by their extreme potency and on target, off-tumor toxicities in healthy tissue.

[0004] TCEs have been highly effective at inducing remission in patients with hematologic cancers, but their use in solid tumors has been limited by their extreme potency and on-target toxicities against normal tissue expressing even low levels of target. Impressive, but rare clinical responses have been observed against tumors typically refractory to immunotherapy (e.g., microsatellite-stable colorectal and prostate cancers), but toxicities such as cytokine release syndrome at low doses have prevented dose escalation to reveal the modality's clinical potential. Grade 4 cytokine release syndrome induced in patients treated with the Ichnos ISB 1302 HER2 TCE at < 1 ug/kg doses highlights the challenge faced by TCEs even when directed against relatively tissue-restricted targets.

[0005] Clinical trials with blinatumomab, (an approved CD3xCD19 bispecific antibody) revealed that cytokine release syndrome (CRS) is one of the major safety-related adverse events. CRS and on-target toxicities at low drug doses have significantly compromised the therapeutic index and potential of the TCE modality in the clinic against solid tumors. For example, the clinical trial for catumaxomab (CD3xEpCAM) was terminated due to drug-related hepatic failure at a 10μg dose. In another trial, a HER2-targeted TCE (Glenmark GBR1302) the dose of the agent was limited to less than 1μg/kg due to onset of G4 CRS. While pasotuxumab (a PSMA-targeted TCE) showed a good response, it was hampered by CRS at doses greater than 40μg/day. The literature is replete with other examples of the CRS and on-target toxicity challenges presented by TCEs.

[0006] Attempts to circumvent CRS include complex molecular designs, but these have been unsuccessful due to toxicity and/or enhanced immunogenicity. This presents a significant unmet need for new strategies that can overcome therapeutic index challenges in solid tumors. If the potency of TCEs could be harnessed and the CRS and on-target toxicity challenges could be controlled, it may be possible to generate powerful therapeutics that could potentially be used against a broad spectrum of cancers.

[0007] A therapeutic, such as a drug substance includes polypeptides that may be produced in a manner that results in a mixture of polypeptides that can influence activity of the drug substance. The mixture of polypeptides can often include the full-length polypeptide, along with size variants (e.g., truncations) thereof. The presence of variants that differ in size from the desired full-length product may affect the biological behavior of a drug substance, potentially affecting the safety and/or efficacy of the polypeptide drug substance. For example, protein-based prodrugs for cancer therapy may be engineered with a tumor-targeted activation mechanism. More specifically, the full-length therapeutic protein may be an inactive (non-cytotoxic) prodrug form, while truncation variants of the full-length construct may lose protective sequences and become cytotoxic (active), thus "contaminating" the prodrug composition. In some instances, such shorter length variants may

pose a greater risk of immunogenicity, have less selective toxicity for tumor cells, or show a less desired pharmacokinetic profile (*e.g.*, resulting in a narrowed therapeutic window) compared with the full-length protein. As a result, detection and quantification of protein structural variations can be of importance in assessing biological properties (*e.g.*, clinical safety and pharmacologic efficacy) of biotherapeutics and in developing new biotherapeutics (*e.g.*, with increased efficacy and reduced side effects). Existing techniques and methods for identifying and quantifying the amount of "contaminating" truncation products may include one or more drawbacks, such as being of limited sensitivity, ease, efficiency, or effectiveness.

SUMMARY

[0008] The present invention addresses a long-felt unmet need in providing TCE cancer therapeutics that have an increased therapeutic index. In doing so, the invention harnesses the therapeutic potential of TCEs by providing XTENylated protease activated bispecific T cell engagers (XPATs). XPATs represent a novel strategy to improve the toxicity profile of T cell engagers while maintaining their potency against solid tumors, thus enabling a significant increase in the therapeutic index and expansion of target landscape for this potent modality. In certain specific embodiments, the XPATs of the present invention target HER2-bearing tumors. More specifically, AMX-818 is a HER2-targeted, conditionally activated prodrug TCE designed to exploit the dysregulated protease activity in tumors, while sparing healthy tissues where protease inhibition prevails, thus broadening the safety margin and therapeutic index

[0009] Provided herein is a polypeptide having an N-terminal amino acid and a C-terminal amino acid, the polypeptide comprising: (a) an extended recombinant polypeptide (**XTEN**), comprising a barcode fragment (**BAR**) releasable from said polypeptide upon digestion by a protease; (b) a bispecific antibody construct (**BsAb**), comprising a first antigen binding fragment (**AF1**) that specifically binds to a cluster of differentiation 3 T cell receptor (CD3), which AF1 comprises light chain complementarity-determining regions 1 (CDR-L1), 2 (CDR-L2), and 3 (CDR-L3) and heavy chain complementarity-determining regions 1 (CDR-H1), 2 (CDR-H2), and 3 (CDR-H3), wherein said CDR-H3 comprises an amino acid sequence of **SEQ ID NO:10**; and a second antigen binding fragment (**AF2**) that specifically binds to human epidermal growth factor receptor 2 (HER2); and (c) a release segment (**RS**) positioned between said XTEN and said bispecific antibody construct, wherein said XTEN is characterized in that: (i) it comprises at least 100, or at least 150 amino acids; (ii) at least 90% of its amino acid residues are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P); (iii) it comprises at least 4 different types of amino acids that are G, A, S, T, E, or P; and (iv) said XTEN is formed from a plurality of non-overlapping sequence motifs that are each from 9 to 14 amino acids in length, wherein said plurality of non-overlapping sequence motifs comprise: (1) a set of non-overlapping sequence motifs, wherein each non-overlapping sequence motif of said set of non-overlapping sequence motifs is repeated at least two times in said XTEN; and (2) a non-overlapping sequence motif that occurs only once

within said XTEN; wherein said barcode fragment (**BAR**) includes at least part of said non-overlapping sequence motif that occurs only once within said XTEN; wherein said barcode fragment (**BAR**) differs in sequence and molecular weight from all other peptides fragments that are releasable from said polypeptide upon complete digestion of said polypeptide by said protease; and wherein said barcode fragment (**BAR**) does not include said N-terminal amino acid or said C-terminal amino acid of said polypeptide.

[0010] In certain embodiments, said set of non-overlapping sequence motifs each independently comprise an amino acid sequence identified herein by **SEQ ID NOS: 179-200 and 1715-1722**. In certain embodiments, said set of non-overlapping sequence motifs each independently comprise an amino acid sequence identified herein by **SEQ ID NOS: 186-189**. In certain embodiments, said set of non-overlapping sequence motifs comprise at least two, at least three, or all four of the sequence motifs **SEQ ID NOS: 186-189**. In certain embodiments, said XTEN comprises a length of from 100 to 3,000, from 150 to 3,000, from 100 to 1,000, or from 150 to 1,000 amino acid residues. In certain embodiments, said XTEN comprises a length of at least 200, at least 250, at least 300, at least 350, at least 400, at least 450, or at least 500 amino acid residues. In certain embodiments, at least 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% of the amino acid residues of said XTEN are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P).

[0011] In certain embodiments, said XTEN has at least 90%, at least 92%, at least 95%, at least 98%, at least 99% or 100% sequence identity to a sequence set forth in **Table 3a**. In certain embodiments, said barcode fragment (**BAR**) does not include a glutamic acid that is immediately adjacent to another glutamic acid, if present, in said XTEN. In certain embodiments, said barcode fragment (**BAR**) has a glutamic acid at its C-terminus. In certain embodiments, said barcode fragment (**BAR**) has an N-terminal amino acid that is immediately preceded by a glutamic acid residue. In certain embodiments, said barcode fragment (**BAR**) is positioned a distance from either said N-terminus of said polypeptide or said C-terminus of said polypeptide, wherein said distance is from 10 to 150 amino acids, or from 10 to 125 amino acids in length. In certain embodiments, said barcode fragment (**BAR**) is characterized in that: (i) it does not include a glutamic acid that is immediately adjacent to another glutamic acid, if present, in said XTEN; (ii) it has a glutamic acid at its C-terminus; (iii) it has an N-terminal amino acid that is immediately preceded by a glutamic acid residue; and (iv) it is positioned a distance from either said N-terminus of said polypeptide or said C-terminus of said polypeptide, wherein said distance is from 10 to 150 amino acids, or from 10 to 125 amino acids in length. In certain embodiments, said glutamic acid residue that precedes said N-terminal amino acid of said barcode fragment (**BAR**) is not immediately adjacent to another glutamic acid residue.

[0012] In certain embodiments, said barcode fragment (**BAR**) does not include a second glutamic acid residue at a position other than the C-terminus of said barcode fragment unless said second glutamic acid is immediately followed by a proline. In certain embodiments, said XTEN is positioned N-terminal of said bispecific antibody

construct (**BsAb**), wherein said barcode fragment (**BAR**) is positioned within 200, within 150, within 100, or within 50 amino acids of said N-terminus of said polypeptide. In certain embodiments, said XTEN is positioned N-terminal of said bispecific antibody construct (**BsAb**), and wherein said barcode fragment (**BAR1**) is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from said N-terminus of said protein. In certain embodiments, said XTEN is positioned C-terminal of said bispecific antibody construct (**BsAb**), and wherein said barcode fragment (**BAR**) is positioned within 200, within 150, within 100, or within 50 amino acids of said C-terminus of said polypeptide. In certain embodiments, said XTEN is positioned C-terminal of said bispecific antibody construct (**BsAb**), and said barcode fragment (**BAR**) is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from said C-terminus of said protein. In certain embodiments, said barcode fragment (**BAR**) is at least 4 amino acids in length. In certain embodiments, said barcode fragment (**BAR**) is between 4 and 20, between 5 and 15, between 6 and 12, or between 7 and 10 amino acids in length.

[0013] In certain embodiments, said barcode fragment (**BAR**) comprises an amino acid sequence set forth in **Table 2**. In certain embodiments, said XTEN has a length defined by a proximal end and a distal end, wherein (1) said proximal end is positioned, relative to said distal end, closer to said bispecific antibody construct (**BsAb**), and wherein (2) said barcode fragment (**BAR**) is positioned within a region of said XTEN that extends, as measured from said distal end, between 5% and 50%, between 7% and 40%, or between 10% and 30% of said length of said XTEN. In certain embodiments, said XTEN further comprises additional one or more barcode fragments, wherein said additional one or more barcode fragments each differ in sequence and molecular weight from all other peptides fragments that are releasable from said polypeptide upon complete digestion of said polypeptide by said protease. In certain embodiments, said release segment (**RS**) comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence identified herein by **SEQ ID NOS: 7001-7626**. In certain embodiments, said protease cleaves on the C-terminal side of glutamic acid residues that are not followed by proline. In certain embodiments, said protease is a Glu-C protease.

[0014] In certain embodiments, the polypeptide is expressed as a fusion protein, wherein said fusion protein, in an uncleaved state, has a structural arrangement from N-terminus to C-terminus that is AF1-AF2-RS-XTEN, AF2-AF1-RS-XTEN, XTEN-RS-AF1-AF2, or XTEN-RS-AF2-AF1. In certain embodiments, said release segment (**RS**) is fused to said bispecific antibody construct (**BsAb**) by a spacer. In certain embodiments, said spacer comprises at least 4 types of amino acids that can be glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P). In certain embodiments, said spacer comprises an amino acid sequence having at least 80%, 90%, or 100% sequence identity to a sequence set forth in **Table C**. In certain embodiments, said

CDR-H1 and said CDR-H2 of said first antigen binding fragment (**AF1**) comprise amino acid sequences of **SEQ ID NOS: 8 and 9**, respectively.

[0015] In certain embodiments, said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:1 or 2**; said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:4 or 5**; and said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**. In certain embodiments, said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:1**; said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:4 or 5**; and said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**. In certain embodiments, said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:2**; said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:4 or 5**; and said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**. In certain embodiments, said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:1**; said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:4**; and said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**. In certain embodiments, said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:2**; said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:5**; and said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**. In certain embodiments, said first antigen binding fragment (**AF1**) comprises four chain variable domain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3), and 4 (FR-H4), each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to an amino acid sequence of **SEQ ID NOS: 60, 64, 65, and 67**, respectively. In certain embodiments, said first antigen binding fragment (**AF1**) comprises four chain variable domain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3), and 4 (FR-H4), each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to an amino acid sequence of **SEQ ID NOS: 61, 64, 65, and 67**, respectively. In certain embodiments, said first antigen binding fragment further comprises four light chain variable domain framework regions (FR-L): FR-L1, FR-L2, FR-L3, and FR-L4, each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to amino acid sequences of **SEQ ID NOS: 51, 52, 53, and 59**, respectively. In certain embodiments, said first antigen binding fragment further comprises four light chain variable domain framework regions (FR-L): FR-L1, FR-L2, FR-L3, and FR-L4, each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to amino acid sequences of **SEQ ID NOS: 51, 52, 54, and 59**, respectively.

[0016] In certain embodiments, said first antigen binding fragment further comprises four light chain variable domain framework regions (FR-L): FR-L1, FR-L2, FR-L3, and FR-L4, each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to amino acid sequences of **SEQ ID NOS: 51, 52, 55, and 59**, respectively. In certain embodiments, said first antigen

binding fragment further comprises four light chain variable domain framework regions (FR-L): FR-L1, FR-L2, FR-L3, and FR-L4, each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to amino acid sequences of **SEQ ID NOS: 51, 52, 56, and 59**, respectively. In certain embodiments, said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein: said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**; said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**; said FR-L3 comprises an amino acid sequence of **SEQ ID NO:53, 54, 55, or 56**; said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**; said FR-H1 comprises an amino acid sequence of **SEQ ID NO:60 or 61**; said FR-H2 comprises an amino acid sequence of **SEQ ID NO:64**; said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

[0017] In certain embodiments, said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein: said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**; said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**; said FR-L3 comprises an amino acid sequence of **SEQ ID NO:53**; said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**; said FR-H1 comprises an amino acid sequence of **SEQ ID NO:60**; said FR-H2 comprises an amino acid sequence of **SEQ ID NO:64**; said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

[0018] In certain embodiments, said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein: said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**; said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**; said FR-L3 comprises an amino acid sequence of **SEQ ID NO:54**; said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**; said FR-H1 comprises an amino acid sequence of **SEQ ID NO:61**; said FR-H2 comprises an amino acid sequence of **SEQ ID NO:64**; said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

[0019] In certain embodiments, said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein: said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**; said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**; said FR-L3 comprises an amino acid sequence of **SEQ ID NO:55**; said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**; said FR-H1 comprises an amino acid sequence of **SEQ ID NO:61**; said FR-H2 comprises an amino acid sequence of

SEQ ID NO:64; said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

[0020] In certain embodiments, said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein: said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**; said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**; said FR-L3 comprises an amino acid sequence of **SEQ ID NO:56**; said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**; said FR-H1 comprises an amino acid sequence of **SEQ ID NO:61**; said FR-H2 comprises an amino acid sequence of **SEQ ID NO:64**; said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

[0021] In certain embodiments, said first antigen binding fragment (**AF1**) exhibits a higher thermal stability than an anti-CD3 binding fragment having a sequence set forth in **SEQ ID NO: 206**, as evidenced in an *in vitro* assay by a higher melting temperature (T_m) of said first antigen binding fragment relative to that of said anti-CD3 binding fragment; or upon incorporating said first antigen binding fragment into a test bispecific antigen binding construct, a higher T_m of said test bispecific antigen binding construct relative to that of a control bispecific antigen binding construct, wherein said test bispecific antigen binding construct comprises said first antigen binding fragment and a reference antigen binding fragment that binds to an antigen other than CD3; and wherein said control bispecific antigen binding construct consists of said anti-CD3 binding fragment consisting of said sequence of **SEQ ID NO:206** and said reference antigen binding fragment. In certain embodiments, said T_m of said first antigen binding fragment is at least 2°C greater, or at least 3°C greater, or at least 4°C greater, or at least 5°C greater than said T_m of said anti-CD3 binding fragment consisting of said sequence of **SEQ ID NO:206**. In certain embodiments, said first antigen binding fragment (**AF1**) comprises a heavy chain variable region (VH_1), wherein said VH_1 comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or being identical to an amino acid sequence of **SEQ ID NO:102 or 105**. In certain embodiments, said first antigen binding fragment (**AF1**) comprises a light chain variable region (VL_1), wherein said VL_1 comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to an amino acid sequence of any one of **SEQ ID NOS: 101, 103, 104, 106, or 107**. In certain embodiments, said VH_1 and said VL_1 is linked by a linker comprising an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table A**. In certain embodiments, said first antigen binding fragment (**AF1**) comprises an amino acid sequence having at least 95%, 96%, 97%, 98%, 99% sequence identity or being identical to an amino acid sequence of any one of **SEQ ID NOS: 201-205**. In certain embodiments, said first antigen binding fragment (**AF1**) specifically binds human or cynomolgus

monkey (cyno) CD3. In certain embodiments, said first antigen binding fragment (**AF1**) specifically binds human CD3. In certain embodiments, said first antigen binding fragment (**AF1**) binds a CD3 complex subunit that is CD3 epsilon, CD3 delta, CD3 gamma, or CD3 zeta unit of CD3. In certain embodiments, said first antigen binding fragment (**AF1**) binds a CD3 epsilon fragment of CD3. In certain embodiments, said first antigen binding fragment (**AF1**) exhibits an isoelectric point (pI) that is less than or equal to 6.6. In certain embodiments, said first antigen binding fragment (**AF1**) exhibits an isoelectric point (pI) that is between 6.0 and 6.6, inclusive.

[0022] In certain embodiments, said first antigen binding fragment (**AF1**) exhibits an isoelectric point (pI) that is at least 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, or 1.0 pH units lower than the pI of a reference antigen binding fragment having a sequence shown in **SEQ ID NO: 206**. In certain embodiments, said first antigen binding fragment (**AF1**) specifically binds human or cyno CD3 with a dissociation constant (K_d) constant between about between about 10 nM and about 400 nM, as determined in an *in vitro* antigen-binding assay comprising a human or cyno CD3 antigen. In certain embodiments, said first antigen binding fragment (**AF1**) specifically binds human or cyno CD3 with a dissociation constant (K_d) of less than about 10 nM, or less than about 50 nM, or less than about 100 nM, or less than about 150 nM, or less than about 200 nM, or less than about 250 nM, or less than about 300 nM, or less than about 350 nM, or less than about 400 nM, as determined in an *in vitro* antigen-binding assay. In certain embodiments, said first antigen binding fragment (**AF1**) exhibits a binding affinity to CD3 that is at least 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, or at least 10-fold weaker relative to that of an antigen binding fragment consisting of an amino acid sequence of **SEQ ID NO:206**, as determined by the respective dissociation constants (K_d) in an *in vitro* antigen-binding assay.

[0023] In certain embodiments, said first antigen binding fragment (**AF1**) is a chimeric or a humanized antigen binding fragment. In certain embodiments, said first antigen binding fragment (**AF1**) is Fv, Fab, Fab', Fab'-SH, linear antibody, or single-chain variable fragment (scFv). In certain embodiments, said second antigen binding fragment (**AF2**) is Fv, Fab, Fab', Fab'-SH, linear antibody, a single domain antibody, or single-chain variable fragment (scFv). In certain embodiments, said first and second antigen binding fragments are configured as an $(Fab')_2$ or a single chain diabody. In certain embodiments, said second antigen binding fragment (**AF2**) comprises a heavy chain variable region (VH_{II}) comprising an amino acid sequence identified herein by **SEQ ID NOS: 778-783**, and a light chain variable region (VL_{II}) comprising an amino acid sequence identified herein by **SEQ ID NOS: 878-883**. In certain embodiments, said VH_{II} and said VL_{II} is linked by a linker comprising an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table A**. In certain embodiments, said first and second antigen binding fragments are fused together by a peptide linker. In certain embodiments, said peptide linker comprises 2 or 3 types of amino acids that are glycine, serine, or proline. In certain embodiments, said peptide linker

comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table B**.

[0024] In certain embodiments, said **XTEN** is a first extended recombinant polypeptide (**XTEN1**); formed from a plurality of non-overlapping sequence motifs, comprising a first plurality of non-overlapping sequence motifs; said **BAR** is a first barcode fragment (**BAR1**); and said **RS** is a first release segment (**RS1**); and the polypeptide further comprising: (d) a second extended recombinant polypeptide (**XTEN2**), comprising a second barcode fragment (**BAR2**) releasable from said polypeptide upon digestion by said protease; and (e) a second release segment (**RS2**) positioned between said second XTEN (**XTEN2**) and said bispecific antibody construct (**BsAb**), wherein said **XTEN2** is characterized in that:(i) it comprises at least 100, or at least 150 amino acids; (ii) at least 90% of its amino acid residues that are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P); and (iii) it comprises at least 4 different types of amino acids that are G, A, S, T, E, or P; wherein said second barcode fragment (**BAR2**) differs in sequence and molecular weight from all other peptides fragments that are releasable from said polypeptide upon complete digestion of said polypeptide by said protease; and wherein said second barcode fragment (**BAR2**) does not include said N-terminal amino acid or said C-terminal amino acid of said polypeptide. In certain embodiments, said **XTEN1** is positioned N-terminal of said bispecific antibody construct and said **XTEN2** is positioned C-terminal of said bispecific antibody construct. In certain embodiments, said **XTEN1** is positioned C-terminal of said bispecific antibody construct and said **XTEN2** is positioned N-terminal of said bispecific antibody construct.

[0025] In certain embodiments, said **XTEN2** is formed from a second plurality of non-overlapping sequence motifs that are each from 9 to 14 amino acids in length, wherein said second plurality of non-overlapping sequence motifs comprise: (1) a second set of non-overlapping sequence motifs repeated at least two times in said second XTEN; and (2) a non-overlapping sequence motif that occurs only once within said second XTEN; and wherein said second barcode fragment (**BAR2**) includes at least part of said non-overlapping sequence motif that occurs only once within said second XTEN. In certain embodiments, said second set of non-overlapping sequence motifs are each independently identified herein by **SEQ ID NOS: 179-200 and 1715-1722**. In certain embodiments, said second set of non-overlapping sequence motifs are each independently identified herein by **SEQ ID NOS: 186-189**. In certain embodiments, said second set of non-overlapping sequence motifs comprise at least two, at least three, or all four of the sequence motifs **SEQ ID NOS: 186-189**. In certain embodiments, said XTEN2 comprises a length of from 100 to 3,000, from 150 to 3,000, from 100 to 1,000, or from 150 to 1,000 amino acid residues. In certain embodiments, said XTEN2 comprises a length of at least 200, at least 250, at least 300, at least 350, at least 400, at least 450, or at least 500 amino acid residues.

[0026] In certain embodiments, at least 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% of the amino acid residues of said XTEN2 are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or

proline (P). In certain embodiments, said XTEN2 has at least 90%, at least 92%, at least 95%, at least 98%, at least 99% or 100% sequence identity to a sequence set forth in **Table 3a**. In certain embodiments, said second barcode fragment (**BAR2**) does not include a glutamic acid that is immediately adjacent to another glutamic acid, if present, in said XTEN2. In certain embodiments, said second barcode fragment (**BAR2**) has a glutamic acid at its C-terminus. In certain embodiments, said second barcode fragment (**BAR2**) has an N-terminal amino acid that is immediately preceded by a glutamic acid residue. In certain embodiments, said second barcode fragment (**BAR2**) is positioned a distance from either said N-terminus of said polypeptide or said C-terminus of said polypeptide, wherein said distance is from 10 to 150 amino acids, or from 10 to 125 amino acids in length. In certain embodiments, said second barcode fragment (**BAR2**) is characterized in that (i) it does not include a glutamic acid that is immediately adjacent to another glutamic acid, if present, in said XTEN2; (ii) it has a glutamic acid at its C-terminus; (iii) it has an N-terminal amino acid that is immediately preceded by a glutamic acid residue; and (iv) it is positioned a distance from either said N-terminus of said polypeptide or said C-terminus of said polypeptide, wherein said distance is from 10 to 150 amino acids, or from 10 to 125 amino acids in length.

[0027] In certain embodiments, said glutamic acid residue that precedes said N-terminal amino acid of said **BAR2** is not immediately adjacent to another glutamic acid residue. In certain embodiments, said second barcode fragment (**BAR2**) does not include a second glutamic acid residue at a position other than the C-terminus of said second barcode fragment (**BAR2**) unless said second glutamic acid is immediately followed by a proline. In certain embodiments, said **XTEN2** is positioned N-terminal of said bispecific antibody construct (**BsAb**), and wherein said second barcode fragment (**BAR2**) is positioned within 200, within 150, within 100, or within 50 amino acids of said N-terminus of said polypeptide. In certain embodiments, said **XTEN2** is positioned N-terminal of said bispecific antibody construct (**BsAb**), and wherein said second barcode fragment (**BAR2**) is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from said N-terminus of said protein. In certain embodiments, said **XTEN2** is positioned C-terminal of said bispecific antibody construct (**BsAb**), and wherein said second barcode fragment (**BAR2**) is positioned within 200, within 150, within 100, or within 50 amino acids of said C-terminus of said polypeptide. In certain embodiments, said **XTEN2** is positioned C-terminal of said bispecific antibody construct (**BsAb**), and said second barcode fragment (**BAR2**) is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from said C-terminus of said protein.

[0028] In certain embodiments, said second barcode fragment (**BAR2**) is at least 4 amino acids in length. In certain embodiments, said second barcode fragment (**BAR2**) is between 4 and 20, between 5 and 15, between 6 and 12, or between 7 and 10 amino acids in length. In certain embodiments, said second barcode fragment

(**BAR2**) comprises an amino acid sequence as set forth in **Table 2**. In certain embodiments, said **XTEN2** has a length defined by a proximal end and a distal end, wherein (1) said proximal end of said **XTEN2** is positioned, relative to said distal end, closer to said bispecific antibody construct (**BsAb**), and wherein (2) said second barcode fragment (**BAR2**) is positioned within a region of said **XTEN2** that extends, as measured from said distal end of said **XTEN2**, between 5% and 50%, between 7% and 40%, or between 10% and 30% of said length of said **XTEN2**. In certain embodiments, said **XTEN2** further comprises additional one or more barcode fragments, wherein said additional one or more barcode fragments of said **XTEN2** each differ in sequence and molecular weight from all other peptides fragments that are releasable from said polypeptide upon complete digestion of said polypeptide by said protease. In certain embodiments, said first release segment (**RS1**) and said second release segment (**RS2**) are identical in sequence. In certain embodiments, said first release segment (**RS1**) and said second release segment (**RS2**) are not identical in sequence. In certain embodiments, said second release segment (**RS2**) comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence identified herein by **SEQ ID NOS:7001-7626**. In certain embodiments, said first release segment (**RS1**) and said second release segment (**RS2**) are each a substrate for cleavage by multiple proteases at one, two, or three cleavage sites within each release segment sequence.

[0029] In certain embodiments, a polypeptide is expressed as a fusion protein, wherein said fusion protein, in an uncleaved state, has a structural arrangement from N-terminus to C-terminus identified herein by XTEN1-RS1-AF1-AF2-RS2-XTEN2, XTEN1-RS1-AF2-AF1-RS2-XTEN2, XTEN2-RS2-AF1-AF2-RS1-XTEN1, XTEN2-RS2-AF2-AF1-RS1-XTEN1, XTEN1-RS1-diabody-RS2-XTEN2, or XTEN2-RS2-diabody-RS1-XTEN1, wherein said diabody comprises a light chain variable region (VL_I) of said AF1, a heavy chain variable region (VH_I) of said AF1, a light chain variable region (VL_{II}) of said AF2, and a heavy chain variable region (VH_{II}) of said AF2. In certain embodiments, said spacer of said first release segment (**RS1**) is a first spacer, and wherein said second release segment (**RS2**) is fused to said bispecific antibody construct (**BsAb**) by a second spacer. In certain embodiments, said second spacer comprises at least 4 types of amino acids that are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P). In certain embodiments, said second spacer comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table C**.

[0030] In certain embodiments, said **XTEN1** comprises an amino acid sequence having at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence set forth in **Table 3a**; said **BsAb** comprises: said **AF1** comprising light chain complementarity-determining regions 1 (CDR-L1), 2 (CDR-L2), and 3 (CDR-L3) and heavy chain complementarity-determining regions 1 (CDR-H1), 2 (CDR-H2), and 3 (CDR-H3), wherein said CDR-H1, said CDR-H2, and said CDR-H3 comprise amino acid sequences

of **SEQ ID NOS: 8, 9, and 10**, respectively; and said **AF2** comprising a light chain variable region (VL_{II}) identified herein by **SEQ ID NOS: 778-783** and a heavy chain variable region (VH_{II}) identified herein by **SEQ ID NOS: 878-883**; said **RS1** comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence identified herein by **SEQ ID NOS: 7001-7626**; said **XTEN2** comprises an amino acid sequence having at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence set forth in **Table 3a**; and said **RS2** comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence identified herein by **SEQ ID NOS: 7001-7626**, wherein said polypeptide has a structural arrangement from N-terminus to C-terminus identified herein as XTEN1-RS1-AF2-AF1-RS2-XTEN2, XTEN1-RS1-AF1-AF2-RS2-XTEN2, XTEN2-RS2-AF2-AF1-RS1-XTEN1, or XTEN2-RS2-AF1-AF2-RS1-XTEN1.

[0031] In certain embodiments, the polypeptide has a terminal half-life that is at least two-fold longer compared to the bispecific antibody construct not linked to any XTEN. In certain embodiments, the polypeptide is less immunogenic compared to the bispecific antibody construct not linked to any XTEN, as ascertained by measuring production of IgG antibodies that selectively bind to said bispecific antibody construct after administration of comparable doses to a subject. In certain embodiments, the polypeptide exhibits an apparent molecular weight factor under physiological conditions that is greater than about 3, greater than about 4, greater than about 5, or greater than about 6. In certain embodiments, the polypeptide comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence identified herein as the sequences in **Table D**.

[0032] In certain embodiments, a pharmaceutical composition comprises a polypeptide as above and one or more pharmaceutically suitable excipients. In certain embodiments, said pharmaceutical composition is formulated for administration to a human or animal by any clinically appropriate route and formulation. In certain embodiments, said pharmaceutical composition is in a liquid form or frozen. In certain embodiments, said pharmaceutical composition is in a pre-filled syringe for a single injection. In certain embodiments, said pharmaceutical composition is formulated as a lyophilized powder to be reconstituted prior to administration.

[0033] In certain embodiments said composition is in a pharmaceutical combination with at least one additional therapeutic agent selected from the group consisting of an antibody, an antibody fragment, an antibody conjugate, a cytotoxic agent, a toxin, a radionuclide, an immunomodulator, a photoactive therapeutic agent, a radiosensitizing agent, a hormone, an anti-angiogenesis agent, and combinations thereof.

[0034] The additional therapeutic agent may be a PD-1/PD-L1(2) inhibitor wherein the PD-1/PD-L1(2) inhibitor is an anti-PD-1 antibody or an anti-PD-L1 antibody or an anti-PD-L2 antibody.

[0035] In certain embodiments the PD-1/PD-L1(2) inhibitor is an anti-PD-1 antibody selected from the group comprising nivolumab (Opdivo, BMS-936558, MDX1106), pembrolizumab (Keytruda, MK-3475, lambrolizumab), pidilizumab (CT-011), PDR-001, JS001, STI-A1110, AMP-224 and AMP-514 (MEDI0680).

[0036] In one embodiment the PD-1/PD-L1(2) inhibitor can be an anti-PD-L1 antibody selected from the group comprising atezolizumab (Tecentriq, MPDL3280A), durvalumab (MEDI4736), avelumab (MSB0010718C), BMS-936559 (MDX1105) and LY3300054. In another embodiment the PD-1/PD-L1(2) inhibitor is an anti-PD-L2 antibody.

[0037] In certain embodiments the combination is a combination pack containing the components separate from one another. In certain embodiments the components are administered in separate dosage forms simultaneously or sequentially for use in the treatment of the same disease.

[0038] In certain embodiments the polypeptide described herein is part of a pharmaceutical combination for use as medicament for treating hyper-proliferative disorders. The hyper-proliferative disorders are selected from the group consisting of cancers of the breast, respiratory tract, brain, reproductive organs, digestive tract, urinary tract, eye, liver, skin, head and neck, thyroid, parathyroid and their distant metastases.

[0039] In certain embodiments, a polypeptide as described herein is used in the preparation of a medicament for the treatment of a disease in a subject. In certain embodiments, said disease is cancer.

[0040] In certain embodiments is a method of treating a disease in a subject, comprising administering to said subject in need thereof one or more therapeutically effective doses of a pharmaceutical composition or a pharmaceutical combination.

[0041] In one embodiment the disease is cancer. In certain embodiments the cancer is selected from the group consisting of glioblastoma, melanoma, cholangio carcinoma, small cell lung cancer, colorectal cancer, prostate cancer, vaginal cancer, angiosarcoma, non-small cell lung cancer, appendiceal cancer, squamous cell cancer, salivary duct carcinoma, adenoid cystic carcinoma, small intestine cancer, and gallbladder cancer.

[0042] Provided herein is a method of treating a disease in a subject, comprising administering to said subject in need thereof one or more therapeutically effective doses of a pharmaceutical composition of as above. In certain embodiments, the pharmaceutical composition is administered to the subject as one or more therapeutically effective doses administered during a therapeutically effective course of treatment. In certain embodiments, the dose is administered to a human or animal by any clinically appropriate route and formulation. In certain embodiments, the subject is a mouse, rat, monkey, or human.

[0043] Also provided herein is a nucleic acid comprising a polynucleotide sequence encoding a polypeptide as described herein; or a reverse complement of said polynucleotide sequence thereof. Further provided herein is an expression vector comprising said polynucleotide sequence of as described herein and a recombinant regulatory sequence operably linked to said polynucleotide sequence. Provided herein is a host cell, comprising

said expression vector. In certain embodiments, the host cell is a prokaryote. In certain embodiments, the host cell is *E. coli*. In certain embodiments, the host cell is a mammalian cell.

[0044] Additional aspects and advantages of the present disclosure will become readily apparent to those skilled in this art from the following detailed description, wherein only illustrative embodiments of the present disclosure are shown and described. As will be realized, the present disclosure is capable of other and different embodiments, and its several details are capable of modifications in various obvious respects, all without departing from the disclosure. Accordingly, the drawings and description are to be regarded as illustrative in nature, and not as restrictive.

INCORPORATION BY REFERENCE

[0045] All publications, patents, and patent applications mentioned in this specification are herein incorporated by reference to the same extent as if each individual publication, patent, or patent application was specifically and individually indicated to be incorporated by reference.

BRIEF DESCRIPTION OF THE DRAWINGS

[0046] Various features of this disclosure are set forth with particularity in the appended claims. A better understanding of the features and advantages of the present disclosure will be obtained by reference to the following detailed description that sets forth illustrative embodiments, in which the principles of the invention are utilized, and the accompanying drawings of which:

[0047] **Figure 1** depicts a mixture of XTENylated Protease-Activated T Cell Engager ("XPAT") polypeptides having varying lengths of XTEN. The full-length XPAT (top) comprises a 288 amino acid-long XTEN at the N-terminus and a 864 amino acid-long XTEN at the C-terminus. Various truncations can occur in the XPAT in one or both of the N- and C-terminal XTENs, for example, during fermentation, purification or other steps in product preparation. While products having limited truncations (truncations near a distal end an XTEN) may function in a manner similar to the full-length construct, severe truncations (truncations closer to the proximal end of the XTEN) may possess significantly different pharmacological properties from their full-length counterparts. The presence of truncations poses a challenge for quantifying the pharmacologically efficacious and inefficacious variants in an XPAT product. As illustrated in Figure 1 using the full-length XPAT, each XTEN has a proximal end and a distal end, wherein the proximal end is positioned, relative to the distal end, closer to the biologically active polypeptide (e.g., T-cell engager, cytokine, monoclonal antibody (mAb), antibody fragment, or other protein that is XTENylated).

[0048] **Figure 2** depicts a mixture of XPAT polypeptides having varying lengths of barcoded XTEN. In the full-length XPAT (top), the 288 amino acid-long N-terminal XTEN contains three cleavably fused barcode sequences, "NA," "NB," and "NC" (from distal end to proximal end), and the 864 amino acid-long C-terminal XTEN contains three cleavably fused barcode sequences, "CC," "CB," and "CA" (from proximal end to distal

end). Each barcode is positioned to indicate a pharmacologically-relevant length of the corresponding XTEN. For example, minor N-terminal truncation products of the XPAT, lacking the barcode "NA" (e.g., due to truncation) but having the more proximal barcodes "NB" and "NC," may show substantially the same pharmacological properties as the full-length construct. In contrast, major N-terminal truncation products of the XPAT, e.g., lacking all three barcodes (e.g., due to truncation) on the N-terminus, may discernibly differ in pharmacological activity from the full-length construct. A unique proteolytically-cleavable sequence is identified from the biologically-active polypeptide (here, the tandem scFvs that comprise the active portion of the T-cell engager) of the XPAT. Due to its presence in all the length variants of the XPAT (including full-length XPAT, minor truncations, and major truncations thereof), the unique proteolytically-cleavable sequence can be used as a reference for quantifying the amounts of various truncation products in relation to the total amount of the biologically active protein.

[0049] **Figure 3** illustrates a potential design for a barcoded XTEN by inserting a barcode-generating sequence into a general-purpose (or regular) XTEN. The exemplary general-purpose (or regular) XTEN (top) comprises non-overlapping 12-mer motifs in the sequence "BCDABDCDABDCBDCDABDCB," wherein the sequence motifs "A," "B," "C," and "D" occur 3, 6, 5, and 7 times, respectively. Glu-C protease digest of the exemplary general-purpose XTEN (upper panel) does not yield unique peptides except both termini ("NT" and "CT"). The insertion of a barcode-generating sequence, "X" (e.g., a unique 12-mer), into the XTEN results in a unique proteolytically-cleavable sequence (or barcode sequence) that does not occur anywhere else in the XTEN. The barcode-generating sequence, "X," can be positioned such that the resulting barcode marks a pharmacologically-relevant length of the XTEN. For example, an XTEN lacking a barcode due to truncation may functionally differ from the corresponding XTEN with the barcode. One of ordinary skill in the art will understand that the barcode-generating sequence ("X") can be the barcode sequence itself. Alternatively, the barcode-generating sequence ("X") can differ from the resulting barcode sequence. For example, the barcode sequence can overlap with and, thus, contain part of the preceding or following 12-mer motif.

[0050] **Figures 4A-4B** illustrate the quantification of the level of truncation for an N-terminal XTEN. Figure 4A demonstrates that a barcoded XTEN (bottom panel) can be constructed by replacing a sequence motif in a general-purpose XTEN (top panel) (e.g., the third sequence motif from the N-terminus, "D") with a barcode-generating motif, "X"; and, in this example, the barcode-generating motif ("X") is itself the unique proteolytically-cleavable barcode sequence. As shown in the bottom panel in Figure 4A, the barcode is positioned such that all the severe truncation forms of the XTEN lack the barcode, and all the limited truncation forms of the XTEN contain the barcode. Figure 4B illustrates the relative abundance of various cleavage products in two different mixtures of XPAT. In one of the mixtures, the barcode is present in 99% of the constructs that contain the biologically active protein. In the other one of the mixtures, 13% of the constructs

are lack a barcode (e.g., due to truncation). Figures 4A-4B illustrate the use of barcoded XTEN to differentiate between two polypeptide mixtures having substantially similar average molecular weights but discernibly different pharmacological activities.

[0051] **Figures 5A-5B** and **6A-6C** illustrate dose-dependent cytotoxicity of masked (XTENylated) and unmasked bispecific T-cell engagers against target cells with various levels of target antigen expression. Proteolytically-unmasked (de-XTENylated) bispecifics demonstrate potent cytotoxicity against various tumor lines, for example, with EC50s in the single-digit pM range when tested with SK-OV-3 and BT-474 cells. XTENylation further demonstrate robust masking capability, e.g., in shielding a bispecific T-cell engager from forming an immune synapse, thereby resulting in reduced toxicity as indicated by a right-ward shift in the concentration-response curve.

[0052] **Figures 5A-5B** illustrate effective masking by XTENs on XTENylated Protease-Activated T-cell engagers ("XPATs") in general, and on HER2-XPATs in particular. For example, cytotoxicity of the XTENylated (masked) HER2-XPAT (e.g., set forth in Table D) and the corresponding de-XTENylated (unmasked, activated) HER2-PAT against two different HER2-expressing (e.g., cancer) cell lines was observed in a dose-dependent manner. The unmasked, de-XTENylated PAT (indicated as solid circles) yielded EC50 values of 3.4 picomolar (pM) (SKOV3 cells) (**Figure 5A**) and 4.8 pM (BT474 cells) (**Figure 5B**) respectively, while the corresponding masked, XTENylated PAT (indicated as solid squares) yielded EC50 values of 44,474 pM (SKOV3 cells) and 49,370 pM (BT474 cells), indicating a masking effect of at least 10^4-fold.

[0053] **Figure 6A** depicts effective masking by XTENs on a bispecific T-cell engager when in contact with non-cancerous tissue (cardiomyocytes). More particularly, **Figure 6A** shows the cytotoxicity of the XTENylated (masked) HER2-XPAT (e.g., set forth in Table D) and the corresponding de-XTENylated (unmasked, activated) HER2-PAT against cardiomyocytes. Killing of the cardiomyocytes by T cell-directed cytolysis in response to unmasked, de-XTENylated PAT was observed (with an approximate EC50 concentration of 64 pM), while in contrast to tumor cells, the cardiomyocytes remained refractory to killing by masked, XTENylated PAT at concentrations as high as 1 micromolar (μM).

[0054] **Figures 6B-6C** illustrate robust masking by XTENs on bispecific T-cell engagers in the context of engaging target cells with relatively medium or low levels of target antigen expression. For example, **Figure 6B** illustrates cytotoxic effects of XTENylated and de-XTENylated Protease-Activated T-cell engagers (PATs) on a cancer cell line with low level of HER2 expression, MCF-7, where masking with XTEN polypeptides reduced T cell-mediated cytotoxicity of the tested HER2-XPAT by approximately 10^4-fold. As another example, cytotoxicity of the XTENylated (masked) HER2-XPAT and the corresponding de-XTENylated (unmasked, activated) HER2-PAT against MDA-MB-453, another cancer cell line with a medium level of HER2 expression, was determined (**Figure 6C**).

[0055] **Figures 7A-7D** illustrate proteolytic activation of XTENylated bispecifics in subjects and robust tumor regressions induced thereby. **Figure 7A** illustrates comparable efficacy induced with equimolar dosing of a cleavable XTENylated HER2 T-cell engager ("HER2-XPAT," indicated as hexagons) and a corresponding unmasked HER2 T-cell engager ("HER2-PAT," indicated as triangles) in (e.g., BT-474) tumor-bearing mice. (** indicates $p < 0.01$.) Notably, between the two tested XTENylated bispecifics, tumor regression was observed on the cleavable XTENylated construct (indicated as hexagons) but not on the non-cleavable XTENylated counterpart (indicated as diamonds), indicating that proteolytic unmasking (de-XTENylation) is a prerequisite for efficacy. **Figure 7B** illustrates efficacy of cleavable XTENylated bispecific (e.g., HER2-XPAT) against large tumors with a single dose (e.g., 2.1 milligrams per kilograms (mpk)). The non-cleavable constructs used in the experiments are identical to the corresponding cleavable constructs, but the release site has been replaced with a non-cleavable sequence of similar length made from GASTEP amino acids (glycine, alanine, serine, threonine, glutamate, and/or proline). **Figure 7C** shows efficacy of the masked HER2-XPAT (solid triangles) at two different concentrations (15nmol/Kg and 36 nmol/Kg), compared to unmasked HER2-PAT (solid squares) and non-cleavable XPAT (solid diamonds). The data for the tumors treated with vehicle and vehicle + PBMCs is also shown. **Figure 7D** shows the percentage cleavage of HER2-PAT in vivo in BT-474 tumor bearing mice.

[0056] **Figure 8** illustrates lymphocyte margination induced in subjects by administration of XTENylated HER2-XPATs. For example, upon a single-dose intravenous infusion (e.g., 25 mg/kg at a dose volume of 10 ml/kg) of an XTENylated, HER2-XPAT (e.g., set forth in Table D), a decrease in lymphocyte count (hematology) was observed, in both female and male monkey subjects (as indicated by triangles and squares, respectively), starting at 6 hours post-dose and lasting till at least 24-72 hours post-dose.

[0057] **Figures 9A-9B.** **Figure 9A** illustrates stability of XTENylated Protease-Activated T-cell engagers (PATs) in plasma circulation in subjects (e.g., cynomolgus monkey) (e.g., at 25 mg/kg dose). Lack of significant increase in XTENylated PAT clearance relative to its non-cleavable form indicates minimal cleavage in periphery. Comparable pharmacokinetics were observed between the tested cleavable HER2-XPAT (solid triangle and solid square) and the non-cleavable counterpart (unfilled triangle and unfilled square). The non-cleavable constructs used in the experiments are identical to the corresponding cleavable constructs, but the release site has been replaced with a non-cleavable sequence of similar length made from GASTEP amino acids (glycine, alanine, serine, threonine, glutamate, and/or proline). **Figure 9B** shows that there is a low frequency of HER2-XPAT proteolytic metabolites in circulation even 96 hours post administration.

[0058] **Figures 10A-10C.** **Figure 10A** shows a single dose, single subject HER2-XPAT dose escalation and demonstrates that all doses up to 42 mg/kg were tolerated. **Figure 10B** shows a single subject HER2-PAT de-escalation scheme in which the maximum tolerated dose of the unmasked HER2-PAT is 0.2mg/kg. **Figure**

10C shows the plasma concentrations of masked HER2-PAT (at a 450-fold higher tolerated Cmax) as compared to unmasked HER2-PAT.

[0059] Figures 11A-11E. Figures 11A and 11B show peripheral T cell Activation data that show there is no peripheral T cell activation in response to HER2-XPAT as compared to HER2-PAT. **Figures 11C-11E** show the levels of cytokines IL-6 (**Figure 11C**), TNF-α (**Figure 11D**) and IFN-γ (**Figure 11E**) in subjects treated with HER2-XPAT and HER2-PAT at varying concentrations, demonstrating that HER2-XPAT does not induce cytokine release even at 50 mg/kg. Note, that the normal range for cytokine levels: IL-6 $\leqslant$ 6 pg/ml, TNFα 1-10 pg/ml, IFNγ $\leqslant$ 10 pg/ml.

[0060] Figure 12. AMX-818 Incubated ex vivo in plasma samples from NHPs and humans showed minimal cleavage to unmasked TCE, even under inflammatory conditions. AMX-818 with a fluorescent label (DyL650) attached to the tandem scFv was incubated in the indicated plasma samples for seven days at 37°C. Samples were then run on a gel and metabolites similar in size to the unmasked, active form of AMX-818 were quantified using a LI-COR detector. Inflammatory disease human samples were derived from patients with rheumatoid arthritis, lupus, inflammatory bowel disease, and multiple sclerosis. Cancer human samples were derived from patients with lung, breast, and colon tumors. Sample sizes: Healthy NHP N = 4, Healthy Human N = 4, "Inflamed" NHP N = 6, "Inflamed" Human N = 27, Cancer Human N = 11.

[0061] Figure 13A-Figure 13B. Provides additional data in support of the safety profile of a preferred HER2-XPAT of the present invention. The data in **Figure 13A** shows a comparable PK between HER2-XPAT and the non-cleavable HER2-PAT formats, demonstrating that the protease release site remains largely stable in circulation of cynomolgus monkeys even at high doses. **Figure 13B** shows that even at high dose of HER2 XPAT there is very limited systemic accumulation of metabolites lacking one or both XTEN masks.

[0062] Figures 14A-14D. AMX818 and 818-PAT Induce Surface Expression of PD-1 on T cells in Response to SKOV3 Tumor Cells. Surface PD-1 expression was evaluated on CD4+ and CD8+ T cells by flow cytometry following a 48-hour co-incubation of PBMCs and SKOV3 cells at a 5:1 Effector:Target ratio with test articles at the indicated concentrations. **Figure 14A and Figure 14C** shows surface PD1 expression on CD4+ T cells in the presence of AMX818 and 818-PAT. **Figure 14B and Figure 14D** shows surface PD1 expression on CD8+ T cells in the presence of AMX818 and 818-PAT.

[0063] Figure 13A-13C. AMX818 and 818-PAT Induce Surface Expression of PD-L1 on T cells in Response to SKOV3 Tumor Cells. **Figure 15A and Figure 15B** show PD-L1 expression on SKOV3 cells at a 5:1 Effector:Target ratio with test articles at the indicated concentrations. **Figure 15C** shows surface HER2 expression on SKOV3 tumor cells.

[0064] Figure 16A-16C. XPATs Are Preferentially Cleaved to Unmasked TCEs in Human Tumors Implanted in Living Mice, with Minimal Cleavage Observed in Healthy Tissues. A single dose of 1.8 mg/kg (13nM) red

fluorophore-labeled (Alexa) XPAT was injected into mice implanted with human tumors (**Figure 16A**). Two days later, tumors and healthy organs were harvested to measure protease cleavage in vivo (results shown in **Figure 16B**). The green-fluorophore-labeled (Dyl800) XPAT was added post-tissue harvesting in the presence of protease inhibitors to account for artifactual cleavage that could result from release of non-relevant intracellular proteases during the processing. By comparing the generated cleavage products of both red and green fluorophore-labeled XPATs, we can determine the cleavage that occurred in the various tissues in vivo vs artifactually during tissue processing. On average 20% of XPAT in the tumor is activated by Day 2 after injection into mice implanted with tumors (n=31 across 9 tumor types; **Figure 16C**).

[0065] The patent or application file contains at least one drawing executed in color. Copies of this patent or patent application publication with color drawings will be provided by the Office upon request and payment of the necessary fee.

DETAILED DESCRIPTION OF PREFERRED EMBODIMENTS

[0066] There is a significant unmet need in cancer therapeutics. While TCEs have been shown to be effective in inducing remission in certain cancers, they have not produced widespread therapeutics due to their extreme potency and on target, off tumor toxicities in healthy tissues. By way of explanation, the TCEs form a bridge between T cells and tumor cells and activate T cell-mediated of the tumor cell and further initiating a cytokine amplification cascades that promotes further killing and potentially provides long term immunity. T cells activated by TCEs to release cytolytic perforin/granzymes in a manner that is independent of antigen-MHC recognition. This creates a two-fold response: direct tumor cell death and amplification of tumor killing through initiation of a powerful cytokine response from the tumor cells. The direct tumor cell death results in release of tumor antigens. The cytokine response includes, among others, increased interferon-γ which stimulates CD8 T cell activity and stimulates antigen presentation by APCs; increased IL2 which causes increased proliferation of activated T-cells, and increased CXCL9 and 10 response which increases T cell recruitment. Together the release of tumor antigens and the initiation of the cytokine response results in activation of the endogenous T-cell response which potentially cases epitope spreading to induce long term immunity.

[0067] The toxicity challenge with TCEs arises out the fact that most tumor targets are, to some extent, also expressed in healthy tissue, and normal cells also can produce the cytokines response resulting in cytokine release syndrome (CRS). These two powerful responses of health tissue to T cell activation by TCEs results in an overall lack therapeutic index for these agents.

[0068] The present invention overcomes the drawbacks in the existing TCEs by providing a conditionally-activated TCE, XPAT or **X**TENylated **P**rotease-**A**ctivated bispecific **T** Cell Engager targeting HER2 (referred to herein as HER2-XPAT, and exemplified as AMX818). More particularly, the XPAT of the present invention

exploits the dysregulated protease activity present in tumors vs. healthy tissues, enabling expansion of the therapeutic index. The XPAT core consists of 2 single chain antibody fragments (scFvs) targeting CD3 and the tumor target(in exemplary embodiments the tumor target is HER2). Two unstructured polypeptide masks (XTENs) are attached to the core that sterically reduce target engagement of either the tumor target and/or CD3 and extend protein half-life. The properties of the XTEN polymer also minimize the potential for immunogenicity, as its lack of stable tertiary structure disfavors antibody binding and the absence of hydrophobic, aromatic and positively charged residues that serve as anchor residues for peptide MHC II binding reduces the potential for T cell epitopes. In humans, minimal immunogenicity of XTEN polymers was observed in >200 patients treated with drugs containing XTEN in the context of half-life extended forms of human growth hormone and Factor VIII. Protease cleavage sites at the base of the XTEN masks enable proteolytic activation of XPAT in the tumor microenvironment, unleashing a small, highly potent TCE that is capable redirecting cytotoxic T cells to kill target-expressing tumor cells. In healthy tissues, where protease activity is tightly regulated, XPATs should remain predominantly inactive as intact prodrugs, thus expanding the therapeutic index compared to unmasked TCEs.

[0069] In addition to localized activation, the short half-life of the unmasked PAT form should further widen the therapeutic index while providing the potency of T-cell immunity to improve the eradication of solid tumors. The release sites used in the XPATs can be cleaved across a broad array of tumors by proteases that are collectively involved in every cancer hallmark (growth; survival and death; angiogenesis; invasion and metastasis; inflammation; and immune evasion). Thus, TCE activity of the XPATs is localized to tumors by exploiting the enhanced protease activity that is upregulated in all stages of cancer and tumor development but is tightly regulated in healthy tissues.

[0070] The components of the exemplary HER2-XPAT, AMX-818, have been optimized to achieve the desired balance between providing sufficient protection in healthy tissue while retaining the necessary potency in tumors across a broad range of cancers. To reduce the potential for T cell activation by the prodrug, a lower binding affinity was selected for the a-CD3 domain in addition to a longer XTEN polymer mask (576 amino acid mask vs 256 amino acids on the HER2 side). To ensure sufficient activation of AMX-818 in the tumor, the protease release site at the base of the XTEN masks was engineered to be cleaved by at least 8 different proteases among 3 different classes that have been reported to be over-expressed or dysregulated in cancer; these include several matrix metalloproteinases (MMPs), Matriptase, uPA, and the cysteine protease, legumain. As a safety checkpoint, co-engagement of both CD3 and HER2 by AMX-818 is required for T cell activation. Activation of T cells should not occur if AMX-818 is unmasked in inflamed tissues where HER2 expression is absent or if it encounters HER2 expressed in healthy tissue where proteases are tightly controlled. This AND-

gate feature is expected to provide preferential activation in the tumor where both elevated protease activity and high HER2 expression are present.

[0071] Thus presence of the XTEN on the XPAT produces an agent that a long half-life, weak target engagement and negligible T-cell activation. Once the XTEN is removed by the action of the proteases in the tumor microenvironment, this preferential activation of the XPATs produces an activated drug (PAT, without the XTEN) that has a short half-life, optimal target engagement, and highly efficient T-cell activation, thereby producing a powerful activated drug with an enhanced therapeutic index. The HER2-XPATs of the present invention a capable of improving the toxicity profile of T cell engagers while maintaining their potency against solid tumors, thus enabling a significant increase in the therapeutic index and expansion of target landscape for this potent modality.

SUMMARY OF DATA GENERATED FROM AMX-818, AN EXEMPLARY HER2-XPAT

[0072] Target binding and in vitro biological activities of AMX-818 have been characterized in multiple studies. Equilibrium binding analysis utilizing surface plasmon resonance demonstrated highly comparable affinities for AMX-818 and its metabolites between human and cynomolgus monkey HER2 and CD3, supporting the use of cynomolgus monkey as a species for toxicity and PK studies. Proteolytically-activated AMX-818(PAT) bound to human and cynomolgus monkey HER2 with 2.4 nM and 2.0 nM affinities, respectively, and to human and cynomolgus monkey CD3 with 26.3 nM and 21.5 nM affinities, respectively. Masking of AMX-818 reduced its affinities to HER2 by 10-fold and to CD3 by approximately 6-fold for both species. AMX-818 bound to human and cynomolgus monkey HER2 with 24.9 nM and 20.1 nM affinities, while CD3 affinities for human and cynomolgus monkey were 160 nM and 140.3 nM, respectively.

[0073] The activity of a TCE depends on its ability to activate T cells through effective stimulation of the T cell receptor (TCR). The extreme potency of TCEs derives from the minimal requirement for as few as 3 TCRs to become stimulated and coalesce to form an immune synapse between the T cell and target cell to initiate cytotoxicity. While T-cell engagers are primarily known for inducing cytotoxicity, their potency also involves cytokine-driven actions downstream of T-cell activation that enhance and amplify the anti-tumor immune response. T-cell activation by AMX-818, its prototype AMX-818-P1, and its proteolytic metabolites was characterized in vitro utilizing a Jurkat NFAT-reporter cell and primary human PBMCs in the presence of HER2-high-expressing tumor cells, BT-474 (breast) and SKOV-3 (ovarian). Human T cells were also assessed for upregulation of the surface activation marker CD69 and the inhibitory receptor PD-1 by flow cytometry. As an indirect measure of T-cell activation, upregulation of the PD-1 ligand PD-L1 was assessed on the surface of SKOV3 tumor targets, as it is induced in response to IFN-γ secreted by activated T cells.

[0074] AMX-818(PAT) activated the Jurkat NFAT-Luciferase reporter T cells in the presence of BT-474 cells with EC_{50} values in the 70pM range while response to the masked AMX-818 and AMX-818-P1 was

significantly attenuated by 4 orders of magnitude, with maximal responses reduced by 80-90% compared to that of the activated AMX-818(PAT). T-cell activation by AMX-818(PAT) was not observed in the absence of HER2+ BT-474 tumor cells, demonstrating that monovalent engagement of CD3 was not sufficient for activation and that co-engagement of both CD3 and tumor target were required for effective T cell receptor (TCR) stimulation. The singly masked AMX-818 metabolites, AMX-818(1x-C), and AMX-818(1x-N) demonstrated intermediate activity. AMX-818-NoClvSite induced no detectable T-cell activation, suggesting that the minimal response observed by the AMX-818 was likely driven by proteolytic cleavage.

[0075] AMX-818(PAT) and prodrug AMX-818 induced CD69 and PD-1 expression on the surface of both CD4+ and CD8+ T-cell subsets and PD-L1 on SKOV3 tumor cells to comparable degrees. However, the dose–response curve for AMX-818 was shifted on average 400 to 650-fold higher relative to that of AMX-818(PAT), further demonstrating effective functional masking of the XTEN masks on AMX-818.

[0076] AMX-818 and its metabolites were characterized for their cytotoxic activity and induction of inflammatory cytokines. Peripheral blood mononuclear cells (PBMCs) were used as effector cells and the HER2-high BT-747 breast tumor line selected as the tumor target cell. Because cardiac tissues are known to express HER2 and, while rare, cardiac toxicities have been observed in patients treated with some HER2-targeted therapies, primary cardiomyocytes (HER2 low to medium) were selected to represent a more physiologic cytolytic target for AMX-818 and its proteolytic metabolites. A luminescence-based cytotoxicity assay was conducted at a 1:1 effector:target cell ratio and cell free supernatants were collected for measurement of TCE-induced cytokines.

[0077] AMX-818(PAT) demonstrated highly potent cytotoxicity against BT-474 tumor cells, showing nearly complete target cell killing with half-maximal inhibitory concentration (IC_{50}) values in the 5–11 pM range. With lower HER2-expressing human cardiomyocytes as targets - cells not expected to exhibit dysregulated protease activity – cytotoxicity responses were reduced by approximately 13-fold and maximal killing was incomplete, approaching only 50–60%. The cytotoxic response by AMX-818 against both BT-474 and cardiomyocyte cells was strongly attenuated, with average IC_{50} values shifted by 2500- to 3000-fold, demonstrating effective functional masking of the prodrug by its XTEN polymer masks. The masks provide synergistic protection in cytotoxicity well above their combined impact on reducing target binding by impeding formation of the functional immune synapse that is required to initiate target cell killing. AMX-818 and its prototype AMX-818-P1 showed comparable cytotoxicity consistent with their nearly identical composition. Cytotoxicity of the singly masked proteolytic metabolites, AMX-818(1x-N) and AMX-818(1x-C) was intermediate between AMX-818(PAT) and AMX-818, demonstrating partial protection by a single mask. The cytotoxicity observed by AMX-818 was likely due to proteolytic cleavage based on the further reduction in activity provided by AMX-818-NoClvSite, the format lacking both protease cleavage sites.

[0078] In general, the relative potencies of AMX-818 and its metabolites in induction of cytokine secretion in supernatants from cytotoxicity assays mirrored those observed in cytotoxicity to both BT-474 and cardiomyocyte target cells (where AMX-818(PAT) was most potent > singly masked forms > and AMX-818 least potent). The cytokine IC_{50} values were on average higher than those for the cytotoxic response demonstrating that cytotoxicity is the more sensitive assay among the two. Responses to AMX-818 in the presence of both BT-474 and cardiomyocytes were reduced by several orders of magnitude relative to those of AMX-818(PAT), while the singly masked metabolites AMX-818(1x-N) and AMX-818(1x-C) showed intermediate response.

[0079] In the presence of cardiomyocytes, the maximal levels of cytokines induced by AMX-818 at its highest concentration tested (300 nM) were markedly reduced relative to those induced by AMX-818(PAT), with the exception of IL-6. In the presence of both BT-474 and cardiomyocytes, elevated IL-6 levels were detected at 300 nM concentrations of both AMX-818 and AMX-818-NoClvSite, that in most cases exceeded the maximal levels produced by AMX-818(PAT) by 2- to 6-fold. Of note, this is in contrast to what is observed *in vivo* in cynomolgus monkeys, where peak systemic levels of IL-6 are > 9-fold higher at the 0.2 mg/kg MTD of AMX-818(PAT) than at the 42 mg/kg MTD of AMX-818.

[0080] In assessing the induction of cytokines in the absence of HER2-expressing target cells, cytokines IL-2, IL-4, TNF-α, and IFN-γ were not induced in human PBMC cultures treated with suspension and plate-coated AMX-818 or AMX-818(PAT). At its highest concentration tested (500 nM), soluble AMX-818 induced low levels of IL-10 from all PBMC donors. IL-6 was a notable exception, where in the soluble format at 500 nM, AMX-818 induced levels of IL-6 from all donors that exceeded those induced by the anti-CD3 antibody positive control by an average of 4.6-fold increase in the means. High levels of IL-6 were also induced by 2 of 5 donors in the wet plate-coated format, and lower levels of IL-6 were also seen at the highest concentration of AMX-818(PAT). However, importantly, such elevated IL-6 levels were not accompanied by increases in TNF-α, IFN-γ, and IL-2, which are cytokines commonly co-associated with IL-6 secretion under conditions of CRS, nor were they observed in cynomolgus monkeys dosed with AMX-818 at up to 50 mg/kg. In contrast, the activated AMX-818(PAT) induced high levels of IL-6 in cynos at doses ≤0.3 mg/kg that were accompanied by increases in the additional inflammatory cytokines.

[0081] *In vivo* pharmacology, PK, and toxicology studies were conducted to characterize the efficacy and safety of AMX-818 and its metabolites. In addition to standard toxicology endpoints, the proteolytic stability of AMX-818 in circulation was evaluated in cynomolgus monkeys administered high doses of AMX-818 or under conditions of induced inflammation and *in vitro* following extended incubation in plasma from patients with cancer or systemic autoimmune disease. Preferential cleavage of AMX-818 in tumors was evident resulting in protease-dependent efficacy, while its peripheral stability in NHP provided a large safety margin

relative to AMX-818(PAT) that predicts an increased therapeutic index. Together, these data support strong conditional masking of AMX818 that enables localized tumor activity with concomitant stability and effective masking in circulation and peripheral tissues.

[0082] Several *in vivo* efficacy studies were conducted to evaluate the impact of AMX-818 in redirecting T cells to kill HER2-expressing tumors. Since AMX-818 is not cross-reactive to mouse HER2 or CD3, immunodeficient mice were inoculated with human HER2-expressing xenograft tumors and engrafted with human PBMC (hPBMC) as a source of effector T cells. Anti-tumor activity of AMX-818 was assessed in the HER2-high BT-474 breast model (~975,000 HER2 receptors) and HER2-low HT-55 colorectal model (25,000 receptors).

[0083] In the BT474 model, administration of equimolar doses of AMX-818 and its prototype AMX-818-P1 at 2.1 mg/kg or the unmasked AMX-818(PAT) at 0.9 mg/kg induced robust and complete tumor regressions. The anti-tumor efficacy of AMX-818 was dependent on protease cleavage of its masks, as demonstrated by the lack of significant tumor growth inhibition in mice treated with a form of AMX-818 lacking its protease release sites (AMX-818-NoClvSite). AMX-818-P1 and AMX-818(PAT) induced comparable activation of T cells in the tumor microenvironment as assessed by upregulation of activation markers CD25 and CD69 on CD4 and CD8+ T cells by flow cytometry. Importantly, T cells were not activated in the periphery even by the unmasked AMX-818(PAT) where human HER2 is not expressed, consistent with a requirement for dual engagement of both HER2 and CD3 to activate T cells and redirect their killing.

[0084] A single 2.1 mg/kg dose of AMX-818 was sufficient to induce tumor regressions in mice bearing large established BT-474 tumors (478 mm^3 mean tumor volume) within 4 days of dosing. The efficacy was dependent on both HER2 expression and T cells, as demonstrated by the lack of activity of a non-tumor binding variant NB-XPAT (a version of AMX-818, in which its HER2 binding domain was replaced with a non-HER2 binding scFv) or of AMX-818 when dosed in tumor-bearing mice lacking hPBMCs. These findings further support the requirement for dual engagement of AMX-818 for its activity and provide a safety measure should AMX-818 become cleaved in normal tissue where HER2 is absent.

[0085] In mice bearing HER2-low HT-55 tumors, AMX-818 also induced dose- and protease-dependent efficacy, with 5.1 mg/kg inducing complete tumor regression in all mice (103% TGI, $p<0.01$), and 2.1 mg/kg inducing 70% tumor growth inhibition. Efficacy by AMX-818 in a model expressing 25,000 HER2 receptors provides potential for treating patients with multiple cancer types that include tumors with low levels of HER2 expression. Finally, preferential unmasking of a fluorophore-labeled AMX-818 to an average of 25.2% AMX-818(PAT) was evident in tumors compared to <2% in heart, brain and liver tissue combined following a 2-day incubation in BT-474 tumor-bearing mice, supporting the key tenants of localized dysregulation of proteases in tumors and the dominance of protease inhibition in normal tissues.

TERMINOLOGY

[0086] As used herein, the following terms have the meanings ascribed to them unless specified otherwise.

[0087] As used in the specification and claims, the singular forms "a," "an," and "the" include plural references unless the context clearly dictates otherwise. For example, the term "a cell" includes a plurality of cells, including mixtures thereof.

[0088] The terms "polypeptide," "peptide," and "protein" are used interchangeably herein to refer to polymers of amino acids of any length. The polymer may be linear or branched, it may comprise modified amino acids, and it may be interrupted by non-amino acids. The terms also encompass an amino acid polymer that has been modified, for example, by disulfide bond formation, glycosylation, lipidation, acetylation, phosphorylation, or any other manipulation, such as conjugation with a labeling component.

[0089] As used herein the term "amino acid" refers to either natural and/or unnatural or synthetic amino acids, including but not limited to glycine and both the D or L optical isomers, and amino acid analogs and peptidomimetics. Standard single or three letter codes are used to designate amino acids.

[0090] A "host cell" includes an individual cell or cell culture which can be or has been a recipient for the subject vectors. Host cells include progeny of a single host cell. The progeny may not necessarily be completely identical (in morphology or in genomic of total DNA complement) to the original parent cell due to naturally occurring or genetically engineered variation

[0091] A "chimeric" protein contains at least one fusion polypeptide comprising regions in a different position in the sequence than that which occurs in nature. The regions may normally exist in separate proteins and are brought together in the fusion polypeptide; or they may normally exist in the same protein but are placed in a new arrangement in the fusion polypeptide. A chimeric protein may be created, for example, by chemical synthesis, or by creating and translating a polynucleotide in which the peptide regions are encoded in the desired relationship.

[0092] "Conjugated", "linked," "fused," and "fusion" are used interchangeably herein. These terms refer to the joining together of two more chemical elements or components, by whatever means including chemical conjugation or recombinant means.

[0093] The terms "polynucleotides", "nucleic acids", "nucleotides" and "oligonucleotides" are used interchangeably. They refer to a polymeric form of nucleotides of any length, either deoxyribonucleotides or ribonucleotides, or analogs thereof. Polynucleotides may have any three-dimensional structure, and may perform any function, known or unknown. A polynucleotide may comprise modified nucleotides, such as methylated nucleotides and nucleotide analogs. If present, modifications to the nucleotide structure may be imparted before or after assembly of the polymer. The sequence of nucleotides may be interrupted by non-

nucleotide components. A polynucleotide may be further modified after polymerization, such as by conjugation with a labeling component.

[0094] As used herein, polynucleotides having "homology" or that are "homologous" are those which hybridize under stringent conditions as defined herein and have at least 70%, preferably at least 80%, more preferably at least 90%, more preferably 95%, more preferably 97%, more preferably 98%, and even more preferably 99% sequence identity to those sequences.

[0095] The terms "percent identity" and "% identity," as applied to polynucleotide sequences, refer to the percentage of residue matches between at least two polynucleotide sequences aligned using a standardized algorithm. Such an algorithm may insert, in a standardized and reproducible way, gaps in the sequences being compared in order to optimize alignment between two sequences, and therefore achieve a more meaningful comparison of the two sequences. Percent identity may be measured over the length of an entire defined polynucleotide sequence, for example, as defined by a particular SEQ ID number, or may be measured over a shorter length, for example, over the length of a fragment taken from a larger, defined polynucleotide sequence, for instance, a fragment of at least 45, at least 60, at least 90, at least 120, at least 150, at least 210 or at least 450 contiguous residues. Such lengths are exemplary only, and it is understood that any fragment length supported by the sequences shown herein, in the tables, figures or Sequence Listing, may be used to describe a length over which percentage identity may be measured.

[0096] "Percent (%) amino acid sequence identity," with respect to the polypeptide sequences identified herein, is defined as the percentage of amino acid residues in a query sequence that are identical with the amino acid residues of a second, reference polypeptide sequence or a portion thereof, 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 measuring alignment, including any algorithms needed to achieve maximal alignment over the full length of the sequences being compared. Percent identity may be measured over the length of an entire defined polypeptide sequence, for example, as defined by a particular SEQ ID number, or may be measured over a shorter length, for example, over the length of a fragment taken from a larger, defined polypeptide sequence, for instance, a fragment of at least 15, at least 20, at least 30, at least 40, at least 50, at least 70 or at least 150 contiguous residues. Such lengths are exemplary only, and it is understood that any fragment length supported by the sequences shown herein, in the tables, figures or Sequence Listing, may be used to describe a length over which percentage identity may be measured.

[0097] As used herein, "repetitiveness" of an XTEN sequence refers to the 3-mer repetitiveness and can be measured by computer programs or algorithms or by other means known in the art. The 3-mer repetitiveness of an XTEN is assessed by determining the number of occurrences of the overlapping 3-mer sequences within the polypeptide. For example, a polypeptide of 200 amino acid residues has 198 overlapping 3-amino acid sequences (3-mers), but the number of unique 3-mer sequences will depend on the amount of repetitiveness within the sequence. A score can be generated (hereinafter "subsequence score") that is reflective of the degree of repetitiveness of the 3-mers in the overall polypeptide sequence. In the context of the present invention, "subsequence score" means the sum of occurrences of each unique 3-mer frame across a 200 consecutive amino acid sequence of the polypeptide divided by the absolute number of unique 3-mer subsequences within the 200 amino acid sequence. Examples of such subsequence scores derived from the first 200 amino acids of repetitive and non-repetitive polypeptides are presented in Example 73 of International Patent Application Publication No. WO 2010/091122 A1, which is incorporated by reference in its entirety. In some embodiments, the present invention provides BPXTEN each comprising XTEN in which the XTEN can have a subsequence score less than 16, or less than 14, or less than 12, or more preferably less than 10.

[0098] The term "substantially non-repetitive XTEN," as used herein, refers to an XTEN, wherein (1) there are few or no instances of four contiguous amino acids in the XTEN sequence that are identical amino acid types and wherein (2) the XTEN has a subsequence score (defined in the preceding paragraph herein) of 12, or 10 or less or that there isn't a pattern in the order, from N- to C-terminus, of the sequence motifs that constitute the polypeptide sequence.

[0099] A "vector" is a nucleic acid molecule, preferably self-replicating in an appropriate host, which transfers an inserted nucleic acid molecule into and/or between host cells. The term includes vectors that function primarily for insertion of DNA or RNA into a cell, replication of vectors that function primarily for the replication of DNA or RNA, and expression vectors that function for transcription and/or translation of the DNA or RNA. Also included are vectors that provide more than one of the above functions. An "expression vector" is a polynucleotide which, when introduced into an appropriate host cell, can be transcribed and translated into a polypeptide(s). An "expression system" usually connotes a suitable host cell comprised of an expression vector that can function to yield a desired expression product.

[0100] The term "$t_{1/2}$" as used herein means the terminal half-life calculated as $\ln(2)/K_{el}$. K_{el} is the terminal elimination rate constant calculated by linear regression of the terminal linear portion of the log concentration vs. time curve. Half-life typically refers to the time required for half the quantity of an administered substance deposited in a living organism to be metabolized or eliminated by normal biological processes. The terms "$t_{1/2}$", "terminal half-life", "elimination half-life" and "circulating half-life" are used interchangeably herein.

[0101] The terms "antigen," "target antigen," or "immunogen" are used interchangeably herein to refer to the structure or binding determinant that an antibody fragment or an antibody fragment-based therapeutic binds to or has specificity against.

[0102] The term "payload" as used herein refers to a protein or peptide sequence that has biological or therapeutic activity; the counterpart to the pharmacophore of small molecules. Examples of payloads include, but are not limited to, cytokines, enzymes, hormones and blood and growth factors. Payloads can further comprise genetically fused or chemically conjugated moieties such as chemotherapeutic agents, antiviral compounds, toxins, or contrast agents. These conjugated moieties can be joined to the rest of the polypeptide via a linker which may be cleavable or non-cleavable.

[0103] As used herein, "treatment" or "treating," "palliating," and "ameliorating" are used interchangeably herein. These terms refer to an approach for obtaining beneficial or desired results including but not limited to a therapeutic benefit and/or a prophylactic benefit. By "therapeutic benefit" is meant eradication or amelioration of the underlying disorder being treated. Also, a therapeutic benefit is achieved with the eradication or amelioration of one or more of the physiological symptoms associated with the underlying disease condition such that an improvement is observed in the subject, notwithstanding that the subject may still be afflicted with the underlying disorder. For prophylactic benefit, the compositions may be administered to a subject at risk of developing a particular disease condition, or to a subject reporting one or more of the physiological symptoms of a disease, even though a diagnosis of this disease may not have been made.

[0104] A "therapeutic effect," as used herein, refers to a physiologic effect, including but not limited to the cure, mitigation, amelioration, or prevention of disease condition in humans or other animals, or to otherwise enhance physical or mental wellbeing of humans or animals, caused by a fusion polypeptide of the invention other than the ability to induce the production of an antibody against an antigenic epitope possessed by the biologically active protein. Determination of a therapeutically effective amount is well within the capability of those skilled in the art, especially in light of the detailed disclosure provided herein.

[0105] The terms "therapeutically effective amount" and "therapeutically effective dose," as used herein, refers to an amount of a biologically active protein, either alone or as a part of a fusion protein composition, that is capable of having any detectable, beneficial effect on any symptom, aspect, measured parameter or characteristics of a disease state or condition when administered in one or repeated doses to a subject. Such effect need not be absolute to be beneficial. The disease condition can refer to a disorder or a disease.

[0106] The term "therapeutically effective dose regimen," as used herein, refers to a schedule for consecutively administered doses of a biologically active protein, either alone or as a part of a fusion protein composition, wherein the doses are given in therapeutically effective amounts to result in sustained beneficial effect on any symptom, aspect, measured parameter or characteristics of a disease state or condition.

FUSION POLYPEPTIDE

[0107] Disclosed herein includes a polypeptide (or fusion polypeptide) comprising one or more extended recombinant polypeptides (XTEN or XTENs) (as described more fully hereinbelow), a bispecific antibody construct (**BsAb**) linked to the XTEN(s), and one or more release segments (**RS**); the release segment is positioned between the XTEN and the bispecific antibody construct (**BsAb**); and the polypeptide (or fusion polypeptide) has an N-terminal amino acid and a C-terminal amino acid.

[0108] In some embodiments, the polypeptide comprises a first XTEN (such as those described below in the "EXTENDED RECOMBINANT POLYPEPTIDE (XTEN)" section or described anywhere else herein). In some embodiments, the polypeptide further comprises a second XTEN (such as those described below in the "EXTENDED RECOMBINANT POLYPEPTIDE (XTEN)" section or described anywhere else herein). In some embodiments, the polypeptide comprises an XTEN at or near its N-terminus (an "N-terminal XTEN"). In some embodiments, the polypeptide comprises an XTEN at or near its C-terminus (a "C-terminal XTEN"). In some embodiments, the polypeptide comprises both an N-terminal XTEN and a C-terminal XTEN. In some embodiments, the first XTEN is an N-terminal XTEN and the second XTEN is a C-terminal XTEN. In some embodiments, the first XTEN is a C-terminal XTEN and the second XTEN is an N-terminal XTEN.

[0109] As the bispecific antibody (BsAb), a biologically active polypeptide ("BP"), is linked to the one or more XTENs within the polypeptide, the polypeptide may be referred to as an XTEN-containing fusion polypeptide: "BPXTEN."

[0110] The XTEN can comprise one or more barcode fragments (as described more fully below) releasable (configured to be released) from the XTEN upon digestion of the fusion polypeptide (or BPXTEN) by a protease. In some embodiments, each barcode fragment differs in sequence and molecular weight from all other peptide fragments (including all other barcode fragments if present) that are releasable from the polypeptide upon complete digestion of the polypeptide by the protease.

[0111] The (fusion) polypeptide can comprise one or more reference fragments (as described more fully below) releasable (configured to be released) from the polypeptide, for example, upon the protease digestion which releases the barcode fragment(s) from the polypeptide. In some embodiments, each reference fragment can be a single reference fragment that differs in sequence and molecular weight from all other peptide fragments that are releasable from the polypeptide upon digestion of the polypeptide by the protease.

[0112] In some embodiments, the polypeptide has an N-terminal amino acid and a C-terminal amino acid. The polypeptide can comprise **(a)** an extended recombinant polypeptide (**XTEN**) comprising a barcode fragment (**BAR**) releasable from the polypeptide upon digestion by a protease; **(b)** a bispecific antibody construct (**BsAb**), comprising a first antigen binding fragment (**AF1**) that specifically binds to cluster of differentiation 3 T cell receptor (CD3), which AF1 comprises light chain complementarity-determining regions 1 (CDR-L1), 2 (CDR-

L2), and 3 (CDR-L3) and heavy chain complementarity-determining regions 1 (CDR-H1), 2 (CDR-H2), and 3 (CDR-H3), wherein the CDR-H3 comprises an amino acid sequence of **SEQ ID NO:10**; and a second antigen binding fragment (**AF2**) that specifically binds to human epidermal growth factor receptor 2 (HER2); and **(c)** a release segment (**RS**) positioned between the XTEN and the bispecific antibody construct, wherein the XTEN is characterized in that: **(i)** it comprises at least 100, or at least 150 amino acids; **(ii)** at least 90% of its amino acid residues are identified herein by glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P); **(iii)** it comprises at least 4 different types of amino acids identified herein by G, A, S, T, E, or P; and **(iv)** the XTEN is formed from a plurality of non-overlapping sequence motifs that are each from 9 to 14 amino acids in length, wherein the plurality of non-overlapping sequence motifs comprise: **(1)** a set of non-overlapping sequence motifs, wherein each non-overlapping sequence motif of the set of non-overlapping sequence motifs is repeated at least two times in the XTEN; and **(2)** a non-overlapping sequence motif that occurs only once within the XTEN; wherein the barcode fragment (**BAR**) includes at least part of the non-overlapping sequence motif that occurs only once within the XTEN; wherein the barcode fragment (**BAR**) differs in sequence and molecular weight from all other peptides fragments that are releasable from the polypeptide upon complete digestion of the polypeptide by the protease; and wherein the barcode fragment (**BAR**) does not include the N-terminal amino acid or the C-terminal amino acid of the polypeptide. The polypeptide can be expressed as a fusion protein. The fusion protein, in an uncleaved state, can have a structural arrangement from N-terminus to C-terminus identified herein by AF1-AF2-RS-XTEN, AF2-AF1-RS-XTEN, XTEN-RS-AF1-AF2, or XTEN-RS-AF2-AF1.

[0113] In some embodiments of the polypeptides of this disclosure, the **XTEN** is a first extended recombinant polypeptide (**XTEN1**); the plurality of non-overlapping sequence motifs, from which the **XTEN1** is formed, is a first plurality of non-overlapping sequence motifs; the **BAR** is a first barcode fragment (**BAR1**); and the **RS** is a first release segment (**RS1**). In some embodiments, the polypeptide can further comprise: **(d)** a second extended recombinant polypeptide (**XTEN2**), comprising: a second barcode fragment (**BAR2**) releasable from the polypeptide upon digestion by the protease; and **(e)** a second release segment (**RS2**) positioned between the second XTEN (**XTEN2**) and the bispecific antibody construct (**BsAb**), wherein the **XTEN2** is characterized in that: **(i)** it comprises at least 100, or at least 150 amino acids; **(ii)** at least 90% of its amino acid residues are identified herein by glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P); and **(iii)** it comprises at least 4 different types of amino acids identified herein by G, A, S, T, E, or P; wherein the second barcode fragment (**BAR2**) differs in sequence and molecular weight from all other peptides fragments that are releasable from the polypeptide upon complete digestion of the polypeptide by the protease; and wherein the second barcode fragment (**BAR2**) does not include the N-terminal amino acid or the C-terminal amino acid of the polypeptide. The **XTEN1** can be positioned N-terminal of the bispecific antibody construct (BsAb) and the

XTEN2 can be positioned C-terminal of the bispecific antibody construct (BsAb). Alternatively, the **XTEN1** can be positioned C-terminal of the bispecific antibody construct (BsAb) and the **XTEN2** can be positioned N-terminal of the bispecific antibody construct (BsAb). In some embodiments of the polypeptide, where **(iv)** the **XTEN2** can be formed from a second plurality of non-overlapping sequence motifs that are each from 9 to 14 amino acids in length, wherein the second plurality of non-overlapping sequence motifs comprise: **(1)** a second set of non-overlapping sequence motifs, wherein each non-overlapping sequence motif of the second set of non-overlapping sequence motifs is repeated at least two times in the second XTEN; and **(2)** a non-overlapping sequence motif that occurs only once within the second XTEN; and wherein the second barcode fragment (**BAR2**) includes at least part of the non-overlapping sequence motif that occurs only once within the second XTEN. The polypeptide can be expressed as a fusion protein, where the fusion protein, in an uncleaved state, can have a structural arrangement from N-terminus to C-terminus identified herein by XTEN1-RS1-AF1-AF2-RS2-XTEN2, XTEN1-RS1-AF2-AF1-RS2-XTEN2, XTEN2-RS2-AF1-AF2-RS1-XTEN1, XTEN2-RS2-AF2-AF1-RS1-XTEN1, XTEN1-RS1-diabody-RS2-XTEN2, or XTEN2-RS2-diabody-RS1-XTEN1. The diabody can comprise a light chain variable region (VL_I) of the AF1, a heavy chain variable region (VH_I) of the AF1, a light chain variable region (VL_{II}) of the AF2, and a heavy chain variable region (VH_{II}) of the AF2.

[0114] In some embodiments of the polypeptides of this disclosure, (a) the first extended recombinant polypeptide (**XTEN1**) can comprise an amino acid sequence having at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence set forth in **Table 3a**; (b) the bispecific antibody construct (**BsAb**) can comprise: (I) the first antigen binding fragment (**AF1**) comprising light chain complementarity-determining regions 1 (CDR-L1), 2 (CDR-L2), and 3 (CDR-L3) and heavy chain complementarity-determining regions 1 (CDR-H1), 2 (CDR-H2), and 3 (CDR-H3), wherein the CDR-H1, the CDR-H2, and the CDR-H3 comprise amino acid sequences of **SEQ ID NOS: 8, 9, and 10**, respectively; and (II) the second antigen binding fragment (**AF2**) comprising a light chain variable region (VL_{II}) identified herein by **SEQ ID NOS: 778-783** and a heavy chain variable region (VH_{II}) identified herein by **SEQ ID NOS: 878-883**; (c) the first release segment (**RS1**) comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence identified herein by **SEQ ID NOS: 7001-7626**; (d) the second extended recombinant polypeptide (**XTEN2**) comprises an amino acid sequence having at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence set forth in **Table 3a**; and (e) the second release segment (**RS2**) comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence identified herein by **SEQ ID NOS: 7001-7626**, where the polypeptide can have a structural arrangement from N-terminus to C-terminus identified herein by: XTEN1-RS1-AF2-AF1-RS2-XTEN2,

XTEN1-RS1-AF1-AF2-RS2-XTEN2, XTEN2-RS2-AF2-AF1-RS1-XTEN1, or XTEN2-RS2-AF1-AF2-RS1-XTEN1.

Extended Recombinant Polypeptide (XTEN)

Chain Length and Amino Acid Composition

[0115] In some embodiments, the XTEN comprises at least 100, or at least 150 amino acids. In some embodiments, the XTEN is from 100 to 3,000, or from 150 to 3,000 amino acids in length. In some embodiments, the XTEN is from 100 to 1,000, or from 150 to 1,000 amino acids in length. In some embodiments, the XTEN is at least (about) 100, at least (about) 150, at least (about) 200, at least (about) 250, at least (about) 300, at least (about) 350, at least (about) 400, at least (about) 450, at least (about) 500, at least (about) 550, at least (about) 600, at least (about) 650, at least (about) 700, at least (about) 750, at least (about) 800, at least (about) 850, at least (about) 900, at least (about) 950, at least (about) 1,000, at least (about) 1,100, at least (about) 1,200, at least (about) 1,300, at least (about) 1,400, at least (about) 1,500, at least (about) 1,600, at least (about) 1,700, at least (about) 1,800, at least (about) 1,900, or at least (about) 2,000 amino acids in length. In some embodiments, the XTEN is at most (about) 100, at most (about) 150, at most (about) 200, at most (about) 250, at most (about) 300, at most (about) 350, at most (about) 400, at most (about) 450, at most (about) 500, at most (about) 550, at most (about) 600, at most (about) 650, at most (about) 700, at most (about) 750, at most (about) 800, at most (about) 850, at most (about) 900, at most (about) 950, at most (about) 1,000, at most (about) 1,100, at most (about) 1,200, at most (about) 1,300, at most (about) 1,400, at most (about) 1,500, at most (about) 1,600, at most (about) 1,700, at most (about) 1,800, at most (about) 1,900, or at most (about) 2,000 amino acids in length. In some embodiments, the XTEN has (about) 100, (about) 150, (about) 200, (about) 250, (about) 300, (about) 350, (about) 400, (about) 450, (about) 500, (about) 550, (about) 600, (about) 650, (about) 700, (about) 750, (about) 800, (about) 850, (about) 900, (about) 950, (about) 1,000, (about)1,100, (about) 1,200, (about) 1,300, (about) 1,400, (about) 1,500, (about) 1,600, (about) 1,700, (about) 1,800, (about) 1,900, or (about) 2,000 amino acids in length, or of a range between any two of the foregoing. In some embodiments, at least 90% of the amino acid residues of the XTEN are identified herein by glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P). In some embodiments, at least 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% of the amino acid residues of the XTEN are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P). In some embodiments, the XTEN comprises at least 4 different types of amino acids are G, A, S, T, E, or P that is substantially randomized with respect to any other nonoverlapping sequence motif comprising the XTEN polypeptide. In some embodiments, the XTEN (e.g., XTEN1, XTEN2, etc.) is characterized in that: (i) it comprises at least 100, or at least 150 amino acids; (ii) at least 90% of the amino acid residues of the XTEN are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P); and (iii) it comprises at least 4 different types of the amino acids from G, A, S, T,

E, or P that is substantially randomized with respect to any other nonoverlapping sequence motif comprising the XTEN polypeptide. One of ordinary skill in the art will understand that, as used herein, the term "glutamate" is a synonym for "glutamic acid," and refers to the glutamic acid residue whether or not the side-chain carboxyl is deprotonated. In some embodiments, the XTEN-containing fusion polypeptide comprises a first XTEN and a second XTEN. In some embodiments, the sum of the total number of amino acids in the first XTEN and the total number of amino acids in the second XTEN is at least 300, at least 350, at least 400, at least 500, at least 600, at least 700, or at least 800 amino acids.

Non-Overlapping Sequence Motif

[0116] In some embodiments, the XTEN comprises, or is formed from, a plurality of non-overlapping sequence motifs. In some embodiments, at least one of the non-overlapping sequence motifs is recurring (or repeated at least two times in the XTEN), and wherein at least another one of the non-overlapping sequence motifs is non-recurring (or found only once within the XTEN). In some embodiments, the plurality of non-overlapping sequence motifs comprises (a) a set of (recurring) non-overlapping sequence motifs, wherein each non-overlapping sequence motif of the set of non-overlapping sequence motifs is repeated at least two times in the XTEN; and (b) a non-overlapping (non-recurring) sequence motif that occurs (or is found) only once within the XTEN. In some embodiments, each non-overlapping sequence motif is from 9 to 14 (or 10 to 14, or 11 to 13) amino acids in length. In some embodiments, each non-overlapping sequence motif is 12 amino acids in length. In some embodiments, the plurality of non-overlapping sequence motifs comprises a set of non-overlapping (recurring) sequence motifs, wherein each non-overlapping sequence motif of the set of non-overlapping sequence motifs is (1) repeated at least two times in the XTEN; and (2) is between 9 and 14 amino acids in length. In some embodiments, the set of (recurring) non-overlapping sequence motifs comprises 12-mer sequence motifs identified herein by **SEQ ID NOs: 179-200** and **1715-1722** in **Table 1**. In some embodiments, the set of (recurring) non-overlapping sequence motifs comprises 12-mer sequence motifs identified herein by **SEQ ID NOs: 186-189** in **Table 1**. In some embodiments, the set of (recurring) non-overlapping sequence motifs comprise at least two, at least three, or all four of 12-mer sequence motifs of **SEQ ID NOs: 186-189** in **Table 1**.

Table 1. Exemplary 12-Mer Sequence Motifs for Construction of XTENs

Motif Family*	Amino Acid Sequence	SEQ ID NO.
AD	GESPGGSSGSES	182
AD	GSEGSSGPGESS	183
AD	GSSESGSSEGGP	184
AD	GSGGEPSESGSS	185
AE, AM	GSPAGSPTSTEE	186
AE, AM, AQ	GSEPATSGSETP	187
AE, AM, AQ	GTSESATPESGP	188

Motif Family*	Amino Acid Sequence	SEQ ID NO.
AE, AM, AQ	GTSTEPSEGSAP	189
AF, AM	GSTSESPSGTAP	190
AF, AM	GTSTPESGSASP	191
AF, AM	GTSPSGESSTAP	192
AF, AM	GSTSSTAESPGP	193
AG, AM	GTPGSGTASSSP	194
AG, AM	GSSTPSGATGSP	195
AG, AM	GSSPSASTGTGP	196
AG, AM	GASPGTSSTGSP	197
AQ	GEPAGSPTSTSE	198
AQ	GTGEPSSTPASE	199
AQ	GSGPSTESAPTE	200
AQ	GSETPSGPSETA	179
AQ	GPSETSTSEPGA	180
AQ	GSPSEPTEGTSA	181
BC	GSGASEPTSTEP	1715
BC	GSEPATSGTEPS	1716
BC	GTSEPSTSEPGA	1717
BC	GTSTEPSEPGSA	1718
BD	GSTAGSETSTEA	1719
BD	GSETATSGSETA	1720
BD	GTSESATSESGA	1721
BD	GTSTEASEGSAS	1722

*Denotes individual motif sequences that, when used together in various permutations, results in a "family sequence"

BARCODE FRAGMENT

[0117] In some embodiments, the polypeptide comprises a barcode fragment (*e.g.*, a first, second, or third barcode fragment of an XTEN) releasable from the polypeptide upon digestion by a protease. In some embodiments, the barcode fragment (1) is a portion of the XTEN that includes at least part of the (non-recurring, non-overlapping) sequence motif that occurs (or is found) only once within the XTEN; and (2) differs in sequence and molecular weight from all other peptide fragments that are releasable from the polypeptide upon complete digestion of the polypeptide by the protease. One of ordinary skill in the art will understand that the term "barcode fragment" (or "barcode," or "barcode sequence") can refer to either the portion of the XTEN cleavably fused within the polypeptide, or the resulting peptide fragment released from the polypeptide.

[0118] In some embodiments, the barcode fragment does not include the N-terminal amino acid or the C-terminal amino acid of the polypeptide. As described more fully below or described anywhere herein, in some embodiments, the barcode fragment is releasable (configured to be released) upon Glu-C digestion of the fusion polypeptide. In some embodiments, the barcode fragment does not include a glutamic acid that is immediately

adjacent to another glutamic acid, if present, in the XTEN. In some embodiments, the barcode fragment has a glutamic acid at its C-terminus. One of ordinary skill in the art will understand that the C-terminus of a barcode fragment can refer to the "last" (or the most C-terminal) amino acid residue within the barcode fragment, when cleavably fused within an XTEN, even if other "non-barcode" amino acid residues are positioned C-terminal to the barcode fragment within the same XTEN. In some embodiments, the barcode fragment has an N-terminal amino acid that is immediately preceded by a glutamic acid residue. In some embodiments, the glutamic acid residue that precedes the N-terminal amino acid is not immediately adjacent to another glutamic acid residue. In some embodiments, the barcode fragment does not include a (second) glutamic acid residue at a position other than the C-terminus of the barcode fragment unless the glutamic acid is immediately followed by a proline. In some embodiments, the barcode fragment is positioned a distance from either the N-terminus of the polypeptide or the C-terminus of the polypeptide, wherein the distance is from 10 to 150, or 10 to 125 amino acids. In some embodiments, the barcode fragment is positioned within, or at a location of, 300, 280, 260, 250, 240, 220, 200, 190, 180, 170, 160, 150, 140, 130, 120, 110, 100, 90, 80, 70, 60, 50, 48, 40, 36, 30, 24, 20, 12, or 10 amino acids from the N-terminus of the polypeptide, or at a location in a range between any of the foregoing. In some embodiments, the barcode fragment is positioned within 200, within 150, within 100, or within 50 amino acids of the N-terminus of the polypeptide. In some embodiments, the barcode fragment is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from the N-terminus of the polypeptide. In some embodiments, the barcode fragment is positioned within, or at a location of, 300, 280, 260, 250, 240, 220, 200, 190, 180, 170, 160, 150, 140, 130, 120, 110, 100, 90, 80, 70, 60, 50, 48, 40, 36, 30, 24, 20, 12, or 10 amino acids from the C-terminus of the polypeptide, or at a location in a range between any of the foregoing. In some embodiments, the barcode fragment is positioned within 200, within 150, within 100, or within 50 amino acids of the C-terminus of the polypeptide. In some embodiments, the barcode fragment is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from the C-terminus of the polypeptide. In some embodiments, the barcode fragment (BAR) is characterized in that: (i) it does not include a glutamic acid that is immediately adjacent to another glutamic acid, if present, in the XTEN; (ii) it has a glutamic acid at its C-terminus; (iii) it has an N-terminal amino acid that is immediately preceded by a glutamic acid residue; and (iv) it is positioned a distance from either the N-terminus of the polypeptide or the C-terminus of the polypeptide, wherein the distance is from 10 to 150 amino acids, or from 10 to 125 amino acids in length. In some embodiments, the barcode fragment (i) does not include the N-terminal amino acid or the C-terminal amino acid of the polypeptide; (ii) does not include a glutamic acid that is immediately adjacent to another glutamic acid in the XTEN; (iii) has a glutamic acid at its C-terminus; (iv) has an N-terminal amino acid that is immediately preceded by a glutamic acid residue; and (v) is positioned a distance from either the N-terminus

of the polypeptide or the C-terminus of the polypeptide, wherein the distance is from 10 to 150, or 10 to 125 amino acids in length. In some embodiments, the glutamic acid residue that precedes the N-terminal amino acid is not immediately adjacent to another glutamic acid residue. In some embodiments, the barcode fragment does not include a glutamic acid residue at a position other than the C-terminus of the barcode fragment unless the glutamic acid is immediately followed by a proline. One or ordinary skill in the art will understand the term "distance," as used herein, can refer to the number of amino acid residues from the N-terminus of the polypeptide to the most N-terminal amino acid residue of the barcode fragment, or from the C-terminus of the polypeptide to the most C-terminal amino acid residue of the barcode fragment. In some embodiments, for a barcoded XTEN fused to a biologically-active polypeptide, at least one barcode fragment (or at least two barcode fragments, or three barcode fragments) contained in the barcoded XTEN is positioned at least 50, 75, 100, 125, 150, 175, 200, 225, 250, 275, 300 amino acids from the biologically active polypeptide. In some embodiments, the barcode fragment is at least 4, at least 5, at least 6, at least 7, or at least 8 amino acids in length. In some embodiments, the barcode fragment is at least 4 amino acids in length. In some embodiments, the barcode fragment is 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25 amino acids in length, or in a range between any of the foregoing values. In some embodiments, the barcode fragment is between 4 and 20, between 5 and 15, between 6 and 12, or between 7 and 10 amino acids in length. In some embodiments, the barcode fragment comprises an amino acid sequence identified herein by **SEQ ID NOs: 68-77** in **Table 2**.

Table 2. Exemplary Barcode Fragments Releasable Upon Glu-C Digest

Amino Acid Sequence	SEQ ID NO:
SPATSGSTPE	**68 (BAR001)**
GSAPATSE	**69 (BAR002)**
GSAPGTATE	**70 (BAR003)**
GSAPGTE	**71 (BAR004)**
PATSGPTE	**72 (BAR005)**
SASPE	**73 (BAR006)**
PATSGSTE	**74 (BAR007)**
GSAPGTSAE	**75 (BAR008)**
SATSGSE	**76 (BAR009)**
SGPGSTPAE	**77 (BAR010)**

[0119] In some embodiments of the polypeptides of this disclosure, the XTEN can have a length defined by a proximal end and a distal end, where (1) the proximal end is positioned, relative to the distal end, closer to the bispecific antibody construct (**BsAb**), and where (2) the barcode fragment (**BAR**) can be positioned within a region of the XTEN that extends, as measured from the distal end, between 5% and 50%, between 7% and 40%, or between 10% and 30% of the length of the XTEN.

[0120] In some embodiments of the polypeptides of this disclosure, the XTEN further comprises additional one or more barcode fragments, wherein said additional one or more barcode fragments each differ in sequence and molecular weight from all other peptides fragments that are releasable from said polypeptide upon complete digestion of said polypeptide by said protease. In some embodiments, a barcoded XTEN comprises only one barcode fragment. In some embodiments, a barcoded XTEN comprises a set of barcode fragments, comprising a first barcode fragment, such as those described above or anywhere else herein. In some embodiments, the set of barcode fragments comprises a second barcode fragment (or a further barcode fragment), such as those described above or anywhere else herein. In some embodiments, the set of barcode fragments comprises a third barcode fragment, such as those described above or anywhere else herein. The set of barcode fragments fused within an N-terminal XTEN can be referred to as an N-terminal set of barcodes ("an N-terminal set"). The set of barcode fragments fused within a C-terminal XTEN can be referred to as a C-terminal set of barcodes ("a C-terminal set"). In some embodiments, the N-terminal set comprises a first barcode fragment and a second barcode fragment. In some embodiments, the N-terminal set further comprises a third barcode fragment. In some embodiments, the C-terminal set comprises a first barcode fragment and a second barcode fragment. In some embodiments, the C-terminal set further comprises a third barcode fragment. In some embodiments, the second barcode fragment is positioned N-terminal to the first barcode fragment of the same set. In some embodiments, the second barcode fragment is positioned C-terminal to the first barcode fragment of the same set. In some embodiments, the third barcode fragment is positioned N-terminal to both the first and second barcode fragments. In some embodiments, the third barcode fragment is positioned C-terminal to both the first and second barcode fragments. In some embodiments, the third barcode fragment is positioned between the first and second barcode fragments. In some embodiments, the polypeptide comprises a set of barcode fragments that includes a first barcode fragment, a further (second) barcode fragment, and at least one additional barcode fragment, wherein each barcode fragment of the set of barcode fragments (1) is a portion of the second XTEN and (2) differs in sequence and molecular weight from all other peptides fragments that are releasable from the polypeptide upon complete digestion of the polypeptide by the protease.

EXEMPLARY BARCODED XTEN

[0121] Amino acid sequences of 13 exemplary barcoded XTENs, containing one barcode (e.g., **SEQ ID NOs: 8002-8003, 8005-8009, and 8013**), or two barcodes (e.g., **SEQ ID NOS: 8001, 8004, and 8012**), or three barcodes (e.g., **SEQ ID NO: 8011**), are illustrated in **Table 3a**. Among these 13 exemplary barcoded XTEN, six (**SEQ ID NOs: 8001-8003, 8008-8009, and 8011**) are to be fused to a biologically-active protein at the C-terminal of the biologically-active protein, and seven (**SEQ ID NOS: 8004-8007, 8010, and 8012-8013**) are to be fused at the N-terminal of the biologically-active protein. In some embodiments, the XTEN has at least 90%,

at least 92%, at least 95%, at least 98%, at least 99% or 100% sequence identity to a sequence identified herein by **SEQ ID NOs: 8001-8020** in **Table 3a**.

Table 3a. Exemplary Barcoded XTENs

SEQ ID NO.	XTEN Type	# of Barcode(s)	Amino Acid Sequence	Total # of AAs
8001	C-terminal XTEN	2	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGS PTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPG SEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATP ESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTS TEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGS APGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTE PSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEE GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGT STEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESG PGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESA TPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPG TSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGS APGTSESATPESGPGTSESATPESGPGfiabTSESATPESGPGS E**PATSGPTE**SGSEPATSGSETPGSPAGSPTSTEEGTSTEPSE **GSAPGTE**STPSEGSAPGSEPATSGSETPGTSESATPESGPGT STEPSEGSAPGEPEA	864
8002	C-terminal XTEN	1	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGS PTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPG SEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATP ESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTS TEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGS APGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTE PSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEE GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGT STEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESG PGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESA TPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPG TSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGS APGTSESATPESGPGTSESATPESGPGTSESATPESGPGSE**PA TSGPTE**SGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAP GTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPS EGSAPGEPEA	864
8003	C-terminal XTEN	1	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGS PTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPG SEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATP ESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTS TEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGS	864

SEQ ID NO.	XTEN Type	# of Barcode(s)	Amino Acid Sequence	Total # of AAs
			APGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTE PSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEE GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGT STEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESG PGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESA TPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPG TSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGS APGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPA TSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSE**GSAP GTE**STPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPS EGSAPGEPEA	
8004	N-terminal XTEN	2	ASSPAGSPTSTESGTSESATPESGPGTETEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPE**SGPGSTPAE**SGSETP GTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESAT PESGPGE**SPATSGSTPE**GTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPE SGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPA GSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSAP	288
8005	N-terminal XTEN	1	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPG TSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATP ESGPGE**SPATSGSTPE**GTSESATPESGPGSPAGSPTSTEEGSP AGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAG SPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSAP	288
8006	N-terminal XTEN	1	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPE**SGPGSTPAE**SGSETP GTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESAT PESGPGEEPATSGSTPEGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPE SGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPA GSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSAP	288
8007	N-terminal XTEN	1	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPE**SGPGSTPAE**SGSETP GTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESAT PESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPE SGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPA GSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSAP	288
8008	C-terminal XTEN	1	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGS PTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPG SEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATP ESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTS TEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGS APGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTE PSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEE GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPS	864

SEQ ID NO.	XTEN Type	# of Barcode(s)	Amino Acid Sequence	Total # of AAs
			EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGT STEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESG PGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESA TPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPG TSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGS APGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPA TSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSE**GSAP GTE**STPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPS EGSAPG	
8009	C-terminal XTEN	1	PGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESA TPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPT STEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGS APGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAG SPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGS EPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTS TEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSE SATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTE EGTSTEPSE**GSAPGTE**STPSEGSAPGSEPATSGSETPGTSES ATPESGPGTSTEPSEGSAPG	576
8010	N-terminal XTEN	2	SAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGS PAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPE **SGPGSTPAE**SGSETPGSEPATSGSETPGSPAGSPTSTEEGTS ESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTST EEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTE PSEGSAPGTSESATPESGPGSE**PATSGSTE**TPGTSTEPSEGSA PGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGS PTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTS TEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSES ATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEE GTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATS GSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGT STEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTST EPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESG PGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEP SEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPG TSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSE GSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTS ESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTST EEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAG SPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGP	1152

SEQ ID NO.	XTEN Type	# of Barcode(s)	Amino Acid Sequence	Total # of AAs
			GTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTESAS	
8011	C-terminal XTEN	3	SAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGS PAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPE SGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSE SATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTE EGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPG TSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPT STEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESAT PESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGT SESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEP SEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPG SEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSE GSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGS APGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSES ATPESGPGSEPATSGSETPGSE**PATSGSTE**TPGSPAGSPTSTE EGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGS PTSTEEGTSTEPSE**GSAPGTATE**SPEGSAPGTSESATPESGP GTSTEPSE**GSAPGTSAE**SATPESGPGSEPATSGSETPGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGTESAS	1152
8012	N-terminal XTEN	2	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSP TSTEEGTSTEPSEGSAPGTSTEPSE**GSAPATSE**SATPESGPGS EPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSE**SASPE** SGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTST EPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSA PGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEG TSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTS TEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPA TSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESAT PESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGT SESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEG SAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSE SATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSA PGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPAT SGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPG TSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAP	864
8013	N-terminal XTEN	1	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSP TSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGS	864

SEQ ID NO.	XTEN Type	# of Barcode(s)	Amino Acid Sequence	Total # of AAs
			ESATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPE SGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTST EPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSA PGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEG TSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTS TEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPA TSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESAT PESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGT SESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEG SAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSE SATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSA PGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPAT SGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPG TSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAP	
8014	N-terminal XTEN	1	SPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATP ESGPGSEPATSGSETPGTSESATPESGPGSTPAESGSETPGT SESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPE SGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPA GSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESG PGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGS PTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSAP	292
8015	C-terminal XTEN	1	PGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESA TPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPT STEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGS APGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAG SPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGS EPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTS TEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSE SATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTE EGTSTEPSEGSAPGTESTPSEGSAPGSEPATSGSETPGTSES ATPESGPGTSTEPSEGSAPGEPEA	582
8016	C-terminal XTEN	1	TPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPG SEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTS TEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEE GSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPS EGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS EPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTS TEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPA GSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESG PGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSESAT	576

SEQ ID NO.	XTEN Type	# of Barcode(s)	Amino Acid Sequence	Total # of AAs
			SGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPG SEPATSGSETPGTSESA	
8017	C-terminal XTEN	1	GTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATS GSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGT STEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEG SAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEP ATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESG PGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSE GSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSE PATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGTSTEPSEGSAPGTSESASPESGPGSPAGSPTSTEEGSPAG SPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATS GSETPGTSESATPESGP	576
8018	C-terminal XTEN	1	GSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGT STEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEG SAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEP ATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESG PGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSE GSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSE PATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAG SPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEG TSESATPESGPGSEPATS	576
8019	C-terminal XTEN	1	EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGT SESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEG SAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSE SATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSA PGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEP SEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPG TSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSP AGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPES GPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGSEPATSGSETPGTSESASPESGPGTSTEPSEGSAP GSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGT SESATPESGPGTSESAT	576
8020	N-terminal XTEN	1	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSTPAESGSETPG TSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATP ESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSP AGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAG SPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSAP	294

[0122] In some embodiments, a barcoded XTEN can be obtained by making one or more mutations to a general-purpose XTEN, such as any listed in **Table 3b**, according to one or more of the following criteria: to minimize the sequence change in the XTEN, to minimize the amino acid composition change in the XTEN, to substantially maintain the net charge of the XTEN, to substantially maintain (or improve) low immunogenicity of the XTEN, and to substantially maintain (or improve) the pharmacokinetic properties of the XTEN. In some embodiments, the XTEN sequence has at least 90%, at least 92%, at least 95%, at least 98%, at least 99%, or 100% sequence identity to any one of **SEQ ID NOs: 601-659** listed in **Table 3b**. In some embodiments, the XTEN sequence, having at least 90% (e.g., at least 92%, at least 95%, at least 98%, or at least 99%) but less than 100% sequence identity to any of **SEQ ID NOs: 601-659** listed in **Table 3b**, is obtained by one or more mutations (e.g., less than 10, less than 8, less than 6, less than 5, less than 4, less than 3, less than 2 mutations) of the corresponding sequence from **Table 3b**. In some embodiments, the one or more mutations comprise deletion of a glutamic acid residue, insertion of a glutamic acid residue, substitution of a glutamic acid residue, or substitution for a glutamic acid residue, or any combination thereof. In some embodiments, where the XTEN sequence differs from, but has at least 90% (e.g., at least 92%, at least 95%, at least 98%, or at least 99%) sequence identity to, any one of **SEQ ID NOs: 601-659** listed in **Table 3b**, at least 80%, at least 90%, at least 95%, at least 97%, or about 100% of the difference between the XTEN sequence and the corresponding sequence of **Table 3b** involve deletion of a glutamic acid residue, insertion of a glutamic acid residue, substitution of a glutamic acid residue, or substitution for a glutamic acid residue, or any combination thereof. In some such embodiments, at least 80%, at least 90%, at least 95%, at least 97%, or about 100% of the difference between the XTEN sequence and the corresponding sequence of **Table 3b** involve a substitution of a glutamic acid residue, or a substitution for a glutamic acid residue, or both. The term "a substitution of a first amino acid," as used herein, refers to replacement of the first amino acid residue for a second amino acid residue, resulting in the second amino acid residue taking place at the substitution position in the obtained sequence. For example, "a substitution of glutamic acid" refers to replacement of the glutamic acid (E) residue for a non-glutamic acid residue (e.g., serine (S)). The term "a substitution for a first amino acid," as used herein, refers to replacement of a second amino acid residue for the first amino acid residue, resulting in the first amino acid residue taking place at the substitution position in the obtained sequence. For example, "a substitution for glutamic acid" refers to replacement of a non-glutamic acid residue (e.g., serine (S)) for a glutamic acid residue.

Table 3b. Exemplary General-Purpose XTEN for Engineering into Barcoded XTEN(s)

XTEN Name	Amino Acid Sequence	SEQ ID NO
AE144	GSEPATSGSETPGTSESATPESGPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGSEPATSGSETPGSEPATSGSETPGSEPATSGSETPGTSTEPSEGSAPGTSESATP ESGPGSEPATSGSETPGTSTEPSEGSAP	601

XTEN Name	Amino Acid Sequence	SEQ ID NO
AE144_1A	SPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAP GTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTST EEGTSESATPESGPGTSTEPSEGSAPG	602
AE144_2A	TSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGP GTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGS APGTSESATPESGPGTSESATPESGPG	603
AE144_2B	TSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGP GTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGS APGTSESATPESGPGTSESATPESGPG	604
AE144_3A	SPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGSPAGSPTSTEEGTSTEPSEGSAPG	605
AE144_3B	SPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGSPAGSPTSTEEGTSTEPSEGSAPG	606
AE144_4A	TSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTST EEGTSESATPESGPGTSTEPSEGSAPG	607
AE144_4B	TSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTST EEGTSESATPESGPGTSTEPSEGSAPG	608
AE144_5A	TSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSPAGSPTSTEEGSPAGSPTSTEEG	609
AE144_6B	TSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETP GSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSE TPGTSESATPESGPGTSTEPSEGSAPG	610
AE288_1	GTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATP ESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESA TPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTST EPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP	611
AE288_2	GSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGS APGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSE GSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPA GSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAP	612
AE576	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAP	613
AE624	MAEPAGSPTSTEEGTPGSGTASSSPGSSTPSGATGSPGASPGTSSTGSPGSPAGSPTS TEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSE GSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESA TPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTST EPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGT	614

XTEN Name	Amino Acid Sequence	SEQ ID NO
	STEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEE GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGS APGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPT STEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPA GSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAP	
AE864	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS PAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP	615
AE865	GGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEG SAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSP TSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEP SEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEP ATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGS PAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATP ESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGS PTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSES ATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPG SPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP	616
AE866	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS PAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG	617

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AE1152	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS PAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATP ESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPS EGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAG SPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSE SATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGT STEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP	618
AE144A	STEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPES GPGSPAGSPTSTEEGSPAGSPTSTEEGS	619
AE144B	SEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP GSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTST EEGSPAGSPTSTEEGTSTEPSEGSAPG	620
AE180A	TSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAG SPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEP ATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGS EPATS	621
AE216A	PESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTS ESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEG TSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESAT	622
AE252A	ESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTST EPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGS EPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETP GTSESATPESGPGTSTEPSE	623
AE288A	TPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGS EPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAP GTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSE TPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESA	624
AE324A	PESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSE SATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGT SESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGP GTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTST EEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATS	625
AE360A	PESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAG SPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSE	626

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	SATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGT SESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEE GTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSE TPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSG SETPGTSESAT	
AE396A	PESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAG SPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSE SATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGT STEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSE GSAPGTSTEPS	627
AE432A	EGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSP AGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPG TSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGSEPATS	628
AE468A	EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSES ATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEG TSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPES GPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSE GSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPAT SGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSES AT	629
AE504A	EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAG SPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGS APGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESA TPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTST EPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPS	630
AE540A	TPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGT STEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPES GPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSE GSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESA TPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTS ESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEG TSTEPSEGSAPGTSTEP	631
AE576A	TPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTS TEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPG SEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP	632

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	GTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPES GPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATS GSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSES ATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSP AGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESA	
AE612A	GSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAG SPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTST EPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTST EEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATP ESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPS EGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAG SPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSE SATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGT STEPSEGSAPGSEPATSGSETPGTSESAT	633
AE648A	PESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGT STEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSES ATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTS TEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPG SEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETP GTSESAT	634
AE684A	EGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTE PSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTS ESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPG TSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESA TPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGS EPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAP GTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSE TPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATS	635
AE720A	TSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTS TEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPG TSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAP GTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSE TPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGT SESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPES GPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSE	636

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	GSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPAT SGSETPGSPAGSPTSTEEGTSTE	
AE756A	TSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTS TEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPG TSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAP GTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSE TPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGT SESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPES GPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSE GSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPAT SGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSES	637
AE792A	EGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSES ATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTS TEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPG TSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEE GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGS APGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPT STEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPA GSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGS EPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP GSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTST EEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATP ESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGSEPATSGSETPGTSESATPESGPGTSTEPS	638
AE828A	PESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSES ATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTS TEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPG TSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAP GTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSE SATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGS PAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTST EEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPT STEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPAT SGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEP ATSGSETPGTSESAT	639
AE869	GSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSE GSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGS PTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSE PATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPG SPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATP	640

XTEN Name	Amino Acid Sequence	SEQ ID NO
	ESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGS PTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSES ATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPG SPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGR	
AE144_R1	SAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSP AGSPTSTEEGTSESATPESGPGTESASR	641
AE288_R1	SAGSPTGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGT STEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSE GSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPSASR	642
AE432_R1	SAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSP AGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEG TSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEP SEGSAPGSPAGSPTSTEEGTESASR	643
AE576_R1	SAGSPTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGT STEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS PAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPSASR	644
AE864_R1	SAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSP AGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEG TSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEP SEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSE SATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGS PAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATP ESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESA TPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTST EPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTESASR	645
AE712	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA	646

XTEN Name	Amino Acid Sequence	SEQ ID NO
	TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS PAGSPTSTEAHHH	
AE864_R2	GSPGAGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSP AGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEG TSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEP SEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSE SATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGS PAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATP ESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESA TPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTST EPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTESASR	647
AE288_3	SPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTST EEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPT STEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPAT SGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPG	648
AE284	GTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATP ESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESA TPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTST EPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSE	649
AE292	SPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTST EEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPT STEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPAT SGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSA P	650
AE864_2	AGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPG TSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEE GTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSG SETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGS PTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTE PSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSP AGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPG SEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEE GSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGS	651

XTEN Name	Amino Acid Sequence	SEQ ID NO
	PTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSES ATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTS TEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGAAEPEA	
AE867	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS PAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGAAEPEA	652
AE867_2	SPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEG SAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSP TSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEP SEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEP ATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGS PAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATP ESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGS PTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSES ATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPG SPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG	653
AE868	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS PAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGAAEPEA	654
AE144_7A	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAP	655

XTEN Name	Amino Acid Sequence	SEQ ID NO
AE292	SPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTST EEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPT STEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPAT SGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSA P	656
AE293	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPEGAAEPE A	657
AE300	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGAAEPEA	658
AE584	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGS APGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPA TSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGAAEPE A	659

[0123] In some embodiments, for constructing the sequence of a barcoded XTEN, amino-acid mutations are performed on XTEN of intermediate lengths to those of **Table 3b**, as well as XTEN of longer lengths than those of **Table 3b**, such as those in which one or more 12-mer motifs of **Table 1** are added to the N- or C- terminus of a general-purpose XTEN of **Table 3b**.

[0124] Additional examples of general-purpose XTEN sequences that can be used according to the present disclosure are disclosed in U.S. Patent Publication Nos. 2010/0239554 A1, 2010/0323956 A1, 2011/0046060 A1, 2011/0046061 A1, 2011/0077199 A1, or 2011/0172146 A1, or International Patent Publication Nos. WO 2010091122 A1, WO 2010144502 A2, WO 2010144508 A1, WO 2011028228 A1, WO 2011028229 A1, WO 2011028344 A2, WO 2014/011819 A2, or WO 2015/023891.

[0125] In some embodiments, a barcoded XTEN fused within a polypeptide chain adjacent to the N-terminus of the polypeptide chain ("N-terminal XTEN") can be attached to a His tag of HHHHHH (SEQ ID NO: 48) or HHHHHHHH (SEQ ID NO: 49) at the N-terminus to facilitate the purification of the fusion polypeptide. In some embodiments, a barcoded XTEN fused within a polypeptide chain at the C-terminus of the polypeptide chain ("C-terminal XTEN") can be comprise or be attached to the sequence EPEA at the C-terminus to facilitate

the purification of the fusion polypeptide. In some embodiment, the fusion polypeptide comprises both an N-terminal barcoded XTEN and a C-terminal barcoded XTEN, wherein the N-terminal barcoded XTEN is attached to a His tag of HHHHHH (SEQ ID NO: 48) or HHHHHHHH (SEQ ID NO: 49) at the N-terminus; and wherein the C-terminal barcoded XTEN is attached to the sequence EPEA at the C-terminus, thereby facilitating purification of the fusion polypeptide, for example, to at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or at least 99% purity by chromatography methods known in the art, including but not limited to IMAC chromatography, C-tagXL affinity matrix, and other such methods, including but not limited to those described in the EXAMPLES section below.

PROTEASE DIGESTION

[0126] A barcode fragment, as described above or anywhere else herein, can be cleavably fused within the XTEN and releasable (configured to be released) from the XTEN upon digestion of the polypeptide by a protease. In some embodiments, the protease is a Glu-C protease. In some embodiments, the protease cleaves on the C-terminal side of glutamic acid residues that are not followed by proline. One of ordinary skill in the art will understand that a barcoded XTEN (an XTEN that contains barcode fragment(s) therewithin) is designed to achieve high efficiency, precision and accuracy of the protease digestion. For example, one of ordinary skill in the art will understand that adjacent Glu-Glu (EE) residues in an XTEN sequence can result in varying cleavage patterns upon Glu-C digestion. Accordingly, when Glu-C protease is used for barcode release, the barcoded XTEN or the barcode fragment(s) may not contain any Glu-Glu (EE) sequence. One of ordinary skill in the art will also understand that a di-peptide Glu-Pro (EP) sequence, if present in the fusion polypeptide, may not be cleaved by Glu-C protease during the barcode release process.

STRUCTURAL CONFIGURATION OF BPXTEN

[0127] In some embodiments, a BPXTEN fusion protein comprises a single BP and a single XTEN. Such BPXTEN can have at least the following permutations of configurations, each listed in an N- to C-terminus orientation: BP-XTEN; XTEN-BP; BP-S-XTEN; and XTEN-S-BP.

[0128] In some embodiments, the BPXTEN comprises a C-terminal XTEN and, optionally, a spacer sequence (S) (such as one described herein, e.g., in Table C) between the XTEN and the BP. Such BPXTEN can be represented by **Formula I** (depicted N- to C-terminus):

$$(BP)\text{-}(S)_x\text{-}(XTEN) \qquad (I),$$

wherein BP is a biologically active protein as described hereinbelow; S is a spacer sequence (such as one described herein, e.g., in Table C) having between 1 to about 50 amino acid residues that can optionally include a BP release segment (as described more fully hereinbelow); x is either 0 or 1; and XTEN can be any one as described hereinabove or anywhere else herein.

[0129] In some embodiments, the BPXTEN comprises an N-terminal XTEN and, optionally, a spacer sequence (S) (such as one described herein, e.g., in Table C) between the XTEN and the BP. Such BPXTEN can be represented by **Formula II** (depicted N- to C-terminus):

$$(XTEN)\text{-}(S)_x\text{-}(BP) \qquad (II),$$

wherein BP is a biologically active protein as described hereinbelow; S is a spacer sequence (such as one described herein, e.g., in Table C) having between 1 to about 50 amino acid residues that can optionally include a BP release segment (as described more fully hereinbelow); x is either 0 or 1; and XTEN can be any one as described hereinabove or anywhere else herein.

[0130] In some embodiment, the BPXTEN comprises both an N-terminal XTEN and a C-terminal XTEN. Such BPXTEN (e.g., the XPATs in Figures 1-2) can be represented by **Formula III**:

$$(XTEN)\text{-}(S)_y\text{-}(BP)\text{-}(S)_z\text{-}(XTEN) \qquad (III)$$

wherein BP is a biologically active protein as described hereinbelow; S is a spacer sequence (such as one described herein, e.g., in Table C) having between 1 to about 50 amino acid residues that can optionally include a BP release segment (as described more fully hereinbelow); y is either 0 or 1; z is either 0 or 1; and XTEN can be any one as described hereinabove or anywhere else herein.

[0131]

BIOLOGICALLY ACTIVE POLYPEPTIDE

[0132] A biologically active protein (BP) fused to XTEN (as described herein above or described anywhere else herein), particularly those disclosed hereinbelow, comprising sequences identified herein by those of **Tables 6a-6f**, together with their corresponding nucleic acid and amino acid sequences, are well known in the art. Descriptions and sequences of these BP are available in public databases such as Chemical Abstracts Services Databases (e.g., the CAS Registry), GenBank, The Universal Protein Resource (UniProt) and subscription provided databases such as GenSeq (*e.g.*, Derwent). Polynucleotide sequences may be a wild type polynucleotide sequence encoding a given BP (*e.g.*, either full length or mature), or in some instances the sequence may be a variant of the wild type polynucleotide sequence (*e.g.*, a polynucleotide which encodes the wild type biologically active protein), wherein the DNA sequence of the polynucleotide has been optimized, for example, for expression in a particular species; or a polynucleotide encoding a variant of the wild type protein, such as a site directed mutant or an allelic variant. It is well within the ability of the skilled artisan to use a wild-type or consensus cDNA sequence or a codon-optimized variant of a BP to create BPXTEN constructs contemplated by the invention using methods known in the art and/or in conjunction with the guidance and methods provided herein.

[0133] The BP for inclusion in the BPXTEN (a fusion polypeptide comprising at least one BP and at least one XTEN) can include any protein of biologic, therapeutic, prophylactic, or diagnostic interest or function, or that

is useful for mediating a biological activity or preventing or ameliorating a disease, disorder or conditions when administered to a subject. Of particular interest are BP for which an increase in a pharmacokinetic parameter, increased solubility, increased stability, masking of activity, or some other enhanced pharmaceutical property is sought, or those BP for which increasing the terminal half-life would improve efficacy, safety, or result in reduce dosing frequency and/or improve patient compliance. Thus, the BPXTEN fusion protein compositions are prepared with various objectives in mind, including improving the therapeutic efficacy of the bioactive compound by, for example, increasing the in vivo exposure or the length that the BPXTEN remains within the therapeutic window when administered to a subject, compared to a BP not linked to XTEN.

[0134] A BP can be a native, full-length protein or can be a fragment or a sequence variant of a biologically active protein that retains at least a portion of the biological activity of the native protein.

[0135] In one embodiment, the BP incorporated into the subject compositions can be a recombinant polypeptide with a sequence corresponding to a protein found in nature. In some embodiments, the BP can be sequence variants, fragments, homologs, and mimetics of a natural sequence that retain at least a portion of the biological activity of the native BP. In non-limiting examples, a BP can be a sequence that exhibits at least about 80% sequence identity, or alternatively 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a protein sequence identified herein. In further non-limiting examples, a BP can be a bispecific sequence comprising a first binding domain and a second binding domain, wherein the first binding domain, having specific binding affinity to a tumor-specific marker or an antigen of a target cell, exhibits at least about 80% sequence identity, or alternatively 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to paired VL and VH sequences of an anti-CD3 antibody identified herein by **Table 6f**; and wherein the second binding domain, having specific binding affinity to an effector cell, exhibits at least about 80% sequence identity, or alternatively 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to paired VL and VH sequences of an anti-target cell antibody identified herein by **Table 6a**. In one embodiment, a BPXTEN fusion protein can comprise a single BP molecule linked to an XTEN. In some embodiments, the BPXTEN can comprise a first BP and a second molecule of the same BP, resulting in a fusion protein comprising the two BP linked to one or more XTEN (for example, two molecules of glucagon, or two molecules of hGH).

[0136] In general, BP will exhibit a binding specificity to a given target (or a given number of targets) or another desired biological characteristic when used in vivo or when utilized in an in vitro assay. For example, the BP can be an agonist, a receptor, a ligand, an antagonist, an enzyme, an antibody (e.g., mono- or bi-specific), or a hormone. Of particular interest are BP used or known to be useful for a disease or disorder wherein the native BP have a relatively short terminal half-life and for which an enhancement of a pharmacokinetic

parameter (which optionally could be released from the fusion protein by cleavage of a spacer sequence) would permit less frequent dosing or an enhanced pharmacologic effect. Also of interest are BP that have a narrow therapeutic window between the minimum effective dose or blood concentration (Cmin) and the maximum tolerated dose or blood concentration (Cmax). In such cases, the linking of the BP to a fusion protein comprising a select XTEN sequence(s) can result in an improvement in these properties, making them more useful as therapeutic or preventive agents compared to BP not linked to XTEN.

[0137] The BP encompassed by the inventive compositions can have utility in the treatment in various therapeutic or disease categories, including but not limited to glucose and insulin disorders, metabolic disorders, cardiovascular diseases, coagulation and bleeding disorders, growth disorders or conditions, endocrine disorders, eye diseases, kidney diseases, liver diseases, tumorigenic conditions, inflammatory conditions, autoimmune conditions, etc.

[0138] "Anti-CD3" means the monoclonal antibody against the T cell surface protein CD3, species and sequence variants, and fragments thereof, including OKT3 (also called muromonab) and humanized anti-CD3 monoclonal antibody (hOKT31(Ala-Ala))(KC Herold et al., New England Journal of Medicine 346:1692-1698. 2002) Anti-CD3 prevents T-cell activation and proliferation by binding the T-cell receptor complex present on all differentiated T cells. Anti-CD3-containing fusion proteins of the invention may find particular use to slow new-onset Type 1 diabetes, including use of the anti-CD3 as a therapeutic effector as well as a targeting moiety for a second therapeutic BP in the BPXTEN composition. The sequences for the variable region and the creation of anti-CD3 have been described in U.S. Patent Nos. 5,885,573 and 6,491,916.

[0139] The BP of the subject compositions are not limited to native, full-length polypeptides, but also include recombinant versions as well as biologically and/or pharmacologically active variants or fragments thereof. For example, it will be appreciated that various amino acid substitutions can be made in the GP to create variants without departing from the spirit of the invention with respect to the biological activity or pharmacologic properties of the BP. Examples of conservative substitutions for amino acids in polypeptide sequences are shown in **Table 5**. However, in embodiments of the BPXTEN in which the sequence identity of the BP is less than 100% compared to a specific sequence disclosed herein, the invention contemplates substitution of any of the other 19 natural L-amino acids for a given amino acid residue of the given BP, which may be at any position within the sequence of the BP, including adjacent amino acid residues. If any one substitution results in an undesirable change in biological activity, then one of the alternative amino acids can be employed and the construct evaluated by the methods described herein, or using any of the techniques and guidelines for conservative and non-conservative mutations set forth, for instance, in U.S. Pat. No. 5,364,934, the contents of which is incorporated by reference in its entirety, or using methods generally known to those of skill in the art. In addition, variants can also include, for instance, polypeptides wherein one or more amino acid residues are

added or deleted at the N- or C-terminus of the full-length native amino acid sequence of a BP that retains at least a portion of the biological activity of the native peptide.

[0140]

Table 5: Exemplary conservative amino acid substitutions

Original Residue	Exemplary Substitutions
Ala (A)	val; leu; ile
Arg (R)	lys; gin; asn
Asn (N)	gin; his; lys; arg
Asp (D)	glu
Cys (C)	ser
Gln (Q)	asn
Glu (E)	asp
Gly (G)	pro
His (H)	asn; gin; lys; arg
xIle (I)	leu; val; met; ala; phe; norleucine
Leu (L)	norleucine; ile; val; met; ala; phe
Lys (K)	arg; gin; asn
Met (M)	leu; phe; ile
Phe (F)	leu; val; ile; ala
Pro (P)	gly
Ser (S)	thr
Thr (T)	ser
Trp (W)	tyr
Tyr(Y)	trp; phe; thr; ser
Val (V)	ile; leu; met; phe; ala; norleucine

[0141] In some embodiments, a BP incorporated into a BPXTEN fusion protein can have a sequence that exhibits at least about 80% sequence identity to a sequence, alternatively at least about 81%, or about 82%, or about 83%, or about 84%, or about 85%, or about 86%, or about 87%, or about 88%, or about 89%, or about 90%, or about 91%, or about 92%, or about 93%, or about 94%, or about 95%, or about 96%, or about 97%, or about 98%, or about 99%, or 100% sequence identity. In some embodiments, a BP incorporated into a BPXTEN can be a bispecific sequence comprising a first binding domain and a second binding domain, wherein the first binding domain, having specific binding affinity to a tumor-specific marker or an antigen of a target cell, exhibits at least about 80% sequence identity, or alternatively 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to paired VL and VH sequences of an anti-CD3 antibody identified herein by **Table 6f**; and wherein the second binding domain, having specific binding affinity to an effector cell, exhibits at least about 80% sequence identity, or alternatively 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to paired VL and VH sequences of an anti-target cell antibody identified herein by **Table 6a**. The BP of the foregoing embodiments can be evaluated for activity using assays or measured or determined parameters as described herein, and those sequences that retain at least about 40%, or about 50%,

or about 55%, or about 60%, or about 70%, or about 80%, or about 90%, or about 95% or more activity compared to the corresponding native BP sequence would be considered suitable for inclusion in the subject BPXTEN. The BP found to retain a suitable level of activity can be linked to one or more XTEN polypeptides described hereinabove or anywhere else herein. In one embodiment, a BP found to retain a suitable level of activity can be linked to one or more XTEN polypeptides, having at least about 80% sequence identity (e.g., at least about 81%, at least about 82%, at least about 83%, at least about 84%, at least about 85%, at least about 86%, at least about 87%, at least about 88%, at least about 89%, at least about 90%, at least about 91%, at least about 92%, at least about 93%, at least about 94%, at least about 95%, at least about 96%, at least about 97%, at least about 98%, at least about 99%, or 100% sequence identity) to a sequence from **Tables 3a-3b**, resulting in a chimeric fusion protein.

T CELL ENGAGERS

[0142] Additional structural configuration formulae of BPXTEN relate to XTENylated Protease-Activated T Cell Engagers ("XPAT" or "XPATs"), wherein BP is a bispecific antibody (e.g., a bispecific T-cell engager). In some embodiments, the XPAT composition comprises (1) a first portion comprising a first binding domain and a second binding domain, and (2) a second portion comprising the release segment, and (3) a third portion comprising the bulking moiety. In some embodiments, the XPAT composition has the configuration of **Formula Ia** (depicted N-terminus to C-terminus):

(first portion)-(second portion)-(third portion) **(Ia)**

wherein first portion is a bispecific comprising two scFv wherein the first binding domain has specific binding affinity to a tumor-specific marker or an antigen of a target cell and the second binding domain has specific binding affinity to an effector cell; the second portion comprises a release segment (RS) capable of being cleaved by a mammalian protease (as described more fully hereinbelow, the protease can be tumor- or antigen-specific, thereby activation); and the third portion is a bulking moiety. In the foregoing embodiment, the first portion binding domains can be in the order (VL-VH)1-(VL-VH)2, wherein "1" and "2" represent the first and second binding domains, respectively, or (VL-VH)1-(VH-VL)2, or (VH-VL)1-(VL-VH)2, or (VH-VL)1-(VH-VL)2, wherein the paired binding domains are linked by a polypeptide linker (as described more fully hereinbelow). In one embodiment, the first portion VL and VH are set forth in **Tables 6a-6f**; RS is identified herein by the group of sequences set forth in **Tables 8a-8b** (as described more fully hereinbelow); and the bulking moiety is XTEN; albumin binding domain; albumin; IgG binding domain; polypeptides consisting of proline, serine, and alanine; fatty acid; Fc domain; polyethylene glycol (PEG), PLGA; or hydoxylethyl starch. Where desired, the bulking moiety is an XTEN having at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence comprising the group of

sequences set forth in **Tables 3a-3b**. In the foregoing embodiments, the composition is a recombinant fusion protein. In some embodiments, the portions are linked by chemical conjugation.

[0143] In some embodiments, the XPAT composition has the configuration of **Formula IIa** (depicted N-terminus to C-terminus):

(third portion)-(second portion)-(first portion) (**IIa**)

wherein first portion is a bispecific comprising two scFv wherein the first binding domain has specific binding affinity to a tumor-specific marker or an antigen of a target cell and the second binding domain has specific binding affinity to an effector cell; the second portion comprises a release segment (RS) capable of being cleaved by a mammalian protease; and the third portion is a bulking moiety. In the foregoing embodiment, the first portion binding domains can be in the order (VL-VH)1-(VL-VH)2, wherein "1" and "2" represent the first and second binding domains, respectively, or (VL-VH)1-(VH-VL)2, or (VH-VL)1-(VL-VH)2, or (VH-VL)1-(VH-VL)2, wherein the paired binding domains are linked by a polypeptide linker as described herein, below. In one embodiment, the first portion VL and VH are identified in **Tables 6a-6f**; RS is identified herein as the group of sequences set forth in **Tables 8a-8b**; and the bulking moiety is XTEN; albumin binding domain; albumin; IgG binding domain; polypeptides consisting of proline, serine, and alanine; fatty acid; or Fc domain. Where desired, the bulking moiety is an XTEN having at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence comprising the group of sequences set forth in **Tables 3a-3b**. In the foregoing embodiments, the composition is a recombinant fusion protein. In some embodiments, the portions are linked by chemical conjugation.

[0144] In some embodiments, the XPAT composition has the configuration of **Formula IIIa** (depicted N-terminus to C-terminus):

(fifth portion)-(fourth portion)-(first portion)-(second portion)-(third portion) (**IIIa**)

wherein first portion is a bispecific comprising two scFv wherein the first binding domain has specific binding affinity to a tumor-specific marker or an antigen of a target cell and the second binding domain has specific binding affinity to an effector cell; the second portion comprises a release segment (RS) capable of being cleaved by a mammalian protease; the third portion is a bulking moiety; the fourth portion comprises a release segment (RS) capable of being cleaved by a mammalian protease which may be identical or different from the second portion; and the fifth portion is a bulking moiety that may be identical or may be different from the third portion. In the foregoing embodiment, the first portion binding domains can be in the order (VL-VH)1-(VL-VH)2, wherein "1" and "2" represent the first and second binding domains, respectively, or (VL-VH)1-(VH-VL)2, or (VH-VL)1-(VL-VH)2, or (VH-VL)1-(VH-VL)2, wherein the paired binding domains are linked by a polypeptide linker as described herein, below. In the foregoing embodiments, the RS is identified as sequences set forth in **Tables 8a-8b**. In the foregoing embodiments, the bulking moiety is XTEN; albumin

binding domain; albumin; IgG binding domain; polypeptides consisting of proline, serine, and alanine; fatty acid; or Fc domain. Where desired, the bulking moiety is an XTEN having at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence identified herein by the sequences set forth in **Tables 3a-3b**. In the foregoing embodiments, the composition is a recombinant fusion protein. In some embodiments, the portions are linked by chemical conjugation.

[0145] The subject compositions, based on their design and specific components, advantageously provide bispecific therapeutics that have more selectivity, greater half-life, and result in less toxicity and fewer side effects once they are cleaved by proteases found in associated with the target tissues or tissues rendered unhealthy by a disease, such that the subject compositions have improved therapeutic index compared to bispecific antibody compositions known in the art. Such compositions are useful in the treatment of certain diseases, including, but not limited to cancer. It will be appreciated by those of skill in the art that the compositions of the instant invention achieve this reduction in non-specific interactions by a combination of mechanism, which include steric hindrance by locating the binding domains to the bulky XTEN molecules, steric hindrance in that the flexible, unstructured characteristic of the long flexible XTEN polypeptides, by being tethered to the composition, are able to oscillate and move around the binding domains, providing blocking between the composition and tissues or cells, as well as a reduction in the ability of the intact composition to penetrate a cell or tissue due to the large molecular mass (contributed to by both the actual molecular weight of the XTEN and due to the large hydrodynamic radius of the unstructured XTEN) compared to the size of the individual binding domains. However, the compositions are designed such that when in proximity to a target tissue or cell bearing or secreting a protease capable of cleaving the RS, or when internalized into a target cell or tissue when a binding domain has bound the ligand, the bispecific binding domains are liberated from the bulk of the XTEN by the action of the protease(s), removing the steric hindrance barrier, and is freer to exert its pharmacologic effect. The subject compositions find use in the treatment of a variety of conditions where selective delivery of a therapeutic bispecific antibody composition to a cell, tissue or organ is desired. In one embodiment, the target tissue is a cancer, which may be a leukemia, a lymphoma, or a tumor of an organ or system.

Binding Domains

[0146] The disclosure contemplates use of single chain binding domains, such as but not limited to Fv, Fab, Fab', Fab'-SH, F(ab')2, linear antibodies, single domain antibody, single domain camelid antibody, single-chain antibody molecules (scFv), and diabodies capable of binding ligands or receptors associated with effector cells and antigens of diseased tissues or cells that are cancers, tumors, or other malignant tissues. In some embodiments, an antigen binding fragment (**AF**) (e.g., a first antigen binding fragment (AF1), or a second antigen binding fragment (AF2)) can (each independently) be a chimeric or a humanized antigen binding

fragment. The antigen binding fragment (**AF**) (e.g., a first antigen binding fragment (AF1), or a second antigen binding fragment (AF2)) can (each independently) be Fv, Fab, Fab', Fab'-SH, linear antibody, or single-chain variable fragment (scFv). Two antigen binding fragments (*e.g.*, the first and second antigen binding fragments) can be configured as an (Fab')2 or a single chain diabody. In some embodiments, the bispecific comprises a first binding domain with binding specificity to a target cell marker and a second binding domain with binding specificity to an effector cell antigen. In some embodiments, the first and the second binding domains can be non-antibody scaffolds such as anticalins, adnectins, fynomers, affilins, affibodies, centyrins, or DARPins. In other embodiments, the binding domain for the tumor cell target is a variable domain of a T cell receptor that has been engineered to bind MHC that is loaded with a peptide fragment of a protein that is overexpressed by tumor cells. In some embodiments, the XPAT compositions are designed with considerations of the location of the target tissue protease as well as the presence of the same protease in healthy tissues not intended to be targeted, as well as the presence of the target ligand in healthy tissue but a greater presence of the ligand in unhealthy target tissue, in order to provide a wide therapeutic window. A "therapeutic window" refers to the largest difference between the minimal effective dose and the maximal tolerated dose for a given therapeutic composition. To help achieve a wide therapeutic window, the binding domains of the first portion of the compositions are shielded by the proximity of the bulking moiety (e.g., XTEN) such that the binding affinity of the intact composition for one or both of the ligands is reduced compared to the composition that has been cleaved by a mammalian protease, thereby releasing the first portion from the shielding effects of the bulking moiety.

[0147] With respect to single chain binding domains, as is well established, Fv is the minimum antibody fragment which contains a complete antigen recognition and binding site; consisting of a dimer of one heavy (VH) and one light chain variable domain (VL) in non-covalent association. Within each VH and VL chain are three complementarity determining regions (CDRs) that interact to define an antigen binding site on the surface of the VH-VL dimer; the six CDRs of a binding domain confer antigen binding specificity to the antibody or single chain binding domain. In some cases, scFv are created in which each has 3, 4, or 5 CHRs within each binding domain. Framework sequences flanking the CDRs have a tertiary structure that is essentially conserved in native immunoglobulins across species, and the framework residues (FR) serve to hold the CDRs in their appropriate orientation. The constant domains are not required for binding function, but may aid in stabilizing VH-VL interaction. In some embodiments, the domain of the binding site of the polypeptide can be a pair of VH-VL, VH-VH or VL-VL domains either of the same or of different immunoglobulins, however it is generally preferred to make single chain binding domains using the respective VH and VL chains from the parental antibody. The order of VH and VL domains within the polypeptide chain is not limiting for the present invention; the order of domains given may be reversed usually without any loss of function, but it is understood that the

VH and VL domains are arranged so that the antigen binding site can properly fold. Thus, the single chain binding domains of the bispecific scFv embodiments of the subject compositions can be in the order (VL-VH)1-(VL-VH)2, wherein "1" and "2" represent the first and second binding domains, respectively, or (VL-VH)1-(VH-VL)2, or (VH-VL)1-(VL-VH)2, or (VH-VL)1-(VH-VL)2, wherein the paired binding domains are linked by a polypeptide linker as described herein, below.

[0148] The arrangement of the binding domains in an exemplary bispecific single chain antibody disclosed herein may therefore be one in which the first binding domain is located C-terminally to the second binding domain. The arrangement of the V chains may be VH (target cell surface antigen)-VL(target cell surface antigen)-VL(effector cell antigen)-VH(effector cell antigen), VH(target cell surface antigen)-VL(target cell surface antigen)-VH(effector cell antigen)-VL(effector cell antigen), VL(target cell surface antigen)-VH(target cell surface antigen)-VL(effector cell antigen)-VH(effector cell antigen) or VL(target cell surface antigen)-VH(target cell surface antigen)-VH(effector cell antigen)-VL(effector cell antigen). For an arrangement, in which the second binding domain is located N-terminally to the first binding domain, the following orders are possible: VH (effector cell antigen)-VL(effector cell antigen)-VL(target cell surface antigen)-VH(target cell surface antigen), VH(effector cell antigen)-VL(effector cell antigen)-VH(target cell surface antigen)-VL(target cell surface antigen), VL(effector cell antigen)-VH(effector cell antigen)-VL(target cell surface antigen)-VH(target cell surface antigen) or VL(effector cell antigen)-VH(effector cell antigen)-VH(target cell surface antigen)-VL(target cell surface antigen). As used herein, "N-terminally to" or "C-terminally to" and grammatical variants thereof denote relative location within the primary amino acid sequence rather than placement at the absolute N- or C-terminus of the bispecific single chain antibody. Hence, as a non-limiting example, a first binding domain which is "located C-terminally to the second binding domain" denotes that the first binding is located on the carboxyl side of the second binding domain within the bispecific single chain antibody, and does not exclude the possibility that an additional sequence, for example a His-tag, or another compound such as a radioisotope, is located at the C-terminus of the bispecific single chain antibody.

[0149] In one embodiment, the chimeric polypeptide assembly compositions comprise a first portion comprising a first binding domain and a second binding domain wherein each of the binding domains is an scFv and wherein each scFv comprises one VL and one VH. In some embodiments, the chimeric polypeptide assembly compositions comprise a first portion comprising a first binding domain and a second binding domain wherein the binding domains are in a diabody configuration and wherein each domain comprises one VL domain and one VH.

[0150] It is envisaged that the scFv embodiments of the XPAT compositions of the invention comprise a first binding domain and a second binding domain wherein the VL and VH domains are derived from monoclonal antibodies with binding specificity to the tumor-specific marker or an antigen of a target cell and effector cell

antigens, respectively. In other cases, the first and second binding domains each comprise six CDRs derived from monoclonal antibodies with binding specificity to a target cell marker, such as a tumor-specific marker and effector cell antigens, respectively. In other embodiments, the first and second binding domains of the first portion of the subject compositions can have 3, 4, or 5 CHRs within each binding domain. In other embodiments, the embodiments of the invention comprise a first binding domain and a second binding domain wherein each comprises a CDR-H1 region, a CDR-H2 region, a CDR-H3 region, a CDR-L1 region, a CDR-L2 region, and a CDR-H3 region, wherein each of the regions is derived from a monoclonal antibody capable of binding the tumor-specific marker or an antigen of a target cell, and effector cell antigens, respectively. In one embodiment, the invention provides a chimeric polypeptide assembly composition wherein the second binding domain comprises VH and VL regions derived from a monoclonal antibody capable of binding human CD3. In some embodiments, the invention provides a chimeric polypeptide assembly composition, wherein the scFv second binding domain comprises VH and VL regions wherein each VH and VL regions exhibit at least about 90%, or 91%, or 92%, or 93%, or 94%, or 95%, or 96%, or 97%, or 98%, or 99% identity to or is identical to paired VL and VH sequences of an anti-CD3 antibody identified in **Table 6a**. In some embodiments, the second domain embodiments of the invention comprise a CDR-H1 region, a CDR-H2 region, a CDR-H3 region, a CDR-L1 region, a CDR-L2 region, and a CDR-H3 region, wherein each of the regions is derived from a monoclonal antibody identified herein as the antibodies set forth in **Table 6a**. In the foregoing embodiments, the VH and/or VL domains can be configured as scFv, diabodies, a single domain antibody, or a single domain camelid antibody.

[0151] In other embodiments, the second domains of the subject compositions are derived from an anti-CD3 antibody identified herein as the antibodies set forth in **Table 6a**. In one embodiment of the foregoing, the second domain of the subject composition comprises the paired VL and the VH region sequences of the anti-CD3 antibody identified herein as the group of antibodies set forth in **Table 6a**. In some embodiments, the invention provides a chimeric polypeptide assembly composition, wherein the second binding domain comprises VH and VL regions wherein each VH and VL regions exhibit at least about 90%, or 91%, or 92%, or 93%, or 94%, or 95%, or 96%, or 97%, or 98%, or 99% identity to or is identical to paired VL and VH sequences of the huUCHT1 anti-CD3 antibody of **Table 6a**. In the foregoing embodiments, the VH and/or VL domains can be configured as scFv, a portion of a diabody, a single domain antibody, or a single domain camelid antibody.

[0152] In other embodiments, the scFv of the first domain of the composition are derived from an anti-tumor cell antibody identified as the antibodies set forth in **Table 6f**. In some embodiments, the invention provides a chimeric polypeptide assembly composition, wherein the first binding domain comprises VH and VL regions wherein each VH and VL regions exhibit at least about 90%, or 91%, or 92%, or 93%, or 94%, or 95%, or 96%,

or 97%, or 98%, or 99% identity to or is identical to paired VL and VH sequences of an anti-tumor cell antibody identified in **Table 6f**. In one embodiment of the foregoing, the first domain of the recited compositions comprises the paired VL and the VH region sequences of an anti-tumor cell antibody disclosed herein. In the foregoing embodiments, the VH and/or VL domains can be configured as scFv, a portion of a diabody, a single domain antibody, or a single domain camelid antibody.

[0153] In some embodiments, the chimeric polypeptide assembly compositions comprise a first portion comprising a first binding domain and a second binding domain wherein the binding domains are in a diabody configuration and each of the binding domains comprises one VL domain and one VH domain. In one embodiment, the diabody embodiments of the invention comprise a first binding domain and a second binding domain wherein the VL and VH domains are derived from monoclonal antibodies with binding specificity to a tumor-specific marker or an antigen of a target cell, and the effector cell antigen, respectively. In some embodiments, the diabody embodiments of the invention comprise a first binding domain and a second binding domain wherein each comprises a CDR-H1 region, a CDR-H2 region, a CDR-H3 region, a CDR-L1 region, a CDR-L2 region, and a CDR-H3 region, wherein each of the regions is derived from a monoclonal antibody capable of binding the tumor-specific marker or target cell antigen, and the effector cell antigen, respectively. It is envisaged that the diabody embodiments of the invention comprise a first binding domain and a second binding domain wherein the VL and VH domains are derived from monoclonal antibodies with binding specificity to the tumor-specific marker or target cell antigen, and the effector cell antigen, respectively. In some embodiments, the diabody embodiments of the invention comprise a first binding domain and a second binding domain wherein each comprises a CDR-H1 region, a CDR-H2 region, a CDR-H3 region, a CDR-L1 region, a CDR-L2 region, and a CDR-H3 region, wherein each of the regions is derived from a monoclonal antibody capable of binding the tumor-specific marker or target cell antigen, and the effector cell antigen, respectively. In one embodiment, the invention provides a chimeric polypeptide assembly composition wherein the diabody second binding domain comprises the paired VH and VL regions derived from a monoclonal antibody capable of binding human CD3. In some embodiments, the invention provides a chimeric polypeptide assembly composition, wherein the diabody second binding domain comprises VH and VL regions wherein each VH and VL regions exhibit at least about 90%, or 91%, or 92%, or 93%, or 94%, or 95%, or 96%, or 97%, or 98%, or 99% identity to or is identical to paired VL and VH sequences of an anti-CD3 antibody identified in **Table 6a**. In some embodiments, the invention provides a chimeric polypeptide assembly composition, wherein the diabody second binding domain comprises VH and VL regions wherein each VH and VL regions exhibit at least about 90%, or 91%, or 92%, or 93%, or 94%, or 95%, or 96%, or 97%, or 98%, or 99% identity to or is identical to the VL and a VH sequence of the huUCHT1 antibody of **Table 6a**. In other embodiments, the diabody second domain of the composition is derived from an anti-CD3 antibody described herein. In some

embodiments, the invention provides a chimeric polypeptide assembly composition, wherein the diabody first binding domain comprises VH and VL regions wherein each VH and VL regions exhibit at least about 90%, or 91%, or 92%, or 93%, or 94%, or 95%, or 96%, or 97%, or 98%, or 99% identity to or is identical to VL and VH sequences of an anti-tumor cell antibody identified in **Table 6f**. In other embodiments, the diabody first domain of the composition is derived from an anti-tumor cell antibody described herein.

[0154] Methods to measure binding affinity and/or other biologic activity of the subject compositions of the invention can be those disclosed herein or methods generally known in the art. For example, the binding affinity of a binding pair (e.g., antibody and antigen), denoted as K_d, can be determined using various suitable assays including, but not limited to, radioactive binding assays, non-radioactive binding assays such as fluorescence resonance energy transfer and surface plasmon resonance (SPR, Biacore), and enzyme-linked immunosorbent assays (ELISA), kinetic exclusion assay (KinExA®) or as described in the Examples. An increase or decrease in binding affinity, for example of a chimeric polypeptide assembly which has been cleaved to remove a bulking moiety compared to the chimeric polypeptide assembly with the bulking moiety attached, can be determined by measuring the binding affinity of the chimeric polypeptide assembly to its target binding partner with and without the bulking moiety.

[0155] Measurement of half-life of a subject chimeric assembly can be performed by various suitable methods. For example, the half-life of a substance can be determined by administering the substance to a subject and periodically sampling a biological sample (e.g., biological fluid such as blood or plasma or ascites) to determine the concentration and/or amount of that substance in the sample over time. The concentration of a substance in a biological sample can be determined using various suitable methods, including enzyme-linked immunosorbent assays (ELISA), immunoblots, and chromatography techniques including high-pressure liquid chromatography and fast protein liquid chromatography. In some cases, the substance may be labeled with a detectable tag, such as a radioactive tag or a fluorescence tag, which can be used to determine the concentration of the substance in the sample (e.g., a blood sample or a plasma sample. The various pharmacokinetic parameters are then determined from the results, which can be done using software packages such as SoftMax Pro software, or by manual calculations known in the art.

[0156] In addition, the physicochemical properties of the chimeric polypeptide assembly compositions may be measured to ascertain the degree of solubility, structure and retention of stability. Assays of the subject compositions are conducted that allow determination of binding characteristics of the binding domains towards a ligand, including binding dissociation constant (K_d, K_{on} and K_{off}), the half-life of dissociation of the ligand-receptor complex, as well as the activity of the binding domain to inhibit the biologic activity of the sequestered ligand compared to free ligand (IC_{50} values). The term "IC_{50}" refers to the concentration needed to inhibit half of the maximum biological response of the ligand agonist, and is generally determined by competition binding

assays. The term "EC_{50}" refers to the concentration needed to achieve half of the maximum biological response of the active substance, and is generally determined by ELISA or cell-based assays, including the methods of the Examples described herein.

Anti-CD3 Binding Domains

[0157] In some embodiments, the invention provides chimeric polypeptide assembly compositions comprising a binding domain of the first portion with binding affinity to T cells. In one embodiment, the binding domain of the second portion comprises VL and VH derived from a monoclonal antibody that binds CD3. In some embodiments, the binding domain comprises VL and VH derived from a monoclonal antibody to CD3 epsilon and/or CD3 delta. Exemplary, non-limiting examples of VL and VH sequences of monoclonal antibodies to CD3 are presented in **Table 6a**. In one embodiment, the invention provides a chimeric polypeptide assembly comprising a binding domain with binding affinity to CD3 comprising anti-CD3 VL and VH sequences set forth in **Table 6a**. In some embodiments, the invention provides a chimeric polypeptide assembly comprising a binding domain of the first portion with binding affinity to CD3epsilon comprising anti-CD3epsilon VL and VH sequences set forth in **Table 6a**. In some embodiments, the invention provides a chimeric polypeptide assembly composition, wherein the scFv second binding domain of the first portion comprises VH and VL regions wherein each VH and VL regions exhibit at least about 90%, or 91%, or 92%, or 93%, or 94%, or 95%, or 96%, or 97%, or 98%, or 99% identity to or is identical to paired VL and VH sequences of the huUCHT1 anti-CD3 antibody of **Table 6a**. In some embodiments, the invention provides a chimeric polypeptide assembly composition comprising a binding domain with binding affinity to CD3 comprising the CDR-L1 region, the CDR-L2 region, the CDR-L3 region, the CDR-H1 region, the CDR-H2 region, and the CDR-H3 region, wherein each is derived from the respective anti-CD3 VL and VH sequences set forth in **Table 6a**. In some embodiments, the invention provides a chimeric polypeptide assembly composition comprising a binding domain with binding affinity to CD3 comprising the CDR-L1 region, the CDR-L2 region, the CDR-L3 region, the CDR-H1 region, the CDR-H2 region, and the CDR-H3 region, wherein the CDR sequences are RASQDIRNYLN (SEQ ID NO: 50), YTSRLESQQGNTLPWT (SEQ ID NO: 78), GYSFTGYTMN (SEQ ID NO: 79), LINPYKGVST (SEQ ID NO: 80), and SGYYGDSDWYFDV (SEQ ID NO: 81).

[0158] The CD3 complex is a group of cell surface molecules that associates with the T-cell antigen receptor (TCR) and functions in the cell surface expression of TCR and in the signaling transduction cascade that originates when a peptide:MHC ligand binds to the TCR. Typically, when an antigen binds to the T-cell receptor, the CD3 sends signals through the cell membrane to the cytoplasm inside the T cell. This causes activation of the T cell that rapidly divide to produce new T cells sensitized to attack the particular antigen to which the TCR were exposed. The CD3 complex is comprised of the CD3epsilon molecule, along with four other membrane-bound polypeptides (CD3-gamma, -delta, and/or -zeta). In humans, CD3-epsilon is encoded by the CD3E gene

on Chromosome 11. The intracellular domains of each of the CD3 chains contain immunoreceptor tyrosine-based activation motifs (ITAMs) that serve as the nucleating point for the intracellular signal transduction machinery upon T cell receptor engagement.

[0159] A number of therapeutic strategies modulate T cell immunity by targeting TCR signaling, particularly the anti-human CD3 monoclonal antibodies (mAbs) that are widely used clinically in immunosuppressive regimes. The CD3-specific mouse mAb OKT3 was the first mAb licensed for use in humans (Sgro, C. Side-effects of a monoclonal antibody, muromonab CD3/orthoclone OKT3: bibliographic review. Toxicology 105:23-29, 1995) and is widely used clinically as an immunosuppressive agent in transplantation (Chatenoud, Clin. Transplant 7:422-430, (1993); Chatenoud, Nat. Rev. Immunol. 3:123-132 (2003); Kumar, Transplant. Proc. 30:1351-1352 (1998)), type 1 diabetes, and psoriasis. Importantly, anti-CD3 mAbs can induce partial T cell signaling and clonal anergy (Smith, JA, Nonmitogenic Anti-CD3 Monoclonal Antibodies Deliver a Partial T Cell Receptor Signal and Induce Clonal Anergy J. Exp. Med. 185:1413-1422 (1997)). OKT3 has been described in the literature as a T cell mitogen as well as a potent T cell killer (Wong, JT. The mechanism of anti-CD3 monoclonal antibodies. Mediation of cytolysis by inter-T cell bridging. Transplantation 50:683-689 (1990)). In particular, the studies of Wong demonstrated that by bridging CD3 T cells and target cells, one could achieve killing of the target and that neither FcR-mediated ADCC nor complement fixation was necessary for bivalent anti-CD3 MAB to lyse the target cells.

[0160] OKT3 exhibits both a mitogenic and T-cell killing activity in a time-dependent fashion; following early activation of T cells leading to cytokine release, upon further administration OKT3 later blocks all known T-cell functions. It is due to this later blocking of T cell function that OKT3 has found such wide application as an immunosuppressant in therapy regimens for reduction or even abolition of allograft tissue rejection. Other antibodies specific for the CD3 molecule are disclosed in Tunnacliffe, Int. Immunol. 1 (1989), 546-50, WO2005/118635 and WO2007/033230 describe anti-human monoclonal CD3 epsilon antibodies, United States Patent 5,821,337 describes the VL and VH sequences of murine anti-CD3 monoclonal Ab UCHT1 (muxCD3, Shalaby et al., J. Exp. Med. 175, 217-225 (1992) and a humanized variant of this antibody (hu UCHT1), and United States Patent Application 20120034228 discloses binding domains capable of binding to an epitope of human and non-chimpanzee primate CD3 epsilon chain.

Table 6a: Anti-CD3 Monoclonal Antibodies and Sequences

Clone Name	Antibody Name	Target	VH Sequence	SEQ ID NO.	VL Sequence	SEQ ID NO.
huOKT3		CD3	QVQLVQSGGGVVQP GRSLRLSCKAS**GYT FTRYTMH**WVRQAP GKGLEWIG**YINPSR GYTNYNQKVKD**RF TISRDNSKNTAFLQ	**301**	DIQMTQSPSSLSASV GDRVTITC**SASSSVS YMN**WYQQTPGKAP KRWIY**DTSKLAS**GV PSRFSGSGSGTDYTF TISSLQPEDIATYYC	**351**

Clone Name	Antibody Name	Target	VH Sequence	SEQ ID NO.	VL Sequence	SEQ ID NO.
			MDSLRPEDTGVYFC ARYYDDHYCLDYW GQGTPVTVSS		QQWSSNPFTFGQGT KLQITR	
huUCHT1		CD3	EVQLVESGGGLVQP GGSLRLSCAASGYS FTGYTMNWVRQAP GKGLEWVALINPYK GVSTYNQKFKDRFT ISVDKSKNTAYLQM NSLRAEDTAVYYCA RSGYYGDSDWYFD VWGQGTLVTVSS	302	DIQMTQSPSSLSASV GDRVTITCRASQDIR NYLNWYQQKPGKA PKLLIYYTSRLESGV PSRFSGSGSGTDYTL TISSLQPEDFATYYC QQGNTLPWTFGQG TKVEIK	352
hu12F6		CD3	QVQLVQSGGGVVQP GRSLRLSCKASGYT FTSYTMHWVRQAP GKGLEWIGYINPSS GYTKYNQKFKDRF TISADKSKSTAFLQM DSLRPEDTGVYFCA RWQDYDVYFDYW GQGTPVTVSS	303	DIQMTQSPSSLSASV GDRVTMTCRASSSV SYMHWYQQTPGKA PKPWIYATSNLASG VPSRFSGSGSGTDYT LTISSLQPEDIATYYC QQWSSNPPTFGQGT KLQITR	353
mOKT3		CD3	QVQLQQSGAELARP GASVKMSCKASGY TFTRYTMHWVKQR PGQGLEWIGYINPSR GYTNYNQKFKDKA TLTTDKSSSTAYMQ LSSLTSEDSAVYYCA RYYDDHYCLDYWG QGTTLTVSS	304	QIVLTQSPAIMSASP GEKVTMTCSASSSV SYMNWYQQKSGTS PKRWIYDTSKLASG VPAHFRGSGSGTSYS LTISGMEAEDAATY YCQQWSSNPFTFGS GTKLEINR	354
MT103	blinatumomab	CD3	DIKLQQSGAELARP GASVKMSCKTSGYT FTRYTMHWVKQRP GQGLEWIGYINPSR GYTNYNQKFKDKA TLTTDKSSSTAYMQ LSSLTSEDSAVYYCA RYYDDHYCLDYWG QGTTLTVSS	305	DIQLTQSPAIMSASP GEKVTMTCRASSSV SYMNWYQQKSGTS PKRWIYDTSKVASG VPYRFSGSGSGTSYS LTISSMEAEDAATY YCQQWSSNPLTFG AGTKLELK	355
MT110	solitomab	CD3	DVQLVQSGAEVKKP GASVKVSCKASGYT FTRYTMHWVRQAP GQGLEWIGYINPSR GYTNYADSVKGRF TITTDKSTSTAYMEL SSLRSEDTATYYCA RYYDDHYCLDYWG QGTTVTVSS	306	DIVLTQSPATLSLSP GERATLSCRASQSV SYMNWYQQKPGKA PKRWIYDTSKVASG VPARFSGSGSGTDYS LTINSLEAEDAATYY CQQWSSNPLTFGG GTKVEIK	356
CD3.7		CD3	EVQLVESGGGLVQP GGSLKLSCAASGFTF NKYAMNWVRQAPG KGLEWVARIRSKYN	307	QTVVTQEPSLTVSPG GTVTLTCGSSTGAV TSGYYPNWVQQKP GQAPRGLIGGTKFL	357

Clone Name	Antibody Name	Target	VH Sequence	SEQ ID NO.	VL Sequence	SEQ ID NO.
			NYATYYADSVKDRF TISRDDSKNTAYLQ MNNLKTEDTAVYY CVRHGNFGNSYISY WAYWGQGTLVTVS S		APGTPARFSGSLLGG KAALTLSGVQPEDE AEYYCALWYSNRW VFGGGTKLTVL	
CD3.8		CD3	EVQLVESGGGLVQP GGSLRLSCAASGFTF NTYAMNWVRQAPG KGLEWVGRIRSKYN NYATYYADSVKGRF TISRDDSKNTLYLQ MNSLRAEDTAVYYC VRHGNFGNSYVSWF AYWGQGTLVTVSS	**308**	QAVVTQEPSLTVSP GGTVTLTCGSSTGA VTTSNYANWVQQK PGQAPRGLIGGTNK RAPGVPARFSGSLLG GKAALTLSGAQPED EAEYYCALWYSNL WVFGGGTKLTVL	**358**
CD3.9		CD3	EVQLLESGGGLVQP GGSLKLSCAASGFTF NTYAMNWVRQAPG KGLEWVARIRSKYN NYATYYADSVKDRF TISRDDSKNTAYLQ MNNLKTEDTAVYY CVRHGNFGNSYVS WFAYWGQGTLVTV SS	**309**	ELVVTQEPSLTVSPG GTVTLTCRSSTGAV TTSNYANWVQQKP GQAPRGLIGGTNKR APGTPARFSGSLLGG KAALTLSGVQPEDE AEYYCALWYSNLW VFGGGTKLTVL	**359**
CD3.10		CD3	EVKLLESGGGLVQP KGSLKLSCAASGFTF NTYAMNWVRQAPG KGLEWVARIRSKYN NYATYYADSVKDRF TISRDDSQSILYLQM NNLKTEDTAMYYC VRHGNFGNSYVSWF AYWGQGTLVTVSS	**310**	QAVVTQESALTTSP GETVTLTCRSSTGA VTTSNYANWVQEKP DHLFTGLIGGTNKR APGVPARFSGSLIGD KAALTITGAQTEDE AIYFCALWYSNLWV FGGGTKLTVL	**360**

* underlined sequences, if present, are CDRs within the VL and VH

CD3 CELL ANTIGEN BINDING FRAGMENTS

[0161] In some embodiments, the disclosure relates to antigen binding fragments (AF1) having specific binding affinity for an effector cell antigen that can be incorporated into any of the subject composition embodiments described herein. In some cases, the effector cell antigen is expressed on the surface of an effector cell that is a plasma cell, a T cell, a B cell, a cytokine induced killer cell (CIK cell), a mast cell, a dendritic cell, a regulatory T cell (RegT cell), a helper T cell, a myeloid cell, or a NK cell.

[0162] Various AF1 that bind effector cell antigens have particular utility for pairing with an antigen binding fragment with binding affinity to HER2 antigens associated with a diseased cell or tissue in composition formats in order to effect cell killing of the diseased cell or tissue. Binding specificity can be determined by complementarity determining regions, or CDRs, such as light chain CDRs or heavy chain CDRs. In many cases,

binding specificity is determined by light chain CDRs and heavy chain CDRs. A given combination of heavy chain CDRs and light chain CDRs provides a given binding pocket that confers greater affinity and/or specificity towards an effector cell antigen as compared to other reference antigens. The resulting bispecific compositions, having a first antigen binding fragment (AF1) to HER2 linked by a short, flexible peptide linker to a second antigen binding fragment (AF2) with binding specificity to an effector cell antigen are bispecific, with each antigen binding fragment having specific binding affinity to their respective ligands. It will be understood that in such compositions, an AF2 directed against a HER2 of a disease tissue is used in combination with a AF1 directed towards an effector cell marker in order to bring an effector cell in close proximity to the cell of a disease tissue in order to effect the cytolysis of the cell of the diseased tissue. Further, the AF1 and AF2 are incorporated into the specifically designed polypeptides comprising cleavable release segments and XTEN in order to confer prodrug characteristics on the compositions that becomes activated by release of the fused AF1 and AF2 upon the cleavage of the release segments when in proximity to the disease tissue having proteases capable of cleaving the release segments in one or more locations in the release segment sequence.

[0163] In one embodiment, the AF1 of the subject compositions has binding affinity for an effector cell antigen expressed on the surface of a T cell. In some embodiments, the AF1 of the subject compositions has binding affinity for CD3. In some embodiments, the AF1 of the subject compositions has binding affinity for a member of the CD3 complex, which includes in individual form or independently combined form all known CD3 subunits of the CD3 complex; for example, CD3 epsilon, CD3 delta, CD3 gamma, and CD3 zeta. In some embodiments, the AF1 has binding affinity for CD3 epsilon, CD3 delta, CD3 gamma, or CD3 zeta.

[0164] The origin of the antigen binding fragments contemplated by the disclosure can be derived from a naturally occurring antibody or fragment thereof, a non-naturally occurring antibody or fragment thereof, a humanized antibody or fragment thereof, a synthetic antibody or fragment thereof, a hybrid antibody or fragment thereof, or an engineered antibody or fragment thereof. Methods for generating an antibody for a given target marker are well known in the art. For example, the monoclonal antibodies may be made using the hybridoma method first described by Kohler et al., *Nature*, 256:495 (1975), or may be made by recombinant DNA methods (U.S. Pat. No. 4,816,567). The structure of antibodies and fragments thereof, variable regions of heavy and light chains of an antibody (VH and VL), single chain variable regions (scFv), complementarity determining regions (CDR), and domain antibodies (dAbs) are well understood. Methods for generating a polypeptide having a desired antigen binding fragment with binding affinity to a given antigen are known in the art.

[0165] It will be understood that use of the term "antigen binding fragments" for the composition embodiments disclosed herein is intended to include portions or fragments of antibodies that retain the ability to bind the antigens that are the ligands of the corresponding intact antibody. In such embodiments, the antigen binding

fragment can be, but is not limited to, CDRs and intervening framework regions, variable or hypervariable regions of light and/or heavy chains of an antibody (VL, VH), variable fragments (Fv), Fab' fragments, F(ab')2 fragments, Fab fragments, single chain antibodies (scAb), VHH camelid antibodies, single chain variable fragment (scFv), linear antibodies, a single domain antibody, complementarity determining regions (CDR), domain antibodies (dAbs), single domain heavy chain immunoglobulins of the BHH or BNAR type, single domain light chain immunoglobulins, or other polypeptides known in the art containing a fragment of an antibody capable of binding an antigen. The antigen binding fragments having CDR-H and CDR-L can be configured in a (CDR-H)-(CDR-L) or a (CDR-H)-(CDR-L) orientation, N-terminus to C-terminus. The VL and VH of two antigen binding fragments can also be configured in a single chain diabody configuration; i.e., the VL and VH of the AF1 and AF2 configured with linkers of an appropriate length to permit arrangement as a diabody.

[0166] Various CD3 binding AF1 of the disclosure have been specifically modified to enhance their stability in the polypeptide embodiments described herein. Protein aggregation of antibodies continues to be a significant problem in their developability and remains a major area of focus in antibody production. Antibody aggregation can be triggered by partial unfolding of its domains, leading to monomer-monomer association followed by nucleation and aggregate growth. Although the aggregation propensities of antibodies and antibody-based proteins can be affected by the external experimental conditions, they are strongly dependent on the intrinsic antibody properties as determined by their sequences and structures. Although it is well known that proteins are only marginally stable in their folded states, it is often less well appreciated that most proteins are inherently aggregation-prone in their unfolded or partially unfolded states, and the resulting aggregates can be extremely stable and long-lived. Reduction in aggregation propensity has also been shown to be accompanied by an increase in expression titer, showing that reducing protein aggregation is beneficial throughout the development process and can lead to a more efficient path to clinical studies. For therapeutic proteins, aggregates are a significant risk factor for deleterious immune responses in patients, and can form via a variety of mechanisms. Controlling aggregation can improve protein stability, manufacturability, attrition rates, safety, formulation, titers, immunogenicity, and solubility. The intrinsic properties of proteins such as size, hydrophobicity, electrostatics and charge distribution play important roles in protein solubility. Low solubility of therapeutic proteins due to surface hydrophobicity has been shown to render formulation development more difficult and may lead to poor bio-distribution, undesirable pharmacokinetics behavior and immunogenicity in vivo. Decreasing the overall surface hydrophobicity of candidate monoclonal antibodies can also provide benefits and cost savings relating to purification and dosing regimens. Individual amino acids can be identified by structural analysis as being contributory to aggregation potential in an antibody, and can be located in CDR as well as framework regions. In particular, residues can be predicted to be at high risk of causing

hydrophobicity issues in a given antibody. In one embodiment, the present disclosure provides an AF1 having the capability to specifically bind CD3 in which the AF1 has at least one amino acid substitution of a hydrophobic amino acid in a framework region relative to the parental antibody or antibody fragment wherein the hydrophobic amino acid is isoleucine, leucine or methionine. In some embodiments, the CD3 AF1 has at least two amino acid substitutions of hydrophobic amino acids in one or more framework regions wherein the hydrophobic amino acids are isoleucine, leucine or methionine.

[0167] The isoelectric point (pI) is the pH at which the antibody or antibody fragment has no net electrical charge. If the pH is below the pI of an antibody or antibody fragment, then it will have a net positive charge. A greater positive charge tends to correlate with increased blood clearance and tissue retention, with a generally shorter half-life. If the pH is greater than the pI of an antibody or antibody fragment it will have a negative charge. A negative charge generally results in decreased tissue uptake and a longer half-life. It is possible to manipulate this charge through mutations to the framework residues. These considerations informed the design of the sequences of the AF1 of the embodiments described herein wherein individual amino acid substitutions were made relative to the parental antibody utilized as the starting point. The isoelectric point of a polypeptide can be determined mathematically (e.g., computationally) or experimentally in an in vitro assay. The isoelectric point (pI) is the pH at which a protein has a net charge of zero and can be calculated using the charges for the specific amino acids in the protein sequence. Estimated values for the charges are called acid dissociation constants or pKa values and are used to calculate the pI. The pI can be determined in vitro by methods such as capillary isoelectric focusing (*see* Datta-Mannan, A., et al. The interplay of non-specific binding, target-mediated clearance and FcRn interactions on the pharmacokinetics of humanized antibodies. mAbs 7:1084 (2015); Li, B., et al. Framework selection can influence pharmacokinetics of a humanized therapeutic antibody through differences in molecule charge. mAbs 6, 1255–1264 (2014)) or other methods known in the art. In some embodiments, the isoelectric points of the AF1 and AF2 are designed to be within a particular range of each other, thereby promoting stability.

[0168] In one embodiment, the present disclosure provides an antigen binding fragment (e.g., AF1 or AF2) for use in any of the polypeptide embodiments described herein comprising CDR-L and CDR-H (see **Table 6b**), wherein the antigen binding fragment (e.g., AF1 or AF2) (a) specifically binds to cluster of differentiation 3 T cell receptor (CD3); and (b) comprises CDR-H1, CDR-H2, and CDR-H3, having amino acid sequences of **SEQ ID NOS: 8, 9, and 10**, respectively. In some embodiments, the CDR-H1 and the CDR-H2 of the antigen binding fragment (AF) can comprise amino acid sequences of **SEQ ID NOS: 8 and 9**, respectively. In some embodiments, the present disclosure provides an antigen binding fragment (e.g., AF1 or AF2) for use in any of the polypeptide embodiments described herein comprising CDR-L and CDR-H, wherein the antigen binding fragment (e.g., AF1 or AF2) (a) specifically binds to cluster of differentiation 3 T cell receptor (CD3); (b)

comprises CDR-H1, CDR-H2, and CDR-H3, having amino acid sequences of **SEQ ID NOS: 8, 9, and 10**, respectively. The antigen binding fragment (e.g., AF1 or AF2) can comprise CDR-L, wherein the CDR-L comprises a CDR-L1 having an amino acid sequence of **SEQ ID NOS: 1 or 2**, a CDR-L2 having an amino acid sequence of **SEQ ID NOS: 4 or 5**, and a CDR-L3 having an amino acid sequence of **SEQ ID NO:6**. In some embodiments, where the peptide comprises an antigen binding fragment (AF) (e.g., AF1 or AF2) comprising a CDR-L1, a CDR-L2, and a CDR-L3, the CDR-L1 of the AF can comprise an amino acid sequence of **SEQ ID NO:1 or 2**; the CDR-L2 of the AF can comprise an amino acid sequence of **SEQ ID NO: 4 or 5**; and said CDR-L3 of the AF can comprise an amino acid sequence of **SEQ ID NO:6**. In some embodiments, where the peptide comprises an antigen binding fragment (AF) (e.g., AF1 or AF2) comprising a CDR-L1, a CDR-L2, and a CDR-L3, the CDR-L1 of the AF can comprise an amino acid sequence of **SEQ ID NO:1**; the CDR-L2 of the AF can comprise an amino acid sequence of **SEQ ID NO: 4 or 5**; and said CDR-L3 of the AF can comprise an amino acid sequence of **SEQ ID NO:6**. In some embodiments, where the peptide comprises an antigen binding fragment (AF) (e.g., AF1 or AF2) comprising a CDR-L1, a CDR-L2, and a CDR-L3, the CDR-L1 of the AF can comprise an amino acid sequence of **SEQ ID NO: 2**; the CDR-L2 of the AF can comprise an amino acid sequence of **SEQ ID NO: 4 or 5**; and said CDR-L3 of the AF can comprise an amino acid sequence of **SEQ ID NO: 6**. In some embodiments, where the peptide comprises an antigen binding fragment (AF) (e.g., AF1 or AF2) comprising a CDR-L1, a CDR-L2, and a CDR-L3, the CDR-L1 of the AF can comprise an amino acid sequence of **SEQ ID NO: 1**; the CDR-L2 of the AF can comprise an amino acid sequence of **SEQ ID NO: 4**; and said CDR-L3 of the AF can comprise an amino acid sequence of **SEQ ID NO: 6**. In some embodiments, where the peptide comprises an antigen binding fragment (AF) (e.g., AF1 or AF2) comprising a CDR-L1, a CDR-L2, and a CDR-L3, the CDR-L1 of the AF can comprise an amino acid sequence of **SEQ ID NO: 2**; the CDR-L2 of the AF can comprise an amino acid sequence of **SEQ ID NO: 5**; and said CDR-L3 of the AF can comprise an amino acid sequence of **SEQ ID NO:6**.

[0169] In some embodiments, the foregoing antigen binding fragment (AF) (e.g., AF1 or AF2) embodiments of the immediately preceding paragraph further comprises light chain framework regions (FR-L) and heavy chain framework regions (FR-H) (see **Table 6c**) wherein the antigen binding fragment (AF) can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:51**, a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:52**, a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of any one of **SEQ ID NOS: 53-56**, a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:**

59. The antigen binding fragment (AF) can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:51**, a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:52**, a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 53**, a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 59**. The antigen binding fragment (AF) can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:51**, a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:52**, a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 54**, a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 59**. The antigen binding fragment (AF) can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:51**, a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:52**, a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 55**, a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 59**. The antigen binding fragment (AF) can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:51**, a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO:52**, a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 56**, a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 59**. The antigen binding fragment (AF) can comprise a FR-H1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of any one of **SEQ ID NOS: 60-63**, a FR-H2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is

identical to the amino acid sequence of **SEQ ID NO: 64**, a FR-H3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 65 or 66**; and a FR-H4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 67**. The antigen binding fragment (AF) can comprise a FR-H1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 60**, a FR-H2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 64**, a FR-H3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 65**; and a FR-H4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 67**. The antigen binding fragment (AF) can comprise a FR-H1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 61**, a FR-H2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 64**, a FR-H3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 65**; and a FR-H4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 67**. The antigen binding fragment (AF) (e.g., AF1 or AF2) for use in any of the polypeptide embodiments described herein can comprise light chain framework regions (FR-L) and heavy chain framework regions (FR-H) wherein the AF can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 51**; a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 52**; a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of any one of **SEQ ID NOS: 53-56**; a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 59**; a FR-H1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 60 or 61**; a FR-H2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 64**; a FR-H3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid

sequence of **SEQ ID NO: 65**; and a FR-H4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 67**. The AF can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 51**; a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 52**; a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 53**; a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 59**; a FR-H1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 60**; a FR-H2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 64**; a FR-H3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 65**; and a FR-H4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 67**. The AF can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 51**; a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 52**; a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 54**; a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 59**; a FR-H1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 61**; a FR-H2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 64**; a FR-H3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 65**; and a FR-H4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 67**. The AF can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 51**; a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence

identity or is identical to the amino acid sequence of **SEQ ID NO: 52**; a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 55**; a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 59**; a FR-H1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 61**; a FR-H2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 64**; a FR-H3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 65**; and a FR-H4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 67**. The AF can comprise a FR-L1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 51;** a FR-L2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 52**; a FR-L3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 56**; a FR-L4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 59**; a FR-H1 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 61**; a FR-H2 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 64**; a FR-H3 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 65**; and a FR-H4 exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to the amino acid sequence of **SEQ ID NO: 67**.

[0170] In some embodiments, the present disclosure provides an antigen binding fragment (AF) (e.g., AF1 or AF2) for use in any of the polypeptide embodiments described herein wherein the AF comprises a variable heavy (VH) amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to an amino acid sequence of **SEQ ID NO: 102** or **SEQ ID NO: 105** of **Table 6d**. In some embodiments, the present disclosure provides an AF for use in any of the polypeptide embodiments described herein wherein the AF comprises a variable light (VL) amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to an amino acid sequence

of any one of **SEQ ID NOS: 101, 103, 104, 106, or 107** of **Table 6d**. In some embodiment, the present disclosure provides an antigen binding fragment (e.g., AF1 or AF2) for use in any of the polypeptide embodiments described herein wherein the AF can comprise an amino acid sequence having at least 95%, 96%, 97%, 98%, 99% sequence identity or is identical to an amino acid sequence of any one of **SEQ ID NOS: 201-205** of **Table 6e**.

[0171] In some embodiments, the present disclosure provides antigen binding fragment (e.g., AF1 or AF2) that bind to the CD3 protein complex that have enhanced stability compared to CD3 binding antibodies or antigen binding fragments known in the art. Additionally, the CD3 antigen binding fragments of the disclosure are designed to confer a higher degree of stability on the chimeric bispecific antigen binding fragment compositions into which they are integrated, leading to improved expression and recovery of the fusion protein, increased shelf-life and enhanced stability when administered to a subject. In one approach, the CD3 AF of the present disclosure are designed to have a higher degree of thermal stability compared to certain CD3-binding antibodies and antigen binding fragments known in the art. As a result, the CD3 AF utilized as components of the chimeric bispecific antigen binding fragment compositions into which they are integrated exhibit favorable pharmaceutical properties, including high thermostability and low aggregation propensity, resulting in improved expression and recovery during manufacturing and storage, as well promoting long serum half-life. Biophysical properties such as thermostability are often limited by the antibody variable domains, which differ greatly in their intrinsic properties. High thermal stability is often associated with high expression levels and other desired properties, including being less susceptible to aggregation (Buchanan A, et al. Engineering a therapeutic IgG molecule to address cysteinylation, aggregation and enhance thermal stability and expression. MAbs 2013; 5:255). Thermal stability is determined by measuring the "melting temperature" (T_m), which is defined as the temperature at which half of the molecules are denatured. The melting temperature of each heterodimer is indicative of its thermal stability. In vitro assays to determine T_m are known in the art, including methods described in the Examples, below. The melting point of the heterodimer may be measured using techniques such as differential scanning calorimetry (Chen et al (2003) Pharm Res 20:1952-60; Ghirlando et al (1999) Immunol Lett 68:47-52). Alternatively, the thermal stability of the heterodimer may be measured using circular dichroism (Murray et al. (2002) J. Chromatogr Sci 40:343-9), or as described in the Examples, below.

[0172] In some embodiments of the polypeptides of this disclosure, the antigen binding fragment (e.g., AF1 or AF2) can exhibit a higher thermal stability than an anti-CD3 binding fragment consisting of a sequence of **SEQ ID NO: 206** (see **Table 6e**), as evidenced in an *in vitro* assay by a higher melting temperature (T_m) of the first antigen binding fragment relative to that of the anti-CD3 binding fragment; or upon incorporating the first antigen binding fragment into a test bispecific antigen binding construct, a higher T_m of the test bispecific antigen binding construct relative to that of a control bispecific antigen binding construct, wherein the test

bispecific antigen binding construct comprises the first antigen binding fragment and a reference antigen binding fragment that binds to an antigen other than CD3; and wherein the control bispecific antigen binding construct consists of the anti-CD3 binding fragment consisting of the sequence of **SEQ ID NO:206** (see **Table 6e**) and the reference antigen binding fragment. The melting temperature (T_m) of the first antigen binding fragment can be at least 2 °C greater, or at least 3°C greater, or at least 4°C greater, or at least 5°C greater than the T_m of the anti-CD3 binding fragment consisting of the sequence of **SEQ ID NO: 206** (see **Table 6e**).

[0173] Thermal denaturation curves of the CD3 binding fragments and the anti-CD3 bispecific antibodies comprising the anti-CD3 binding fragment and a reference binding of the present disclosure show that the constructs of the present disclosure are more resistant to thermal denaturation than the antigen binding fragment consisting of a sequence shown in **SEQ ID NO: 781** (see **Table 6f**) or a control bispecific antibody wherein the control bispecific antigen binding fragment comprises **SEQ ID NO: 781** (see **Table 6f**) and a reference antigen binding fragment that binds to a HER2 embodiment described herein. In one embodiment, the polypeptides of any of the subject composition embodiments described herein comprise an anti-CD3 AF of the embodiments described herein, wherein the T_m of the AF is at least 2°C greater, or at least 3°C greater, or at least 4°C greater, or at least 5°C greater, or at least 6°C greater, or at least 7°C greater, or at least 8°C greater, or at least 9°C greater, or at least 10°C greater than the T_m of an antigen binding fragment consisting of a sequence of **SEQ ID NO: 781** (see **Table 6f**), as determined by an increase in melting temperature in an in vitro assay.

[0174] In some embodiments, the polypeptides of any of the subject composition embodiments described herein comprise an antigen binding fragment (AF) that specifically bind human or cynomolgus monkey (cyno) CD3. The antigen binding fragment (**AF**) can specifically bind human CD3. The antigen binding fragment (**AF**) can bind a CD3 complex subunit identified herein as CD3 epsilon, CD3 delta, CD3 gamma, or CD3 zeta unit of CD3. The antigen binding fragment (**AF**) can bind a CD3 epsilon fragment of CD3. The antigen binding fragment (**AF**) can specifically bind human or cyno CD3 with a dissociation constant (K_d) constant between about 10 nM and about 400 nM, or between about 50 nM and about 350 nM, or between about 100 nM and 300 nM, as determined in an in vitro antigen-binding assay comprising a human or cyno CD3 antigen. In some embodiments, the polypeptides of any of the subject composition embodiments described herein comprise an antigen binding fragment (AF) that specifically binds human or cyno CD3 with a dissociation constant (K_d) weaker than about 10 nM, or about 50 nM, or about 100 nM, or about 150 nM, or about 200 nM, or about 250 nM, or about 300 nM, or about 350 nM, or weaker than about 400 nM as determined in an in vitro antigen-binding assay. For clarity, an antigen binding fragment (AF) with a K_d of 400 binds its ligand more weakly than one with a K_d of 10 nM. In some embodiments, the polypeptides of any of the subject composition embodiments described herein comprise an antigen binding fragment (AF) that specifically binds human or

cyno CD3 with at least 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, or at least 10-fold weaker binding affinity than an antigen binding fragment consisting of an amino acid sequence of **SEQ ID NO: 781** (see **Table 6f**), as determined by the respective dissociation constants (K_d) in an in vitro antigen-binding assays. In some embodiments, the present disclosure provides bispecific polypeptides comprising an antigen binding fragment (AF) that exhibits a binding affinity to CD3 (anti-CD3 AF) that is at least 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, 10-fold, 20-fold, 50-fold, 100-fold, or at least 1000-fold at weaker relative to that of an anti-HER2 AF embodiments described herein that are incorporated into the subject polypeptides, as determined by the respective dissociation constants (K_d) in an in vitro antigen-binding assay. The binding affinity of the subject compositions for the target ligands can be assayed using binding or competitive binding assays, such as Biacore assays with chip-bound receptors or binding proteins or ELISA assays, as described in US Patent 5,534,617, assays described in the Examples herein, radio-receptor assays, or other assays known in the art. The binding affinity constant can then be determined using standard methods, such as Scatchard analysis, as described by van Zoelen, et al., Trends Pharmacol Sciences (1998) 19)12):487, or other methods known in the art.

[0175] In a related aspect, the present disclosure provides an antigen binding fragment (AF) that bind to CD3 (anti-CD3 AF) and are incorporated into chimeric, bispecific polypeptide compositions that are designed to have an isoelectric point (pI) that confer enhanced stability on the compositions of the disclosure compared to corresponding compositions comprising CD3 binding antibodies or antigen binding fragments known in the art. In one embodiment, the polypeptides of any of the subject composition embodiments described herein comprise AF that bind to CD3 (anti-CD3 AF) wherein the anti-CD3 AF exhibits a pI that is between 6.0 and 6.6, inclusive. In some embodiments, the polypeptides of any of the subject composition embodiments described herein comprise AF that bind to CD3 (anti-CD3 AF) wherein the anti-CD3 AF exhibits a pI that is at least 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, or 1.0 pH unit lower than the pI of a reference antigen binding fragment (*e.g.*, consisting of a sequence shown in **SEQ ID NO: 206** (see **Table 6e**)). In some embodiments, the polypeptides of any of the subject composition embodiments described herein comprise an AF that binds to CD3 (anti-CD3 AF) fused to another AF that binds to a HER2 antigen (anti-HER2 AF) wherein the anti-CD3 AF exhibits a pI that is within at least 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1.0, 1.1, 1.2, 1.3, 1.4, or 1.5 pH units of the pI of the AF that binds HER2 antigen or an epitope thereof. In some embodiments, the polypeptides of any of the subject composition embodiments described herein comprise an AF that binds to CD3 (anti-CD3 AF) fused to an AF that binds to a HER2 antigen (anti-HER2 AF) wherein the AF exhibits a pI that is within at least about 0.1 to about 1.5, or at least about 0.3 to about 1.2, or at least about 0.5 to about 1.0, or at least about 0.7 to about 0.9 pH units of the pI of the anti-CD3 AF. It is specifically intended that by such design wherein the pI of the two antigen binding fragments are within such ranges, the resulting fused antigen binding fragments will confer

a higher degree of stability on the chimeric bispecific antigen binding fragment compositions into which they are integrated, leading to improved expression and enhanced recovery of the fusion protein in soluble, non-aggregated form, increased shelf-life of the formulated chimeric bispecific polypeptide compositions, and enhanced stability when the composition is administered to a subject. State differently, having the two AFs (the anti-CD3 AF and the anti-HER2 AF) within a relatively narrow pI range of may allow for the selection of a buffer or other solution in which both the AFs (anti-CD3 AF and anti-HER2 AF) are stable, thereby promoting overall stability of the composition. The antigen binding fragment (**AF**) can exhibit an isoelectric point (pI) that is less than or equal to 6.6. The antigen binding fragment (**AF**) can exhibit an isoelectric point (pI) that is between 6.0 and 6.6, inclusive. The antigen binding fragment (**AF**) can exhibit an isoelectric point (pI) that is at least 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, or 1.0 pH units lower than the pI of a reference antigen binding fragment consisting of a sequence shown in **SEQ ID NO: 206** (see **Table 6e**). The antigen binding fragment (**AF**) can specifically bind human or cyno CD3 with a dissociation constant (K_d) constant between about between about 10 nM and about 400 nM (such as determined in an *in vitro* antigen-binding assay comprising a human or cyno CD3 antigen). The antigen binding fragment (**AF**) can specifically bind human or cyno CD3 with a dissociation constant (K_d) of less than about 10 nM, or less than about 50 nM, or less than about 100 nM, or less than about 150 nM, or less than about 200 nM, or less than about 250 nM, or less than about 300 nM, or less than about 350 nM, or less than about 400 nM (such as determined in an *in vitro* antigen-binding assay). The antigen binding fragment (**AF**) can exhibit a binding affinity to CD3 that is at least 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, or at least 10-fold weaker relative to that of an antigen binding fragment consisting of an amino acid sequence of **SEQ ID NO: 206** (see **Table 6e**) (such as determined by the respective dissociation constants (K_d) in an *in vitro* antigen-binding assay).

[0176] In certain embodiments, the VL and VH of the antigen binding fragments are fused by relatively long linkers, consisting 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, or 35 hydrophilic amino acids that, when joined together, have a flexible characteristic. In one embodiment, the VL and VH of any of the scFv embodiments described herein are linked by relatively long linkers of hydrophilic amino acids having the sequences GSGEGSEGEGGGEGSEGEGSGEGGEGEGSG (SEQ ID NO: 82), TGSGEGSEGEGGGEGSEGEGSGEGGEGEGSGT (SEQ ID NO: 83), GATPPETGAETESPGETTGGSAESEPPGEG (SEQ ID NO: 84), or GSAAPTAGTTPSASPAPPTGGSSAAGSPST (SEQ ID NO: 85). In some embodiments, the AF1 and AF2 are linked together by a short linker of hydrophilic amino acids having 3, 4, 5, 6, or 7 amino acids. In one embodiment, the short linker sequences are identified herein as the sequences SGGGGS (SEQ ID NO: 86), GGGGS (SEQ ID NO: 87), GGSGGS (SEQ ID NO: 88), GGS, or GSP. In some embodiments, the disclosure provides compositions comprising a single chain diabody in which after folding, the first domain (VL or VH)

is paired with the last domain (VH or VL) to form one scFv and the two domains in the middle are paired to form the other scFv in which the first and second domains, as well as the third and last domains, are fused together by one of the foregoing short linkers and the second and the third variable domains are fused by one of the foregoing relatively long linkers. As will be appreciated by one of skill in the art, the selection of the short linker and relatively long linker is to prevent the incorrect pairing of adjacent variable domains, thereby facilitating the formation of the single chain diabody configuration comprising the VL and VH of the first antigen binding fragment and the second antigen binding fragment.

Table 6b. Exemplary CD3 CDR Sequences

Construct	CDR REGION	Amino Acid Sequence	SEQ ID NO:
3.23, 3.30, 3.31, 3.32	CDR-L1	RSSNGAVTSSNYAN	1
3.24	CDR-L1	RSSNGEVTTSNYAN	2
3.33, 3.9	CDR-L1	RSSTGAVTTSNYAN	3
3.23, 3.30, 3.31, 3.32, 3.9, 3.33	CDR-L2	GTNKRAP	4
3.24	CDR-L2	GTIKRAP	5
3.23, 3.24, 3.30, 3.31, 3.32	CDR-L3	ALWYPNLWVF	6
3.33, 3.9	CDR-L3	ALWYSNLWVF	7
3.23, 3.24, 3.30, 3.31, 3.32, 3.9, 3.33	CDR-H1	GFTFNTYAMN	8
3.23, 3.24, 3.30, 3.31, 3.32, 3.9, 3.33	CDR-H2	RIRSKYNNYATYYADSVKD	9
3.23, 3.24, 3.30, 3.31, 3.32	CDR-H3	HENFGNSYVSWFAH	10
3.9, 3.33	CDR-H3	HGNFGNSYVSWFAY	11

Table 6c. Exemplary CD3 FR Sequences

Construct	FR REGION	Amino Acid Sequence	SEQ ID NO:
3.23, 3.24, 3.30, 3.31, 3.32, 3.9, 3.33	FR-L1	ELVVTQEPSLTVSPGGTVTLTC	51
3.23, 3.24, 3.30, 3.31, 3.32, 3.9, 3.33	FR-L2	WVQQKPGQAPRGLIG	52
3.23, 3.24	FR-L3	GTPARFSGSLLGGKAALTLSGVQPEDEAVYYC	53
3.30	FR-L3	GTPARFSGSSLGGKAALTLSGVQPEDEAVYYC	54
3.31	FR-L3	GTPARFSGSLLGGSAALTLSGVQPEDEAVYYC	55
3.32	FR-L3	GTPARFSGSSLGGSAALTLSGVQPEDEAVYYC	56
3.9	FR-L3	GTPARFSGSLLGGKAALTLSGVQPEDEAEYYC	57
3.33	FR-L3	GTPARFSGSSLGGSAALTLSGVQPEDEAEYYC	58
3.23, 3.24, 3.30, 3.31, 3.32, 3.9, 3.33	FR-L4	GGGTKLTVL	59
3.23, 3.24	FR-H1	EVQLLESGGGIVQPGGSLKLSCAAS	60
3.30, 3.31, 3.32	FR-H1	EVQLQESGGGIVQPGGSLKLSCAAS	61
3.33	FR-H1	EVQLQESGGGLVQPGGSLKLSCAAS	62
3.9	FR-H1	EVQLLESGGGLVQPGGSLKLSCAAS	63
3.23, 3.24, 3.30, 3.31, 3.32, 3.9, 3.33	FR-H2	WVRQAPGKGLEWVA	64
3.23, 3.24, 3.30, 3.31, 3.32	FR-H3	RFTISRDDSKNTVYLQMNNLKTEDTAVYYCVR	65
3.9, 3.33	FR-H3	RFTISRDDSKNTAYLQMNNLKTEDTAVYYCVR	66

Construct	FR REGION	Amino Acid Sequence	SEQ ID NO:
3.23, 3.24, 3.30, 3.31, 3.32, 3.9, 3.33	FR-H4	WGQGTLVTVSS	67

Table 6d: Exemplary CD3 VL & VH Sequences

Construct	REGION	Amino Acid Sequence	SEQ ID NO:
3.23	VL	ELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAP RGLIGGTNKRAPGTPARFSGSLLGGKAALTLSGVQPEDEAVYYCAL WYPNLWVFGGGTKLTVL	101
3.23, 3.24	VH	EVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGL EWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTVYLQMNNLKT EDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSS	102
3.24	VL	ELVVTQEPSLTVSPGGTVTLTCRSSNGEVTTSNYANWVQQKPGQAP RGLIGGTIKRAPGTPARFSGSLLGGKAALTLSGVQPEDEAVYYCAL WYPNLWVFGGGTKLTVL	103
3.30	VL	ELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAP RGLIGGTNKRAPGTPARFSGSSLGGKAALTLSGVQPEDEAVYYCAL WYPNLWVFGGGTKLTVL	104
3.30, 3.31, 3.32	VH	EVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGL EWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTVYLQMNNLKT EDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSS	105
3.31	VL	ELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAP RGLIGGTNKRAPGTPARFSGSLLGGSAALTLSGVQPEDEAVYYCAL WYPNLWVFGGGTKLTVL	106
3.32	VL	ELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAP RGLIGGTNKRAPGTPARFSGSSLGGSAALTLSGVQPEDEAVYYCAL WYPNLWVFGGGTKLTVL	107
3.9	VL	ELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAP RGLIGGTNKRAPGTPARFSGSLLGGKAALTLSGVQPEDEAEYYCAL WYSNLWVFGGGTKLTVL	108
3.9	VH	EVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGL EWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTAYLQMNNLKT EDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSS	109
3.33	VL	ELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAP RGLIGGTNKRAPGTPARFSGSSLGGSAALTLSGVQPEDEAEYYCAL WYSNLWVFGGGTKLTVL	110
3.33	VH	EVQLQESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGL EWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTAYLQMNNLKT EDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSS	111

Table 6e: Exemplary CD3 scFv Sequences

Construct	Amino Acid Sequence	SEQ ID NO:
3.23	ELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNK RAPGTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVL GATPPETGAETESPGETTGGSAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGF TFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTV YLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSS	201

Construct	Amino Acid Sequence	SEQ ID NO:
3.24	ELVVTQEPSLTVSPGGTVTLTCRSSNGEVTTSNYANWVQQKPGQAPRGLIGGTIKR APGTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLG ATPPETGAETESPGETTGGSAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFT FNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTV YLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSS	202
3.30	ELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNK RAPGTPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVL GATPPETGAETESPGETTGGSAESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGF TFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTV YLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSS	203
3.31	ELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNK RAPGTPARFSGSLLGGSAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVL GATPPETGAETESPGETTGGSAESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGF TFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTV YLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSS	204
3.32	ELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNK RAPGTPARFSGSSLGGSAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVL GATPPETGAETESPGETTGGSAESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGF TFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTV YLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSS	205
3.9	ELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNK RAPGTPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVL GATPPETGAETESPGETTGGSAESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGF TFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTA YLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSS	206
3.33	ELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNK RAPGTPARFSGSSLGGSAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLG ATPPETGAETESPGETTGGSAESEPPGEGEVQLQESGGGLVQPGGSLKLSCAASGFT FNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSKNTA YLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSS	207
4.11	QSVLTQPPSASGTPGQRVTISCSGSSSNIGSNYVYWYQQLPGTAPKLLIYRNNQRPS GVPDRFSGSKSGTSASLAISGLRSEDEADYYCAAWDDSLSGLWVFGGGTKLTVLG ATPPETGAETESPGETTGGSAESEPPGEGQVQLQQWGGGLVKPGGSLRLSCAASGF TFSSYSMNWVRQAPGKGLEWVSRINSDGSSTNYADSVKGRFTISRDNAKNTLYLQ MNSLRAEDTAVYYCARELRWGNWGQGTLVTVSS	208
4.12	QAGLTQPPSASGTPGQRVTLSCSGSYSNIGTYYVYWYQQLPGTAPKLLIYSNDQRL SGVPDRFSGSKSGTSASLAISGLQSEDEAAYYCAAWDDSLNGWAFGGGTKLTVLG ATPPETGAETESPGETTGGSAESEPPGEGQVQLQQWGGGLVKPGGSLRLSCAASGF TFSSYSMNWVRQAPGKGLEWVSRINSDGSSTNYADSVKGRFTISRDNAKNTLYLQ MNSLRAEDTAVYYCARELRWGNWGQGTLVTVSS	209
4.13	QPGLTQPPSASGTPGQRVTLSCSGRSSNIGSYYVYWYQHLPGMAPKLLIYRNSRRP SGVPDRFSGSKSGTSASLVISGLQSDDEADYYCAAWDDSLKSWVFGGGTKLTVLG ATPPETGAETESPGETTGGSAESEPPGEGQVQLQQWGGGLVKPGGSLRLSCAASGF TFSSYSMNWVRQAPGKGLEWVSRINSDGSSTNYADSVKGRFTISRDNAKNTLYLQ MNSLRAEDTAVYYCARELRWGNWGQGTLVTVSS	210
4.14	QSVLTQPPSASGTPGQRVTISCSGSSSNIGTNYVYWYQQFPGTAPKLLIYSNNQRPS GVPDRFSGSKSGTSGSLAISGLQSEDEADYSCAAWDDSLNGWVFGGGTKLTVLGA TPPETGAETESPGETTGGSAESEPPGEGQVQLVQWGGGLVKPGGSLRLSCAASGFT FSSYSMNWVRQAPGKGLEWVSRINSDGSSTNYADSVKGRFTISRDNAKNTLYLQ MNSLRAEDTAVYYCARELRWGNWGQGTLVTVSS	211

Construct	Amino Acid Sequence	SEQ ID NO:
4.15	QPGLTQPPSASGTPGQRVTISCSGSSSNIGSNYVYWYQQLPGTAPKLLIYRNNQRPS GVPDRLSGSKSGTSASLAISGLRSEDEADYYCAAWDDSLSGWVFGGGTKLTVLGA TPPETGAETESPGETTGGSAESEPPGEGQVQLVQWGGGLVKPGGSLRLSCAASGFT FSSYSMNWVRQAPGKGLEWVSRINSDGSSTNYADSVKGRFTISRDNAKNTLYLQ MNSLRAEDTAVYYCARELRWGNWGQGTLVTVSS	212
4.16	QAVLTQPPSASGTPGQRVTISCSGSSSNIGSYYVYWYQQVPGAAPKLLMRLNNQR PSGVPDRFSGAKSGTSASLVISGLRSEDEADYYCAAWDDSLSGQWVFGGGTKLTV LGATPPETGAETESPGETTGGSAESEPPGEGQVQLQQWGGGLVKPGGSLRLSCAAS GFTFSSYSMNWVRQAPGKGLEWVSRINSDGSSTNYADSVKGRFTISRDNAKNTLY LQMNSLRAEDTAVYYCARELRWGNWGQGTLVTVSS	213
4.17	QAGLTQPPSASGTPGQRVTISCSGSSSNIGSNYVYWYQQLPGTAPKLLIYRNNQRPS GVPDRFSGSKSGTSASLAISGLRSEDEADYYCATWDASLSGWVFGGGTKLTVLGA TPPETGAETESPGETTGGSAESEPPGEGEVQLVQWGGGLVKPGGSLRLSCAASGFT FSSYSMNWVRQAPGKGLEWVSRINSDGSSTNYADSVKGRFTISRDNAKNTLYLQ MNSLRAEDTAVYYCARELRWGNWGQGTLVTVSS	214

Anti-HER-2 Binding Domains

[0177] In some embodiments, the invention provides chimeric polypeptide assembly compositions comprising a first portion binding domain with binding affinity to the tumor-specific marker HER-2 and a second binding domain binds to an effector cell antigen, such as CD3 antigen. In one embodiment, the binding domain comprises VL and VH derived form a monoclonal antibody to HER-2. Monoclonal antibodies to HER-2 are known in the art. Exemplary, non-limiting examples of VL and VH sequences are presented in **Table 6f**. In one embodiment, the invention provides a chimeric polypeptide assembly composition comprising a first portion binding domain with binding affinity to the tumor-specific marker HER-2 comprising anti-HER-2 VL and VH sequences set forth in **Table 6f**. In some embodiments, the invention provides a chimeric polypeptide assembly composition comprising a first portion binding domain with binding affinity to the tumor-specific marker comprising the CDR-L1 region, the CDR-L2 region, the CDR-L3 region, the CDR-H1 region, the CDR-H2 region, and the CDR-H3 region, wherein each is derived from the respective VL and VH sequences set forth in **Table 6f**. Preferably, in the embodiments, the binding has a K_d value of greater than 10^{-10} to 10^{-7} M, as determined in an vitro binding assay. In some embodiments, where the polypeptide comprises an antigen binding fragment that specifically binds to HER2 (anti-HER2 AF), the anti-HER2 AF (e.g., AF1 or AF2) can comprise (1) a heavy chain variable region (VH_{II}) comprising an amino acid sequence set forth as **SEQ ID NOS: 778-783**, and (2) a light chain variable region (VL_{II}) comprising an amino acid sequence set forth as **SEQ ID NOS: 878-883** of **Table 6f**. It is specifically contemplated that the chimeric polypeptide assembly composition can comprise any one of the foregoing binding domains or sequence variants thereof so long as the variants exhibit binding specificity for the described antigen. In one embodiment, a sequence variant would be created by substitution of an amino acid in the VL or VH sequence with a different amino acid. In deletion variants, one or more amino acid residues in a VL or VH sequence as described herein are removed. Deletion

variants, therefore, include all fragments of a binding domain polypeptide sequence. In substitution variants, one or more amino acid residues of a VL or VH (or CDR) polypeptide are removed and replaced with alternative residues. In one aspect, the substitutions are conservative in nature and conservative substitutions of this type are well known in the art. In addition, it is specifically contemplated that the compositions comprising the first and the second binding domains disclosed herein can be utilized in any of the methods disclosed herein.

Table 6f. Anti-HER2 Monoclonal Antibodies and Sequences

Trade Name	Antibody Name	Target	VH Sequence	SEQ ID NO:	VL Sequence	SEQ ID NO:
	C1	HER2	QVQLVESGGGLVQP GGSLRLSCAASGFTF S**SYAMG**WVRQAPG KGLEWVS**SISGSSR YIYYADSVKG**RFTIS RDNSKNTLYLQMNS LRAEDTAVYYCAK **MDASGSYFNF**WGQ GTLVTVSS	778	QSPSFLSAFVGDRITI TC**RASPGIRNYLA**W YQQKPGKAPKLLIY **AASTLQS**GVPSRFS GSGSGTDFTLTISSL QPEDFATYYC**QQYN SYPLS**FGGGTKVEIK	878
Erbicin		HER2	QVQLLQSAAEVKKP GESLKISCKGSGYSF T**SYWIG**WVRQMPG KGLEWMG**IIYPGDS DTRYSPSFQG**QVTIS ADKSISTAYLQWSSL KASDTAVYYCAR**W RDSPL**WGQGTLVTV SS	779	QAVVTQEPSFSVSPG GTVTLTC**GLSSGSV STSYYPS**WYQQTPG QAPRTLIY**STNTRSS** GVPDRFSGSILGNKA ALTITGAQADDESD YYC**VLYMGSGQYV** FGGGTKLTVL	879
Herceptin	trastuzumab	HER2	EVQLVESGGGLVQP GGSLRLSCAAS**GFNI KDTY**IHWVRQAPG KGLEWVAR**IYPTNG YT**RYADSVKGRFTIS ADTSKNTAYLQMNS LRAEDTAVYYC**SR WGGDGFYAMDY**W GQGTLVTVSS	780	DIQMTQSPSSLSASV GDRVTITCRAS**QDV NTA**VAWYQQKPGK APKLLIY**SAS**FLYSG VPSRFSGSRSGTDFT LTISSLQPEDFATYY C**QQHYTTPPT**FGQG TKVEIK	880
MAGH22	margetuximab	HER2	QVQLQQSGPELVKP GASLKLSCTAS**GFNI KDTY**IHWVKQRPEQ GLEWIGR**IYPTNGY T**RYDPKFQDKATIT ADTSSNTAYLQVSR LTSEDTAVYYC**SRW GGDGFYAMDY**WG QGASVTVSS	781	DIVMTQSHKFMSTS VGDRVSITCKAS**QD VNTA**VAWYQQKPG HSPKLLIY**SAS**FRYT GVPDRFTGSRSGTDF TFTISSVQAEDLAVY YC**QQHYTTPPT**FGG GTKVEIK	881

Trade Name	Antibody Name	Target	VH Sequence	SEQ ID NO:	VL Sequence	SEQ ID NO:
MM-302	F5	HER2	QVQLVESGGGLVQP GGSLRLSCAASGFTF R**SYAMS**WVRQAPG KGLEWVS**AISGRGD NTYYADSVKG**RFTI SRDNSKNTLYLQMN SLRAEDTAVYYC**AK MTSNAFAFDY**WGQ GTLVTVSS	782	QSVLTQPPSVSGAPG QRVTISC**TGSSSNIG AGYGVH**WYQQLPG TAPKLLIY**GNTNRPS** GVPDRFSGFKSGTSA SLAITGLQAEDEAD YYC**QFYDSSLSGW V**FGGGTKLTVL	882
Perjeta	pertuzumab	HER2	EVQLVESGGGLVQP GGSLRLSCAAS**GFT FTDYT**MDWVRQAP GKGLEWVAD**VNPN SGGS**IYNQRFKGRFT LSVDRSKNTLYLQM NSLRAEDTAVYYC**A RNLGPSFYFDY**WG QGTLVTVSS	783	DIQMTQSPSSLSASV GDRVTITCKASQ**DV SIG**VAWYQQKPGK APKLLIY**SAS**YRYTG VPSRFSGSGSGTDFT LTISSLQPEDFATYY C**QQYYIYPYT**FGQG TKVEIK	883

* underlined & bolded sequences, if present, are CDRs within the VL and VH

Table A: Intramolecular Long Linkers

Linker #	Name	SEQ ID	Amino Acid Sequence
L1	(G4S)3	112	GGGGSGGGGSGGGGS
L2	MT110_18	113	GEGTSTGSGGSGGSGGAD
L3	MT103_18	114	VEGGSGGSGGSGGSGGVD
L4	UCHT1_29	115	RTSGPGDGGKGGPGKGPGGEGTKGTGPGG
L5	Y30	116	GSGEGSEGEGGGEGSEGEGSGEGGEGEGSG
L6	Y32	117	TGSGEGSEGEGGGEGSEGEGSGEGGEGEGSGT
L7	G1_30_3	118	GAPPPETGAETESPGETTGGSAESEPPGEG
L8	G9_30_1	119	GSAAPTAGTTPSASPAPPTGGSSAAGSPST
L9	Y30_modified	120	GEGGESGGSEGEGSGEGEGGSGGEGESEGG
L10	G1_30_1	121	STETSPSTPTESPEAGSGSGSPESPSGTEA
L11	G1_30_2	122	PTGTTGEPSGEGSEPEGSAPTSSTSEATPS
L12	G1_30_4	123	SESESEGEAPTGPGASTTPEPSESPTPETS
L13	UCHT1_modified	124	PEGGESGEGTGPGTGGEPEGEGGPGGEGGT

Table B: Intermolecular Short Linkers

Name	Amino Acid Sequence
S-1	SGGGGS (SEQ ID NO: 86)
S-2	GGGGS (SE ID NO: 87)
S-3	GGS
S-4	GSP

[0178] In some embodiments of the polypeptides of this disclosure, a pair of the light chain variable region (VL) and the heavy chain variable region (VH) of an antigen binding fragment can be linked by a linker, or a

long linker (e.g., of hydrophilic amino acids). Such linker, linking the light chain variable region (VL) and the heavy chain variable region (VH) of an antigen binding fragment (e.g., a first antigen binding fragment (AF1), a second antigen binding fragment (AF2)), can (each independently) comprise an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table A**. Such linker, linking the light chain variable region (VL) and the heavy chain variable region (VH) of an antigen binding fragment (e.g., a first antigen binding fragment (AF1), a second antigen binding fragment (AF2)), can (each independently) comprise an amino acid sequence identical to a sequence set forth in **Table A**. In some embodiments of the polypeptides of this disclosure, two antigen binding fragments (e.g., a first and a second antigen binding fragments) can be fused together by a peptide linker, or a short linker. Such peptide linker, linking two antigen binding fragments (e.g., a first and a second antigen binding fragments), can comprise an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table B**. Such peptide linker, linking two antigen binding fragments (e.g., a first and a second antigen binding fragments), can comprise an amino acid sequence identical to a sequence set forth in **Table B**. In some cases, the first antigen binding fragment is a single-chain variable fragment (scFv). In some cases, the second antigen binding fragment is a single-chain variable fragment (scFv). The two single-chain variable fragments of the first and second antigen binding fragments can be linked together by the peptide linker. In some embodiments of the polypeptides of this disclosure, the linker used to link the VL and VH of the first antigen binding fragment or/and the linker used to link the VL and VH of the second antigen binding fragment can be L7 of **Table A**. In such embodiments, the peptide linker used to link the two antigen binding fragments can be S-1 or S-2 of **Table B**. In some embodiments, the disclosure provides polypeptides comprising a single chain diabody in which after folding, the first domain (VL or VH) is paired with the last domain (VH or VL) to form one scFv and the two domains in the middle are paired to form the other scFv in which the first and second domains, as well as the third and last domains, are fused together by a short linker of hydrophilic amino acids identified herein by the sequences set forth in **Table B** and the second and the third variable domains are fused by a long linker identified in **Table A**. As will be appreciated by one of skill in the art, the selection of the short linker and long linker is to prevent the incorrect pairing of adjacent variable domains, thereby facilitating the formation of the single chain diabody configuration comprising the VL and VH of the first binding moiety and the second binding moiety.

Table C: Exemplary Spacers between a Release Segment and a Bispecific Antibody Construct

Amino Acid Sequence	SEQ ID NO:
STEPS	89
SATPESGPGT	90
ATSGSETPGT	91
GTAEAASASG	92
STEPSEGSAPGTS	93

Amino Acid Sequence	SEQ ID NO:
SGPGTS	94
GTSTEPS	95

[0179] In some embodiments of the polypeptides of this disclosure, a release segment (RS) (e.g., a first release segment (RS1), a second release segment (RS2), etc.) can be fused to a bispecific antibody construct (BsAb) by a spacer. Such spacer can (each independently) comprise at least 4 types of amino acids that are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P). The peptides of this disclosure can comprise a first release segment fused to the bispecific antibody construct via a first spacer and a second release segment fused to the bispecific antibody construct via a second space. A spacer (*e.g.*, a first spacer, a second spacer, etc.) can (each independently) comprise an amino acid sequence having at least (about) 80%, at least (about) 90%, or 100% sequence identity to a sequence set forth in **Table C**. The spacer (*e.g.*, the first spacer, the second spacer, etc.) can (each independently) comprise an amino acid sequence identical to a sequence set forth in **Table C**.

UNSTRUCTURED CONFORMATION

[0180] Typically, the XTEN component of the fusion proteins are designed to behave like denatured peptide sequences under physiological conditions, despite the extended length of the polymer. Denatured describes the state of a peptide in solution that is characterized by a large conformational freedom of the peptide backbone. Most peptides and proteins adopt a denatured conformation in the presence of high concentrations of denaturants or at elevated temperature. Peptides in denatured conformation have, for example, characteristic circular dichroism (CD) spectra and are characterized by a lack of long-range interactions as determined by NMR. "Denatured conformation" and "unstructured conformation" are used synonymously herein. In some cases, the invention provides XTEN sequences that, under physiologic conditions, can resemble denatured sequences largely devoid in secondary structure. In other cases, the XTEN sequences can be substantially devoid of secondary structure under physiologic conditions. "Largely devoid," as used in this context, means that less than 50% of the XTEN amino acid residues of the XTEN sequence contribute to secondary structure as measured or determined by the means described herein. "Substantially devoid," as used in this context, means that at least about 60%, or about 70%, or about 80%, or about 90%, or about 95%, or at least about 99% of the XTEN amino acid residues of the XTEN sequence do not contribute to secondary structure, as measured or determined by the means described herein.

[0181] A variety of methods have been established in the art to discern the presence or absence of secondary and tertiary structures in a given polypeptide. In particular, XTEN secondary structure can be measured spectrophotometrically, e.g., by circular dichroism spectroscopy in the "far-UV" spectral region (190-250 nm). Secondary structure elements, such as alpha-helix and beta-sheet, each give rise to a characteristic shape and magnitude of CD spectra. Secondary structure can also be predicted for a polypeptide sequence via certain

computer programs or algorithms, such as the well-known Chou-Fasman algorithm (Chou, P. Y., *et al.* (1974) *Biochemistry*, 13: 222-45) and the Garnier-Osguthorpe-Robson ("GOR") algorithm (Garnier J, Gibrat JF, Robson B. (1996), GOR method for predicting protein secondary structure from amino acid sequence. Methods Enzymol 266:540-553), as described in US Patent Application Publication No. 20030228309A1. For a given sequence, the algorithms can predict whether there exists some or no secondary structure at all, expressed as the total and/or percentage of residues of the sequence that form, for example, alpha-helices or beta-sheets or the percentage of residues of the sequence predicted to result in random coil formation (which lacks secondary structure).

[0182] In some cases, the XTEN sequences used in the inventive fusion protein compositions can have an alpha-helix percentage ranging from 0% to less than about 5% as determined by a Chou-Fasman algorithm. In other cases, the XTEN sequences of the fusion protein compositions can have a beta-sheet percentage ranging from 0% to less than about 5% as determined by a Chou-Fasman algorithm. In some cases, the XTEN sequences of the fusion protein compositions can have an alpha-helix percentage ranging from 0% to less than about 5% and a beta-sheet percentage ranging from 0% to less than about 5% as determined by a Chou-Fasman algorithm. In preferred embodiments, the XTEN sequences of the fusion protein compositions will have an alpha-helix percentage less than about 2% and a beta-sheet percentage less than about 2%. In other cases, the XTEN sequences of the fusion protein compositions can have a high degree of random coil percentage, as determined by a GOR algorithm. In some embodiments, an XTEN sequence can have at least about 80%, more preferably at least about 90%, more preferably at least about 91%, more preferably at least about 92%, more preferably at least about 93%, more preferably at least about 94%, more preferably at least about 95%, more preferably at least about 96%, more preferably at least about 97%, more preferably at least about 98%, and most preferably at least about 99% random coil, as determined by a GOR algorithm.

NET CHARGE

[0183] In other cases, the XTEN polypeptides can have an unstructured characteristic imparted by incorporation of amino acid residues with a net charge and/or reducing the proportion of hydrophobic amino acids in the XTEN sequence. The overall net charge and net charge density may be controlled by modifying the content of charged amino acids in the XTEN sequences. In some cases, the net charge density of the XTEN of the compositions may be above +0.1 or below -0.1 charges/residue. In other cases, the net charge of a XTEN can be about 0%, about 1%, about 2%, about 3%, about 4%, about 5%, about 6%, about 7%, about 8%, about 9%, about 10% about 11%, about 12%, about 13%, about 14%, about 15%, about 16%, about 17%, about 18%, about 19%, or about 20% or more.

[0184] Since most tissues and surfaces in a human or animal have a net negative charge, the XTEN sequences can be designed to have a net negative charge to minimize non-specific interactions between the XTEN

containing compositions and various surfaces such as blood vessels, healthy tissues, or various receptors. Not to be bound by a particular theory, the XTEN can adopt open conformations due to electrostatic repulsion between individual amino acids of the XTEN polypeptide that individually carry a high net negative charge and that are distributed across the sequence of the XTEN polypeptide. Such a distribution of net negative charge in the extended sequence lengths of XTEN can lead to an unstructured conformation that, in turn, can result in an effective increase in hydrodynamic radius. Accordingly, in one embodiment the invention provides XTEN in which the XTEN sequences contain about 8, 10, 15, 20, 25, or even about 30% glutamic acid. The XTEN of the compositions of the present invention generally have no or a low content of positively charged amino acids. In some cases the XTEN may have less than about 10% amino acid residues with a positive charge, or less than about 7%, or less than about 5%, or less than about 2% amino acid residues with a positive charge. However, the invention contemplates constructs where a limited number of amino acids with a positive charge, such as lysine, may be incorporated into XTEN to permit conjugation between the epsilon amine of the lysine and a reactive group on a peptide, a linker bridge, or a reactive group on a drug or small molecule to be conjugated to the XTEN backbone. In the foregoing, a fusion proteins can be constructed that comprises XTEN, a biologically active protein, plus a chemotherapeutic agent useful in the treatment of metabolic diseases or disorders, wherein the maximum number of molecules of the agent incorporated into the XTEN component is determined by the numbers of lysines or other amino acids with reactive side chains (e.g., cysteine) incorporated into the XTEN.

[0185] In some cases, an XTEN sequence may comprise charged residues separated by other residues such as serine or glycine, which may lead to better expression or purification behavior. Based on the net charge, XTENs of the subject compositions may have an isoelectric point (pI) of 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, or even 6.5. In preferred embodiments, the XTEN will have an isoelectric point between 1.5 and 4.5. In these embodiments, the XTEN incorporated into the BPXTEN fusion protein compositions of the present invention would carry a net negative charge under physiologic conditions that may contribute to the unstructured conformation and reduced binding of the XTEN component to mammalian proteins and tissues.

[0186] As hydrophobic amino acids can impart structure to a polypeptide, the invention provides that the content of hydrophobic amino acids in the XTEN will typically be less than 5%, or less than 2%, or less than 1% hydrophobic amino acid content. In one embodiment, the amino acid content of methionine and tryptophan in the XTEN component of a BPXTEN fusion protein is typically less than 5%, or less than 2%, and most preferably less than 1%. In some embodiments, the XTEN will have a sequence that has less than 10% amino acid residues with a positive charge, or less than about 7%, or less that about 5%, or less than about 2% amino acid residues with a positive charge, the sum of methionine and tryptophan residues will be less than 2%, and the sum of asparagine and glutamine residues will be less than 10% of the total XTEN sequence.

Increased Hydrodynamic Radius

[0187] In some embodiments, the XTEN can have a high hydrodynamic radius, conferring a corresponding increased Apparent Molecular Weight to the BPXTEN fusion protein which incorporates the XTEN. The linking of XTEN to BP sequences can result in BPXTEN compositions that can have increased hydrodynamic radii, increased Apparent Molecular Weight, and increased Apparent Molecular Weight Factor compared to a BP not linked to an XTEN. For example, in therapeutic applications in which prolonged half-life is desired, compositions in which a XTEN with a high hydrodynamic radius is incorporated into a fusion protein comprising one or more BP can effectively enlarge the hydrodynamic radius of the composition beyond the glomerular pore size of approximately 3-5 nm (corresponding to an apparent molecular weight of about 70 kDa) (Caliceti. 2003. Pharmacokinetic and biodistribution properties of poly(ethylene glycol)-protein conjugates. Adv. Drug Deliv. Rev. 55:1261-1277), resulting in reduced renal clearance of circulating proteins. The hydrodynamic radius of a protein is determined by its molecular weight as well as by its structure, including shape and compactness. Not to be bound by a particular theory, the XTEN can adopt open conformations due to electrostatic repulsion between individual charges of the peptide or the inherent flexibility imparted by the particular amino acids in the sequence that lack potential to confer secondary structure. The open, extended and unstructured conformation of the XTEN polypeptide can have a greater proportional hydrodynamic radius compared to polypeptides of a comparable sequence length and/or molecular weight that have secondary and/or tertiary structure, such as typical globular proteins. Methods for determining the hydrodynamic radius are well known in the art, such as by the use of size exclusion chromatography (SEC), as described in U.S. Patent Nos. 6,406,632 and 7,294,513. The addition of increasing lengths of XTEN results in proportional increases in the parameters of hydrodynamic radius, Apparent Molecular Weight, and Apparent Molecular Weight Factor, permitting the tailoring of BPXTEN to desired characteristic cut-off Apparent Molecular Weights or hydrodynamic radii. Accordingly, in certain embodiments, the BPXTEN fusion protein can be configured with an XTEN such that the fusion protein can have a hydrodynamic radius of at least about 5 nm, or at least about 8 nm, or at least about 10 nm, or 12 nm, or at least about 15 nm. In the foregoing embodiments, the large hydrodynamic radius conferred by the XTEN in an BPXTEN fusion protein can lead to reduced renal clearance of the resulting fusion protein, leading to a corresponding increase in terminal half-life, an increase in mean residence time, and/or a decrease in renal clearance rate.

[0188] In some embodiments, an XTEN of a chosen length and sequence can be selectively incorporated into a BPXTEN to create a fusion protein that will have, under physiologic conditions, an Apparent Molecular Weight of at least about 150 kDa, or at least about 300 kDa, or at least about 400 kDa, or at least about 500 kDa, or at least about 600 kDa, or at least about 700 kDa, or at least about 800 kDa, or at least about 900 kDa, or at least about 1000 kDa, or at least about 1200 kDa, or at least about 1500 kDa, or at least about 1800 kDa,

or at least about 2000 kDa, or at least about 2300 kDa or more. In some embodiments, an XTEN of a chosen length and sequence can be selectively linked to a BP to result in a BPXTEN fusion protein that has, under physiologic conditions, an Apparent Molecular Weight Factor of at least three, alternatively of at least four, alternatively of at least five, alternatively of at least six, alternatively of at least seven, alternatively of at least eight, alternatively of at least nine, alternatively of at least 10, alternatively of at least 15, or an Apparent Molecular Weight Factor of at least 20 or greater. In some embodiments, the BPXTEN fusion protein has, under physiologic conditions, an Apparent Molecular Weight Factor that is about 4 to about 20, or is about 6 to about 15, or is about 8 to about 12, or is about 9 to about 10 relative to the actual molecular weight of the fusion protein. In some embodiments, the (fusion) polypeptide exhibits an apparent molecular weight factor under physiological conditions that is greater than about 6.

Increased Terminal Half-Life

[0189] In some embodiments, the (fusion) polypeptide has a terminal half-life that is at least two-fold longer, or at least three-fold longer, or at least four-fold longer, or at least five-fold longer, compared to the biologically active polypeptide not linked to any XTEN. In some embodiments, the (fusion) polypeptide has a terminal half-life that is at least two-fold longer compared to the biologically active polypeptide not linked to any XTEN.

[0190] Administration of a therapeutically effective dose of any of the embodiments of BPXTEN fusion proteins described herein to a subject in need thereof can result in a gain in time of at least two-fold, or at least three-fold, or at least four-fold, or at least five-fold or more spent within a therapeutic window for the fusion protein compared to the corresponding BP not linked to the XTEN of and administered at a comparable dose to a subject.

Low Immunogenicity

[0191] In some embodiments, the invention provides compositions in which the XTEN sequences have a low degree of immunogenicity or are substantially non-immunogenic. Several factors can contribute to the low immunogenicity of XTEN, e.g., the substantially non-repetitive sequence, the unstructured conformation, the high degree of solubility, the low degree or lack of self-aggregation, the low degree or lack of proteolytic sites within the sequence, and the low degree or lack of epitopes in the XTEN sequence.

[0192] One of ordinary skill in the art will understand that, in general, a polypeptide having highly repetitive short amino acid sequences (e.g., wherein a 200 amino acid-long sequence contain on average 20 repeats or more of a limited set of 3- or 4-mers) and/or having contiguous repetitive amino acid residues (e.g., wherein 5- or 6-mer sequences have identical amino acid residues) have a tendency to aggregate or form higher order structures or form contacts resulting in crystalline or pseudo-crystalline structures.

[0193] In some embodiments, the XTEN sequence is substantially non-repetitive, wherein (1) the XTEN sequence has no three contiguous amino acids that are identical amino acid types, unless the amino acid is

serine, in which case no more than three contiguous amino acids can be serine residues; and wherein (2) the XTEN contains no 3-amino acid sequences (3-mers) that occur more than 16, more than 14, more than 12, or more than 10 times within a 200 amino acid-long sequence of the XTEN. One of ordinary skill in the art will understand that such substantially non-repetitive sequences have less tendency to aggregate and, thus, enable the design of long-sequence XTENs with a relatively low frequency of charged amino acids that would be likely to aggregate if the sequences or amino acid residues were otherwise more repetitive.

[0194] Conformational epitopes are formed by regions of the protein surface that are composed of multiple discontinuous amino acid sequences of the protein antigen. The precise folding of the protein brings these sequences into a well-defined, stable spatial configurations, or epitopes, that can be recognized as "foreign" by the host humoral immune system, resulting in the production of antibodies to the protein or triggering a cell-mediated immune response. In the latter case, the immune response to a protein in an individual is heavily influenced by T-cell epitope recognition that is a function of the peptide binding specificity of that individual's HLA-DR allotype. Engagement of an MHC Class II peptide complex by a cognate T-cell receptor on the surface of the T-cell, together with the cross-binding of certain other co-receptors such as the CD4 molecule, can induce an activated state within the T-cell. Activation leads to the release of cytokines further activating other lymphocytes such as B cells to produce antibodies or activating T killer cells as a full cellular immune response.

[0195] The ability of a peptide to bind a given MHC Class II molecule for presentation on the surface of an APC (antigen presenting cell) is dependent on a number of factors; most notably its primary sequence. In one embodiment, a lower degree of immunogenicity may be achieved by designing XTEN sequences that resist antigen processing in antigen presenting cells, and/or choosing sequences that do not bind MHC receptors well. The invention provides BPXTEN fusion proteins with substantially non-repetitive XTEN polypeptides designed to reduce binding with MHC II receptors, as well as avoiding formation of epitopes for T-cell receptor or antibody binding, resulting in a low degree of immunogenicity. Avoidance of immunogenicity is, in part, a direct result of the conformational flexibility of XTEN sequences; i.e., the lack of secondary structure due to the selection and order of amino acid residues. For example, of particular interest are sequences having a low tendency to adapt compactly folded conformations in aqueous solution or under physiologic conditions that could result in conformational epitopes. The administration of fusion proteins comprising XTEN, using conventional therapeutic practices and dosing, would generally not result in the formation of neutralizing antibodies to the XTEN sequence, and may also reduce the immunogenicity of the BP fusion partner in the BPXTEN compositions.

[0196] In one embodiment, the XTEN sequences utilized in the subject fusion proteins can be substantially free of epitopes recognized by human T cells. The elimination of such epitopes for the purpose of generating

less immunogenic proteins has been disclosed previously; see for example WO 98/52976, WO 02/079232, and WO 00/3317 which are incorporated by reference herein. Assays for human T cell epitopes have been described (Stickler, M., *et al.* (2003) *J Immunol Methods*, 281: 95-108). Of particular interest are peptide sequences that can be oligomerized without generating T cell epitopes or non-human sequences. This can be achieved by testing direct repeats of these sequences for the presence of T-cell epitopes and for the occurrence of 6 to 15-mer and, in particular, 9-mer sequences that are not human, and then altering the design of the XTEN sequence to eliminate or disrupt the epitope sequence. In some cases, the XTEN sequences are substantially non-immunogenic by the restriction of the numbers of epitopes of the XTEN predicted to bind MHC receptors. With a reduction in the numbers of epitopes capable of binding to MHC receptors, there is a concomitant reduction in the potential for T cell activation as well as T cell helper function, reduced B cell activation or upregulation and reduced antibody production. The low degree of predicted T-cell epitopes can be determined by epitope prediction algorithms such as, e.g., TEPITOPE (Sturniolo, T., *et al.* (1999) Nat Biotechnol, 17: 555-61), as shown in **Example 74** of International Patent Application Publication No. WO 2010/144502 A2, which is incorporated by reference in its entirety. The TEPITOPE score of a given peptide frame within a protein is the log of the K_d (dissociation constant, affinity, off-rate) of the binding of that peptide frame to multiple of the most common human MHC alleles, as disclosed in Sturniolo, T. *et al.* (1999) *Nature Biotechnology* 17:555). The score ranges over at least 20 logs, from about 10 to about -10 (corresponding to binding constraints of $10e^{10}$ K_d to $10e^{-10}$ K_d), and can be reduced by avoiding hydrophobic amino acids that can serve as anchor residues during peptide display on MHC, such as M, I, L, V, or F. In some embodiments, an XTEN component incorporated into a BPXTEN does not have a predicted T-cell epitope at a TEPITOPE score of about -5 or greater, or -6 or greater, or -7 or greater, or -8 or greater, or at a TEPITOPE score of -9 or greater. As used herein, a score of "-9 or greater" would encompass TEPITOPE scores of 10 to -9, inclusive, but would not encompass a score of -10, as -10 is less than -9.

[0197] In some embodiments, the inventive XTEN sequences, including those incorporated into the subject BPXTEN fusion proteins, can be rendered substantially non-immunogenic by the restriction of known proteolytic sites from the sequence of the XTEN, reducing the processing of XTEN into small peptides that can bind to MHC II receptors. In some embodiments, the XTEN sequence can be rendered substantially non-immunogenic by the use a sequence that is substantially devoid of secondary structure, conferring resistance to many proteases due to the high entropy of the structure. Accordingly, the reduced TEPITOPE score and elimination of known proteolytic sites from the XTEN may render the XTEN compositions, including the XTEN of the BPXTEN fusion protein compositions, substantially unable to be bound by mammalian receptors, including those of the immune system. In one embodiment, an XTEN of a BPXTEN fusion protein can

have >100 nM K_d binding to a mammalian receptor, or greater than 500 nM K_d, or greater than 1 μM K_d towards a mammalian cell surface or circulating polypeptide receptor.

[0198] Additionally, the substantially non-repetitive sequence and corresponding lack of epitopes of such embodiments of XTEN can limit the ability of B cells to bind to or be activated by XTEN. While an XTEN can make contacts with many different B cells over its extended sequence, each individual B cell may only make one or a small number of contacts with an individual XTEN. As a result, XTENs typically may have a much lower tendency to stimulate proliferation of B cells and thus an immune response. In one embodiment, the BPXTEN may have reduced immunogenicity as compared to the corresponding BP that is not fused. In one embodiment, the administration of up to three parenteral doses of a BPXTEN to a mammal may result in detectable anti-BPXTEN IgG at a serum dilution of 1:100 but not at a dilution of 1:1000. In some embodiments, the administration of up to three parenteral doses of an BPXTEN to a mammal may result in detectable anti-BP IgG at a serum dilution of 1:100 but not at a dilution of 1:1000. In some embodiments, the administration of up to three parenteral doses of an BPXTEN to a mammal may result in detectable anti-XTEN IgG at a serum dilution of 1:100 but not at a dilution of 1:1000. In the foregoing embodiments, the mammal can be a mouse, a rat, a rabbit, or a cynomolgus monkey.

[0199] An additional feature of certain embodiments of XTENs with substantially non-repetitive sequences relative to those less non-repetitive sequences (such as one having three contiguous amino acids that are identical) can be that non-repetitive XTENs form weaker contacts with antibodies (e.g. monovalent interactions), thereby resulting in less likelihood of immune clearance such that the BPXTEN compositions can remain in circulation for an increased period of time.

[0200] In some embodiments, the (fusion) polypeptide is less immunogenic compared to the biologically active polypeptide not linked to any XTEN, wherein immunogenicity is ascertained by measuring production of IgG antibodies that selectively bind to the biologically active polypeptide after administration of comparable doses to a subject.

SPACERS & BP RELEASE SEGMENT

[0201] In some embodiments, at least a portion of the biological activity of the respective BP is retained by the intact BPXTEN. In some embodiments, the BP component either becomes biologically active or has an increase in biological activity upon its release from the XTEN by cleavage of an optional cleavage sequence incorporated within spacer sequences into the BPXTEN, as described more fully hereinbelow.

[0202] Any spacer sequence group is optional in the fusion proteins encompassed by the invention. The spacer may be provided to enhance expression of the fusion protein from a host cell or to decrease steric hindrance such that the BP component may assume its desired tertiary structure and/or interact appropriately with its target molecule. For spacers and methods of identifying desirable spacers, see, for example, George, *et al.* (2003)

Protein Engineering 15:871-879, specifically incorporated by reference herein. In one embodiment, the spacer comprises one or more peptide sequences that are between 1 to 50 amino acid residues in length, or about 1 to 25 residues, or about 1 to 10 residues in length. Spacer sequences, exclusive of cleavage sites, can comprise any of the 20 natural L amino acids, and will preferably comprise hydrophilic amino acids that are sterically unhindered that can include, but not be limited to, glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P). In some embodiments, the spacer can be polyglycines or polyalanines, or predominately a mixture of combinations of glycine and alanine residues. The spacer polypeptide exclusive of a cleavage sequence is largely to substantially devoid of secondary structure. In one embodiment, one or both spacer sequences in a BPXTEN fusion protein composition may each further contain a cleavage sequence, which may be identical or may be different, wherein the cleavage sequence may be acted on by a protease to release the BP from the fusion protein.

[0203] In some cases, the incorporation of the cleavage sequence into the BPXTEN is designed to permit release of a BP that becomes active or more active upon its release from the XTEN. The cleavage sequences are located sufficiently close to the BP sequences, generally within 18, or within 12, or within 6, or within 2 amino acids of the BP sequence terminus, such that any remaining residues attached to the BP after cleavage do not appreciably interfere with the activity (e.g., such as binding to a receptor) of the BP, yet provide sufficient access to the protease to be able to effect cleavage of the cleavage sequence. In some embodiments, the cleavage site is a sequence that can be cleaved by a protease endogenous to the mammalian subject such that the BPXTEN can be cleaved after administration to a subject. In such cases, the BPXTEN can serve as a prodrug or a circulating depot for the BP. Examples of cleavage sites contemplated by the invention include, but are not limited to, a polypeptide sequence cleavable by a mammalian endogenous protease that is FXIa, FXIIa, kallikrein, FVIIa, FIXa, FXa, FIIa (thrombin), Elastase-2, granzyme B, MMP-12, MMP-13, MMP-17 or MMP-20, or by non-mammalian proteases such as TEV, enterokinase, PreScission™ protease (rhinovirus 3C protease), or sortase A. Sequences known to be cleaved by the foregoing proteases are known in the art. Exemplary cleavage sequences and cut sites within the sequences are presented in **Table 7a**, as well as sequence variants. For example, thrombin (activated clotting factor II) acts on the sequence LTPRSLLV (SEQ ID NO: 222) [Rawlings N.D., et al. (2008) *Nucleic Acids Res.*, **36**: D320], which would be cut after the arginine at position 4 in the sequence. Active FIIa is produced by cleavage of FII by FXa in the presence of phospholipids and calcium and is downstream from factor IX in the coagulation pathway. Once activated its natural role in coagulation is to cleave fibrinogen, which then in turn, begins clot formation. FIIa activity is tightly controlled and only occurs when coagulation is necessary for proper hemostasis. However, as coagulation is an on-going process in mammals, by incorporation of the LTPRSLLV (SEQ ID NO: 222) sequence into the BPXTEN between the BP and the XTEN, the XTEN domain would be removed from the adjoining BP concurrent with

activation of either the extrinsic or intrinsic coagulation pathways when coagulation is required physiologically, thereby releasing BP over time. Similarly, incorporation of other sequences into BPXTEN that are acted upon by endogenous proteases would provide for sustained release of BP that may, in certain cases, provide a higher degree of activity for the BP from the "prodrug" form of the BPXTEN.

[0204] In some cases, only the two or three amino acids flanking both sides of the cut site (four to six amino acids total) would be incorporated into the cleavage sequence. In other cases, the known cleavage sequence can have one or more deletions or insertions or one or two or three amino acid substitutions for any one or two or three amino acids in the known sequence, wherein the deletions, insertions or substitutions result in reduced or enhanced susceptibility but not an absence of susceptibility to the protease, resulting in an ability to tailor the rate of release of the BP from the XTEN. Exemplary substitutions are shown in Table 7a.

Table 7a: Protease Cleavage Sequences for BP Release

Protease Acting Upon Sequence	SEQ ID NO	Exemplary Cleavage Sequence	SEQ ID NO	Minimal Cut Site*
FXIa	224	KLTR↓VVGG	244	KD/FL/T/R↓VA/VE/GT/GV
FXIIa	225	TMTR↓IVGG	NA	NA
Kallikrein	226	SPFR↓STGG	245	-/-/FL/RY↓SR/RT/-/-
FVIIa	227	LQVR↓IVGG	NA	NA
FIXa	228	PLGR↓IVGG	247	-/-/G/R↓-/-/-/-
FXa	229	IEGR↓TVGG	248	IA/E/GFP/R↓STI/VFS/-/G
FIIa (thrombin)	230	LTPR↓SLLV	249	-/-/PLA/R↓SAG/-/-/-
Elastase-2	231	LGPV↓SGVP	250	-/-/-/VIAT↓-/-/-/-
Granzyme-B	232	VAGD↓SLEE	251	V/-/-/D↓-/-/-/-
MMP-12	233	GPAG↓LGGA	241	G/PA/-/G↓L/-/G/- (SEQ ID NO: 241)
MMP-13	234	GPAG↓LRGA	242	G/P/-/G↓L/-/GA/- (SEQ ID NO: 242)
MMP-17	235	APLG↓LRLR	252	-/PS/-/-↓LQ/-/LT/-
MMP-20	236	PALP↓LVAQ	NA	NA
TEV	237	ENLYFQ↓G	243	ENLYFQ↓/GS
Enterokinase	238	DDDK↓IVGG	238	DDDK↓IVGG
Protease 3C (PreScission™)	239	LEVLFQ↓GP	239	LEVLFQ↓GP
Sortase A	240	LPKT↓GSES	246	L/P/KEAD/T↓G/-/EKS/S

↓ indicates cleavage site;
NA: not applicable;
* the listing of multiple amino acids before, between, or after a slash indicate alternative amino acids that can be substituted at the position;
"-" indicates that any amino acid may be substituted for the corresponding amino acid indicated in the middle column

[0205] In some embodiments, the BPXTEN fusion protein can comprise spacer sequences that can further comprise one or more cleavage sequences configured to release the BP from the fusion protein when acted on

by a protease. In some embodiments, the one or more cleavage sequences can be a sequence having at least about 80% (e.g., at least about 85%, at least about 90%, at least about 95%, at least about 96%, at least about 97%, at least about 98%, at least about 99%, or 100%) sequence identify to a sequence from **Table 7a**.

[0206] In some embodiments, the disclosure provides BP release segment peptides (or release segment (RS)) that are substrates for one or more mammalian proteases associated with or produced by disease tissues or cells found in proximity to disease tissues. Such proteases can include, but not be limited to the classes of proteases such as metalloproteinases, cysteine proteases, aspartate proteases, and serine proteases, including, but not limited to, the proteases of **Table 7b**. The RS are useful for, amongst other things, incorporation into the subject recombinant polypeptides, conferring a prodrug format that can be activated by the cleavage of the RS by mammalian proteases. As described herein, the RS are incorporated into the subject recombinant polypeptide compositions, linking the incorporated binding moieties to the XTEN (the configurations of which are described more fully, below) such that upon cleavage of the RS by action of the one or more proteases for which the RS are substrates, the binding moieties and XTEN are released from the composition and the binding moieties, no longer shielded by the XTEN, regain their full potential to bind their ligands. In those recombinant polypeptide compositions comprising a first and a second antibody fragment, the compositions are also referred to herein as activatable antibody compositions (AAC).

Table 7b: Proteases of Target Tissues

Class of Proteases	Protease
Metalloproteinases	Meprin
	Neprilysin (CD10)
	PSMA
	BMP-1
	A disintegrin and metalloproteinases (ADAMs)
	ADAM8
	ADAM9
	ADAM10
	ADAM12
	ADAM15
	ADAM17 (TACE)
	ADAM19
	ADAM28 (MDC-L)
	ADAM with thrombospondin motifs (ADAMTS)
	ADAMTS1
	ADAMTS4
	ADAMTS5
	Matrix Metalloproteinases (MMPs)
	MMP-1 (Collagenase 1)
	MMP-2 (Gelatinase A)
	MMP-3 (m1)
	MMP-7 (Matrilysin 1)
	MMP-8 (Collagenase 2)
	MMP-9 (Gelatinase B)

Class of Proteases	Protease
	MMP-10 (Stromelysin 2)
	MMP-11(Stromelysin 3)
	MMP-12 (Macrophage elastase)
	MMP-13 (Collagenase 3)
	MMP-14 (MT1-MMP)
	MMP-15 (MT2-MMP)
	MMP-19
	MMP-23 (CA-MMP)
	MMP-24 (MT5-MMP)
	MMP-26 (Matrilysin 2)
	MMP-27 (CMMP)
Cysteine Proteases	Legumain
	Cysteine cathepsins
	Cathepsin B
	Cathepsin C
	Cathepsin K
	Cathepsin L
	Cathepsin S
	Cathepsin X
Aspartate Proteases	Cathepsin D
	Cathepsin E
	Secretase
Serine Proteases	Urokinase (uPA)
	Tissue-type plasminogen activator (tPA)
	Plasmin
	Thrombin
	Prostate-specific antigen (PSA, KLK3)
	Human neutrophil elastase (HNE)
	Elastase
	Tryptase
	Type II transmembrane serine proteases (TTSPs)
	DESC1
	Hepsin (HPN)
	Matriptase
	Matriptase-2
	TMPRSS2
	TMPRSS3
	TMPRSS4 (CAP2)
	Fibroblast Activation Protein (FAP)
	kallikrein-related peptidase (KLK family)
	KLK4
	KLK5
	KLK6
	KLK7
	KLK8
	KLK10
	KLK11
	KLK13
	KLK14

[0207] In one embodiment, the disclosure provides activatable recombinant polypeptides comprising a first release segment (RS1) sequence having at least 88%, or at least 94%, or 100% sequence identity, when optimally aligned, to a sequence identified herein by the sequences set forth in **Table 8a**, wherein the RS1 is a substrate for one or more mammalian proteases. In other embodiments, the disclosure provides activatable recombinant polypeptides comprising a RS1 and a second release segment (RS2) sequence, each having at least 88%, or at least 94%, or 100% sequence identity, when optimally aligned, to a sequence identified herein by the sequences set forth in **Table 8a**, wherein the RS1 and the RS2 each are a substrate for one or more mammalian proteases. In some embodiments, disclosure provides activatable recombinant polypeptides comprising a first RS (RS1) sequence having at least 90%, at least 93%, at least 97%, or 100% identity, when optimally aligned, to a sequence identified herein by the sequences set forth in **Table 8b**, wherein the RS is a substrate for one or more mammalian proteases. In other embodiments, the disclosure provides activatable recombinant polypeptides comprising a RS1 and a second release segment (RS2) sequence, each having at least 88%, or at least 94%, or 100% sequence identity, when optimally aligned, to a sequence identified herein by the sequences set forth in **Table 8b**, wherein the RS1 and the RS2 are each a substrate for one or more mammalian proteases (e.g., at one, two, or three cleavage sites within each release segment sequence). In the embodiments of activatable recombinant polypeptides comprising RS1 and RS2, the two release segments can be identical or the sequences can be different.

[0208] The present disclosure contemplates release segments that are substrates for one, two or three different classes of proteases that are metalloproteinases, cysteine proteases, aspartate proteases, or serine proteases, including the proteases of **Table 7b**. In a particular feature, the RS serve as substrates for proteases found in close association with or are co-localized with disease tissues or cells, such as but not limited to tumors, cancer cells, and inflammatory tissues, and upon cleavage of the RS, the binding moieties that are otherwise shielded by the XTEN of the subject recombinant polypeptide compositions (and thus have a lower binding affinity for their respective ligands) are released from the composition and regain their full potential to bind the target and/or effector cell ligands. In some embodiments, the RS of the subject recombinant polypeptide compositions comprises an amino acid sequence that is a substrate for a cellular protease located within a targeted cell, including but not limited to the proteases of **Table 7b**. In another particular feature of the subject recombinant polypeptide compositions, the RS that are substrates for two or three classes of proteases were designed with sequences that are capable of being cleaved in different locations of the RS sequence by the different proteases. Thus, the RS that are substrates for two, three, or more classes of proteases have two, three, or a plurality of distinct cleavage sites in the RS sequence, but cleavage by a single protease nevertheless results in the release of the binding moieties and the XTEN from the recombinant polypeptide composition comprising the RS.

[0209] In one embodiment, the RS of the disclosure for incorporation into the subject recombinant polypeptide compositions is a substrate for one or more proteases including but not limited to meprin, neprilysin (CD10), PSMA, BMP-1, A disintegrin and metalloproteinases (ADAMs), ADAM8, ADAM9, ADAM10, ADAM12, ADAM15, ADAM17 (TACE), ADAM19, ADAM28 (MDC-L), ADAM with thrombospondin motifs (ADAMTS), ADAMTS1, ADAMTS4, ADAMTS5, MMP-1 (collagenase 1), matrix metalloproteinase-1 (MMP-1), matrix metalloproteinase-2 (MMP-2, gelatinase A), matrix metalloproteinase-3 (MMP-3, stromelysin 1), matrix metalloproteinase-7 (MMP-7, Matrilysin 1), matrix metalloproteinase-8 (MMP-8, collagenase 2), matrix metalloproteinase-9 (MMP-9, gelatinase B), matrix metalloproteinase-10 (MMP-10, stromelysin 2), matrix metalloproteinase-11 (MMP-11, stromelysin 3), matrix metalloproteinase-12 (MMP-12, macrophage elastase), matrix metalloproteinase-13 (MMP-13, collagenase 3), matrix metalloproteinase-14 (MMP-14, MT1-MMP), matrix metalloproteinase-15 (MMP-15, MT2-MMP), matrix metalloproteinase-19 (MMP-19), matrix metalloproteinase-23 (MMP-23, CA-MMP), matrix metalloproteinase-24 (MMP-24, MT5-MMP), matrix metalloproteinase-26 (MMP-26, matrilysin 2), matrix metalloproteinase-27 (MMP-27, CMMP), legumain, cathepsin B, cathepsin C, cathepsin K, cathepsin L, cathepsin S, cathepsin X, cathepsin D, cathepsin E, secretase, urokinase (uPA), tissue-type plasminogen activator (tPA), plasmin, thrombin, prostate-specific antigen (PSA, KLK3), human neutrophil elastase (HNE), elastase, tryptase, Type II transmembrane serine proteases (TTSPs), DESC1, hepsin (HPN), matriptase, matriptase-2, TMPRSS2, TMPRSS3, TMPRSS4 (CAP2), fibroblast activation protein (FAP), kallikrein-related peptidase (KLK family), KLK4, KLK5, KLK6, KLK7, KLK8, KLK10, KLK11, KLK13, and KLK14. In one embodiment, the RS is a substrate for ADAM17. In one embodiment, the RS is a substrate for BMP-1. In one embodiment, the RS is a substrate for cathepsin. In one embodiment, the RS is a substrate for HtrA1. In one embodiment, the RS is a substrate for legumain. In one embodiment, the RS is a substrate for MMP-1. In one embodiment, the RS is a substrate for MMP-2. In one embodiment, the RS is a substrate for MMP-7. In one embodiment, the RS is a substrate for MMP-9. In one embodiment, the RS is a substrate for MMP-11. In one embodiment, the RS is a substrate for MMP-14. In one embodiment, the RS is a substrate for uPA. In one embodiment, the RS is a substrate for matriptase. In one embodiment, the RS is a substrate for MT-SP1. In one embodiment, the RS is a substrate for neutrophil elastase. In one embodiment, the RS is a substrate for thrombin. In one embodiment RS is a substrate for TMPRSS3. In one embodiment, the RS is a substrate for TMPRSS4. In one embodiment, the RS of the subject recombinant polypeptide compositions is a substrate for at least two proteases including but not limited to legumain, MMP-1, MMP-2, MMP-7, MMP-9, MMP-11, MMP-14, uPA, and matriptase. In some embodiments, the RS of the subject recombinant polypeptide compositions is a substrate for legumain, MMP-1, MMP-2, MMP-7, MMP-9, MMP-11, MMP-14, uPA, and matriptase.

Table 8a: BP Release Segment Sequences.

Name	Construct ID	Amino Acid Sequence	SEQ ID NO
RSR-1517	AC1611	EAGRSANHEPLGLVAT	7001
BSRS-A1-1	AC1605	ASGRSTNAGPSGLAGP	7002
BSRS-A2-1	AC1606	ASGRSTNAGPQGLAGQ	7003
BSRS-A3-1	AC1607	ASGRSTNAGPPGLTGP	7004
VP-1	AC1608	ASSRGTNAGPAGLTGP	7005
RSR-1752	AC1609	ASSRTTNTGPSTLTGP	7006
RSR-1512	AC1610	AAGRSDNGTPLELVAP	7007
RSR-1517	AC1611	EAGRSANHEPLGLVAT	7008
VP-2	AC1612	ASGRGTNAGPAGLTGP	7009
RSR-1018	AC1613	LFGRNDNHEPLELGGG	7010
RSR-1053	AC1614	TAGRSDNLEPLGLVFG	7011
RSR-1059	AC1615	LDGRSDNFHPPELVAG	7012
RSR-1065	AC1616	LEGRSDNEEPENLVAG	7013
RSR-1167	AC1617	LKGRSDNNAPLALVAG	7014
RSR-1201	AC1618	VYSRGTNAGPHGLTGR	7015
RSR-1218	AC1619	ANSRGTNKGFAGLIGP	7016
RSR-1226	AC1620	ASSRLTNEAPAGLTIP	7017
RSR-1254	AC1621	DQSRGTNAGPEGLTDP	7018
RSR-1256	AC1622	ESSRGTNIGQGGLTGP	7019
RSR-1261	AC1623	SSSRGTNQDPAGLTIP	7020
RSR-1293	AC1624	ASSRGQNHSPMGLTGP	7021
RSR-1309	AC1625	AYSRGPNAGPAGLEGR	7022
RSR-1326	AC1626	ASERGNNAGPANLTGF	7023
RSR-1345	AC1627	ASHRGTNPKPAILTGP	7024
RSR-1354	AC1628	MSSRRTNANPAQLTGP	7025
RSR-1426	AC1629	GAGRTDNHEPLELGAA	7026
RSR-1478	AC1630	LAGRSENTAPLELTAG	7027
RSR-1479	AC1631	LEGRPDNHEPLALVAS	7028
RSR-1496	AC1632	LSGRSDNEEPLALPAG	7029
RSR-1508	AC1633	EAGRTDNHEPLELSAP	7030
RSR-1513	AC1634	EGGRSDNHGPLELVSG	7031
RSR-1516	AC1635	LSGRSDNEAPLELEAG	7032
RSR-1524	AC1636	LGGRADNHEPPELGAG	7033
RSR-1622	AC1637	PPSRGTNAEPAGLTGE	7034
RSR-1629	AC1638	ASTRGENAGPAGLEAP	7035
RSR-1664	AC1639	ESSRGTNGAPEGLTGP	7036
RSR-1667	AC1640	ASSRATNESPAGLTGE	7037
RSR-1709	AC1641	ASSRGENPPPGGLTGP	7038
RSR-1712	AC1642	AASRGTNTGPAELTGS	7039
RSR-1727	AC1643	AGSRTTNAGPGGLEGP	7040
RSR-1754	AC1644	APSRGENAGPATLTGA	7041
RSR-1819	AC1645	ESGRAANTGPPTLTAP	7042
RSR-1832	AC1646	NPGRAANEGPPGLPGS	7043
RSR-1855	AC1647	ESSRAANLTPPELTGP	7044
RSR-1911	AC1648	ASGRAANETPPGLTGA	7045
RSR-1929	AC1649	NSGRGENLGAPGLTGT	7046
RSR-1951	AC1650	TTGRAANLTPAGLTGP	7047
RSR-2295	AC1761	EAGRSANHTPAGLTGP	7048
RSR-2298	AC1762	ESGRAANTTPAGLTGP	7049
RSR-2038	AC1679	TTGRATEAANLTPAGLTGP	7050

Name	Construct ID	Amino Acid Sequence	SEQ ID NO
RSR-2072	AC1680	TTGRAEEAANLTPAGLTGP	7051
RSR-2089	AC1681	TTGRAGEAANLTPAGLTGP	7052
RSR-2302	AC1682	TTGRATEAANATPAGLTGP	7053
RSR-3047	AC1697	TTGRAGEAEGATSAGATGP	7054
RSR-3052	AC1698	TTGEAGEAANATSAGATGP	7055
RSR-3043	AC1699	TTGEAGEAAGLTPAGLTGP	7056
RSR-3041	AC1700	TTGAAGEAANATPAGLTGP	7057
RSR-3044	AC1701	TTGRAGEAAGLTPAGLTGP	7058
RSR-3057	AC1702	TTGRAGEAANATSAGATGP	7059
RSR-3058	AC1703	TTGEAGEAAGATSAGATGP	7060
RSR-2485	AC1763	ESGRAANTEPPELGAG	7061
RSR-2486	AC1764	ESGRAANTAPEGLTGP	7062
RSR-2488	AC1688	EPGRAANHEPSGLTEG	7063
RSR-2599	AC1706	ESGRAANHTGAPPGGLTGP	7064
RSR-2706	AC1716	TTGRTGEGANATPGGLTGP	7065
RSR-2707	AC1717	RTGRSGEAANETPEGLEGP	7066
RSR-2708	AC1718	RTGRTGESANETPAGLGGP	7067
RSR-2709	AC1719	STGRTGEPANETPAGLSGP	7068
RSR-2710	AC1720	TTGRAGEPANATPTGLSGP	7069
RSR-2711	AC1721	RTGRPGEGANATPTGLPGP	7070
RSR-2712	AC1722	RTGRGGEAANATPSGLGGP	7071
RSR-2713	AC1723	STGRSGESANATPGGLGGP	7072
RSR-2714	AC1724	RTGRTGEEANATPAGLPGP	7073
RSR-2715	AC1725	ATGRPGEPANTTPEGLEGP	7074
RSR-2716	AC1726	STGRSGEPANATPGGLTGP	7075
RSR-2717	AC1727	PTGRGGEGANTTPTGLPGP	7076
RSR-2718	AC1728	PTGRSGEGANATPSGLTGP	7077
RSR-2719	AC1729	TTGRASEGANSTPAPLTEP	7078
RSR-2720	AC1730	TYGRAAEAANTTPAGLTAP	7079
RSR-2721	AC1731	TTGRATEGANATPAELTEP	7080
RSR-2722	AC1732	TVGRASEEANTTPASLTGP	7081
RSR-2723	AC1733	TTGRAPEAANATPAPLTGP	7082
RSR-2724	AC1734	TWGRATEPANATPAPLTSP	7083
RSR-2725	AC1735	TVGRASESANATPAELTSP	7084
RSR-2726	AC1736	TVGRAPEGANSTPAGLTGP	7085
RSR-2727	AC1737	TWGRATEAPNLEPATLTTP	7086
RSR-2728	AC1738	TTGRATEAPNLTPAPLTEP	7087
RSR-2729	AC1739	TQGRATEAPNLSPAALTSP	7088
RSR-2730	AC1740	TQGRAAEAPNLTPATLTAP	7089
RSR-2731	AC1741	TSGRAPEATNLAPAPLTGP	7090
RSR-2732	AC1742	TQGRAAEAANLTPAGLTEP	7091
RSR-2733	AC1743	TTGRAGSAPNLPPTGLTTP	7092
RSR-2734	AC1744	TTGRAGGAENLPPEGLTAP	7093
RSR-2735	AC1745	TTSRAGTATNLTPEGLTAP	7094
RSR-2736	AC1746	TTGRAGTATNLPPSGLTTP	7095
RSR-2737	AC1747	TTARAGEAENLSPSGLTAP	7096
RSR-2738	AC1748	TTGRAGGAGNLAPGGLTEP	7097
RSR-2739	AC1749	TTGRAGTATNLPPEGLTGP	7098
RSR-2740	AC1750	TTGRAGGAANLAPTGLTEP	7099
RSR-2741	AC1751	TTGRAGTAENLAPSGLTTP	7100
RSR-2742	AC1752	TTGRAGSATNLGPGGLTGP	7101

Name	Construct ID	Amino Acid Sequence	SEQ ID NO
RSR-2743	AC1753	TTARAGGAENLTPAGLTEP	7102
RSR-2744	AC1754	TTARAGSAENLSPSGLTGP	7103
RSR-2745	AC1755	TTARAGGAGNLAPEGLTTP	7104
RSR-2746	AC1756	TTSRAGAAENLTPTGLTGP	7105
RSR-2747	AC1757	TYGRTTTPGNEPPASLEAE	7106
RSR-2748	AC1758	TYSRGESGPNEPPPGLTGP	7107
RSR-2749	AC1759	AWGRTGASENETPAPLGGE	7108
RSR-2750	AC1760	RWGRAETTPNTPPEGLETE	7109
RSR-2751	AC1765	ESGRAANHTGAEPPELGAG	7110
RSR-2754	AC1801	TTGRAGEAANLTPAGLTES	7111
RSR-2755	AC1802	TTGRAGEAANLTPAALTES	7112
RSR-2756	AC1803	TTGRAGEAANLTPAPLTES	7113
RSR-2757	AC1804	TTGRAGEAANLTPEPLTES	7114
RSR-2758	AC1805	TTGRAGEAANLTPAGLTGA	7115
RSR-2759	AC1806	TTGRAGEAANLTPEGLTGA	7116
RSR-2760	AC1807	TTGRAGEAANLTPEPLTGA	7117
RSR-2761	AC1808	TTGRAGEAANLTPAGLTEA	7118
RSR-2762	AC1809	TTGRAGEAANLTPEGLTEA	7119
RSR-2763	AC1810	TTGRAGEAANLTPAPLTEA	7120
RSR-2764	AC1811	TTGRAGEAANLTPEPLTEA	7121
RSR-2765	AC1812	TTGRAGEAANLTPEPLTGP	7122
RSR-2766	AC1813	TTGRAGEAANLTPAGLTGG	7123
RSR-2767	AC1814	TTGRAGEAANLTPEGLTGG	7124
RSR-2768	AC1815	TTGRAGEAANLTPEALTGG	7125
RSR-2769	AC1816	TTGRAGEAANLTPEPLTGG	7126
RSR-2770	AC1817	TTGRAGEAANLTPAGLTEG	7127
RSR-2771	AC1818	TTGRAGEAANLTPEGLTEG	7128
RSR-2772	AC1819	TTGRAGEAANLTPAPLTEG	7129
RSR-2773	AC1820	TTGRAGEAANLTPEPLTEG	7130

Table 8b: Release Segment Sequences

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-0001	GSAPGSAGGYAELRMG GAIATSGSETPGT	7131	RSC-0001	GTAEAASASGGSAGGY AELRMGGAIPGSP	7379
RSN-0002	GSAPGTGGGYAPLRMG GGAATSGSETPGT	7132	RSC-0002	GTAEAASASGGTGGGY APLRMGGGAPGSP	7380
RSN-0003	GSAPGAEGGYAALRMG GEIATSGSETPGT	7133	RSC-0003	GTAEAASASGGAEGGY AALRMGGEIPGSP	7381
RSN-0004	GSAPGGPGGYALLRMG GPAATSGSETPGT	7134	RSC-0004	GTAEAASASGGGPGGY ALLRMGGPAPGSP	7382
RSN-0005	GSAPGEAGGYAFLRMG GSIATSGSETPGT	7135	RSC-0005	GTAEAASASGGEAGGY AFLRMGGSIPGSP	7383
RSN-0006	GSAPGPGGGYASLRMG GTAATSGSETPGT	7136	RSC-0006	GTAEAASASGGPGGGY ASLRMGGTAPGSP	7384
RSN-0007	GSAPGSEGGYATLRMG GAIATSGSETPGT	7137	RSC-0007	GTAEAASASGGSEGGY ATLRMGGAIPGSP	7385
RSN-0008	GSAPGTPGGYANLRMG GGAATSGSETPGT	7138	RSC-0008	GTAEAASASGGTPGGY ANLRMGGGAPGSP	7386
RSN-0009	GSAPGASGGYAHLRMG GEIATSGSETPGT	7139	RSC-0009	GTAEAASASGGASGGY AHLRMGGEIPGSP	7387

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-0010	GSAPGGTGGYGELRMG GPAATSGSETPGT	7140	RSC-0010	GTAEAASASGGGTGGY GELRMGGPAPGSP	7388
RSN-0011	GSAPGEAGGYPELRMG GSIATSGSETPGT	7141	RSC-0011	GTAEAASASGGEAGGY PELRMGGSIPGSP	7389
RSN-0012	GSAPGPGGGYVELRMG GTAATSGSETPGT	7142	RSC-0012	GTAEAASASGGPGGGY VELRMGGTAPGSP	7390
RSN-0013	GSAPGSEGGYLELRMG GAIATSGSETPGT	7143	RSC-0013	GTAEAASASGGSEGGY LELRMGGAIPGSP	7391
RSN-0014	GSAPGTPGGYSELRMG GGAATSGSETPGT	7144	RSC-0014	GTAEAASASGGTPGGY SELRMGGGAPGSP	7392
RSN-0015	GSAPGASGGYTELRMG GEIATSGSETPGT	7145	RSC-0015	GTAEAASASGGASGGY TELRMGGEIPGSP	7393
RSN-0016	GSAPGGTGGYQELRMG GPAATSGSETPGT	7146	RSC-0016	GTAEAASASGGGTGGY QELRMGGPAPGSP	7394
RSN-0017	GSAPGEAGGYEELRMG GSIATSGSETPGT	7147	RSC-0017	GTAEAASASGGEAGGY EELRMGGSIPGSP	7395
RSN-0018	GSAPGPGIGPAELRMGG TAATSGSETPGT	7148	RSC-0018	GTAEAASASGGPGIGPA ELRMGGTAPGSP	7396
RSN-0019	GSAPGSEIGAAELRMG GAIATSGSETPGT	7149	RSC-0019	GTAEAASASGGSEIGAA ELRMGGAIPGSP	7397
RSN-0020	GSAPGTPIGSAELRMGG GAATSGSETPGT	7150	RSC-0020	GTAEAASASGGTPIGSA ELRMGGGAPGSP	7398
RSN-0021	GSAPGASIGTAELRMG GEIATSGSETPGT	7151	RSC-0021	GTAEAASASGGASIGTA ELRMGGEIPGSP	7399
RSN-0022	GSAPGGTIGNAELRMG GPAATSGSETPGT	7152	RSC-0022	GTAEAASASGGGTIGN AELRMGGPAPGSP	7400
RSN-0023	GSAPGEAIGQAELRMG GSIATSGSETPGT	7153	RSC-0023	GTAEAASASGGEAIGQ AELRMGGSIPGSP	7401
RSN-0024	GSAPGPGGPYAELRMG GTAATSGSETPGT	7154	RSC-0024	GTAEAASASGGPGGPY AELRMGGTAPGSP	7402
RSN-0025	GSAPGSEGAYAELRMG GAIATSGSETPGT	7155	RSC-0025	GTAEAASASGGSEGAY AELRMGGAIPGSP	7403
RSN-0026	GSAPGTPGVYAELRMG GGAATSGSETPGT	7156	RSC-0026	GTAEAASASGGTPGVY AELRMGGGAPGSP	7404
RSN-0027	GSAPGASGLYAELRMG GEIATSGSETPGT	7157	RSC-0027	GTAEAASASGGASGLY AELRMGGEIPGSP	7405
RSN-0028	GSAPGGTGIYAELRMG GPAATSGSETPGT	7158	RSC-0028	GTAEAASASGGGTGIY AELRMGGPAPGSP	7406
RSN-0029	GSAPGEAGFYAELRMG GSIATSGSETPGT	7159	RSC-0029	GTAEAASASGGEAGFY AELRMGGSIPGSP	7407
RSN-0030	GSAPGPGGYYAELRMG GTAATSGSETPGT	7160	RSC-0030	GTAEAASASGGPGGYY AELRMGGTAPGSP	7408
RSN-0031	GSAPGSEGSYAELRMG GAIATSGSETPGT	7161	RSC-0031	GTAEAASASGGSEGSY AELRMGGAIPGSP	7409
RSN-0032	GSAPGTPGNYAELRMG GGAATSGSETPGT	7162	RSC-0032	GTAEAASASGGTPGNY AELRMGGGAPGSP	7410
RSN-0033	GSAPGASGEYAELRMG GEIATSGSETPGT	7163	RSC-0033	GTAEAASASGGASGEY AELRMGGEIPGSP	7411
RSN-0034	GSAPGGTGHYAELRMG GPAATSGSETPGT	7164	RSC-0034	GTAEAASASGGGTGHY AELRMGGPAPGSP	7412

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-0035	GSAPGEAGGYAEARMG GSIATSGSETPGT	7165	RSC-0035	GTAEAASASGGEAGGY AEARMGGSIPGSP	7413
RSN-0036	GSAPGPGGGYAEVRMG GTAATSGSETPGT	7166	RSC-0036	GTAEAASASGGPGGGY AEVRMGGTAPGSP	7414
RSN-0037	GSAPGSEGGYAEIRMG GAIATSGSETPGT	7167	RSC-0037	GTAEAASASGGSEGGY AEIRMGGAIPGSP	7415
RSN-0038	GSAPGTPGGYAEFRMG GGAATSGSETPGT	7168	RSC-0038	GTAEAASASGGTPGGY AEFRMGGGAPGSP	7416
RSN-0039	GSAPGASGGYAEYRMG GEIATSGSETPGT	7169	RSC-0039	GTAEAASASGGASGGY AEYRMGGEIPGSP	7417
RSN-0040	GSAPGGTGGYAESRMG GPAATSGSETPGT	7170	RSC-0040	GTAEAASASGGGTGGY AESRMGGPAPGSP	7418
RSN-0041	GSAPGEAGGYAETRMG GSIATSGSETPGT	7171	RSC-0041	GTAEAASASGGEAGGY AETRMGGSIPGSP	7419
RSN-0042	GSAPGPGGGYAELAMG GTRATSGSETPGT	7172	RSC-0042	GTAEAASASGGPGGGY AELAMGGTRPGSP	7420
RSN-0043	GSAPGSEGGYAELVMG GARATSGSETPGT	7173	RSC-0043	GTAEAASASGGSEGGY AELVMGGARPGSP	7421
RSN-0044	GSAPGTPGGYAELLMG GGRATSGSETPGT	7174	RSC-0044	GTAEAASASGGTPGGY AELLMGGGRPGSP	7422
RSN-0045	GSAPGASGGYAELIMG GERATSGSETPGT	7175	RSC-0045	GTAEAASASGGASGGY AELIMGGERPGSP	7423
RSN-0046	GSAPGGTGGYAELWM GGPRATSGSETPGT	7176	RSC-0046	GTAEAASASGGGTGGY AELWMGGPRPGSP	7424
RSN-0047	GSAPGEAGGYAELSMG GSRATSGSETPGT	7177	RSC-0047	GTAEAASASGGEAGGY AELSMGGSRPGSP	7425
RSN-0048	GSAPGPGGGYAELTMG GTRATSGSETPGT	7178	RSC-0048	GTAEAASASGGPGGGY AELTMGGTRPGSP	7426
RSN-0049	GSAPGSEGGYAELQMG GARATSGSETPGT	7179	RSC-0049	GTAEAASASGGSEGGY AELQMGGARPGSP	7427
RSN-0050	GSAPGTPGGYAELNMG GGRATSGSETPGT	7180	RSC-0050	GTAEAASASGGTPGGY AELNMGGGRPGSP	7428
RSN-0051	GSAPGASGGYAELEMG GERATSGSETPGT	7181	RSC-0051	GTAEAASASGGASGGY AELEMGGERPGSP	7429
RSN-0052	GSAPGGTGGYAELRPG GPIATSGSETPGT	7182	RSC-0052	GTAEAASASGGGTGGY AELRPGGPIPGSP	7430
RSN-0053	GSAPGEAGGYAELRAG GSAATSGSETPGT	7183	RSC-0053	GTAEAASASGGEAGGY AELRAGGSAPGSP	7431
RSN-0054	GSAPGPGGGYAELRLG GTIATSGSETPGT	7184	RSC-0054	GTAEAASASGGPGGGY AELRLGGTIPGSP	7432
RSN-0055	GSAPGSEGGYAELRIGG AAATSGSETPGT	7185	RSC-0055	GTAEAASASGGSEGGY AELRIGGAAPGSP	7433
RSN-0056	GSAPGTPGGYAELRSG GGIATSGSETPGT	7186	RSC-0056	GTAEAASASGGTPGGY AELRSGGGIPGSP	7434
RSN-0057	GSAPGASGGYAELRNG GEAATSGSETPGT	7187	RSC-0057	GTAEAASASGGASGGY AELRNGGEAPGSP	7435
RSN-0058	GSAPGGTGGYAELRQG GPIATSGSETPGT	7188	RSC-0058	GTAEAASASGGGTGGY AELRQGGPIPGSP	7436
RSN-0059	GSAPGEAGGYAELRDG GSAATSGSETPGT	7189	RSC-0059	GTAEAASASGGEAGGY AELRDGGSAPGSP	7437

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-0060	GSAPGPGGGYAELREG GTIATSGSETPGT	7190	RSC-0060	GTAEAASASGGPGGGY AELREGGTIPGSP	7438
RSN-0061	GSAPGSEGGYAELRHG GAAATSGSETPGT	7191	RSC-0061	GTAEAASASGGSEGGY AELRHGGAAPGSP	7439
RSN-0062	GSAPGTPGGYAELRMP GGIATSGSETPGT	7192	RSC-0062	GTAEAASASGGTPGGY AELRMPGGIPGSP	7440
RSN-0063	GSAPGASGGYAELRMA GEAATSGSETPGT	7193	RSC-0063	GTAEAASASGGASGGY AELRMAGEAPGSP	7441
RSN-0064	GSAPGGTGGYAELRMV GPIATSGSETPGT	7194	RSC-0064	GTAEAASASGGGTGGY AELRMVGPIPGSP	7442
RSN-0065	GSAPGEAGGYAELRML GSAATSGSETPGT	7195	RSC-0065	GTAEAASASGGEAGGY AELRMLGSAPGSP	7443
RSN-0066	GSAPGPGGGYAELRMI GTIATSGSETPGT	7196	RSC-0066	GTAEAASASGGPGGGY AELRMIGTIPGSP	7444
RSN-0067	GSAPGSEGGYAELRMY GAIATSGSETPGT	7197	RSC-0067	GTAEAASASGGSEGGY AELRMYGAIPGSP	7445
RSN-0068	GSAPGTPGGYAELRMS GGAATSGSETPGT	7198	RSC-0068	GTAEAASASGGTPGGY AELRMSGGAPGSP	7446
RSN-0069	GSAPGASGGYAELRMN GEIATSGSETPGT	7199	RSC-0069	GTAEAASASGGASGGY AELRMNGEIPGSP	7447
RSN-0070	GSAPGGTGGYAELRMQ GPAATSGSETPGT	7200	RSC-0070	GTAEAASASGGGTGGY AELRMQGPAPGSP	7448
RSN-0071	GSAPGANHTPAGLTGP GARATSGSETPGT	7201	RSC-0071	GTAEAASASGGANHTP AGLTGPGARPGSP	7449
RSN-0072	GSAPGANTAPEGLTGPS TRATSGSETPGT	7202	RSC-0072	GTAEAASASGGANTAP EGLTGPSTRPGSP	7450
RSN-0073	GSAPGTGAPPGGLTGPG TRATSGSETPGT	7203	RSC-0073	GTAEAASASGGTGAPP GGLTGPGTRPGSP	7451
RSN-0074	GSAPGANHEPSGLTEGS PRATSGSETPGT	7204	RSC-0074	GTAEAASASGGANHEP SGLTEGSPRPGSP	7452
RSN-0075	GSAPGANTEPPELGAGT ERATSGSETPGT	7205	RSC-0075	GTAEAASASGGANTEP PELGAGTERPGSP	7453
RSN-0076	GSAPGASGPPPGLTGPP GRATSGSETPGT	7206	RSC-0076	GTAEAASASGGASGPPP GLTGPPGRPGSP	7454
RSN-0077	GSAPGASGTPAPLGGEP GRATSGSETPGT	7207	RSC-0077	GTAEAASASGGASGTP APLGGEPGRPGSP	7455
RSN-0078	GSAPGPAGPPEGLETEA GRATSGSETPGT	7208	RSC-0078	GTAEAASASGGPAGPPE GLETEAGRPGSP	7456
RSN-0079	GSAPGPTSGQGGLTGPE SRATSGSETPGT	7209	RSC-0079	GTAEAASASGGPTSGQ GGLTGPESRPGSP	7457
RSN-0080	GSAPGSAGGAANLVRG GAIATSGSETPGT	7210	RSC-0080	GTAEAASASGGSAGGA ANLVRGGAIPGSP	7458
RSN-0081	GSAPGTGGGAAPLVRG GGAATSGSETPGT	7211	RSC-0081	GTAEAASASGGTGGGA APLVRGGGAPGSP	7459
RSN-0082	GSAPGAEGGAAALVRG GEIATSGSETPGT	7212	RSC-0082	GTAEAASASGGAEGGA AALVRGGEIPGSP	7460
RSN-0083	GSAPGGPGGAALLVRG GPAATSGSETPGT	7213	RSC-0083	GTAEAASASGGGPGGA ALLVRGGPAPGSP	7461
RSN-0084	GSAPGEAGGAAFLVRG GSIATSGSETPGT	7214	RSC-0084	GTAEAASASGGEAGGA AFLVRGGSIPGSP	7462

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-0085	GSAPGPGGGAASLVRG GTAATSGSETPGT	7215	RSC-0085	GTAEAASASGGPGGGA ASLVRGGTAPGSP	7463
RSN-0086	GSAPGSEGGAATLVRG GAIATSGSETPGT	7216	RSC-0086	GTAEAASASGGSEGGA ATLVRGGAIPGSP	7464
RSN-0087	GSAPGTPGGAAGLVRG GGAATSGSETPGT	7217	RSC-0087	GTAEAASASGGTPGGA AGLVRGGGAPGSP	7465
RSN-0088	GSAPGASGGAADLVRG GEIATSGSETPGT	7218	RSC-0088	GTAEAASASGGASGGA ADLVRGGEIPGSP	7466
RSN-0089	GSAPGGTGGAGNLVRG GPAATSGSETPGT	7219	RSC-0089	GTAEAASASGGGTGGA GNLVRGGPAPGSP	7467
RSN-0090	GSAPGEAGGAPNLVRG GSIATSGSETPGT	7220	RSC-0090	GTAEAASASGGEAGGA PNLVRGGSIPGSP	7468
RSN-0091	GSAPGPGGGAVNLVRG GTAATSGSETPGT	7221	RSC-0091	GTAEAASASGGPGGGA VNLVRGGTAPGSP	7469
RSN-0092	GSAPGSEGGALNLVRG GAIATSGSETPGT	7222	RSC-0092	GTAEAASASGGSEGGA LNLVRGGAIPGSP	7470
RSN-0093	GSAPGTPGGASNLVRG GGAATSGSETPGT	7223	RSC-0093	GTAEAASASGGTPGGA SNLVRGGGAPGSP	7471
RSN-0094	GSAPGASGGATNLVRG GEIATSGSETPGT	7224	RSC-0094	GTAEAASASGGASGGA TNLVRGGEIPGSP	7472
RSN-0095	GSAPGGTGGAQNLVRG GPAATSGSETPGT	7225	RSC-0095	GTAEAASASGGGTGGA QNLVRGGPAPGSP	7473
RSN-0096	GSAPGEAGGAENLVRG GSIATSGSETPGT	7226	RSC-0096	GTAEAASASGGEAGGA ENLVRGGSIPGSP	7474
RSN-1517	GSAPEAGRSANHEPLGL VATATSGSETPGT	7227	RSC-1517	GTAEAASASGEAGRSA NHEPLGLVATPGSP	7475
BSRS-A1-2	GSAPASGRSTNAGPSGL AGPATSGSETPGT	7228	BSRS-A1-3	GTAEAASASGASGRST NAGPSGLAGPPGSP	7476
BSRS-A2-2	GSAPASGRSTNAGPQG LAGQATSGSETPGT	7229	BSRS-A2-3	GTAEAASASGASGRST NAGPQGLAGQPGSP	7477
BSRS-A3-2	GSAPASGRSTNAGPPGL TGPATSGSETPGT	7230	BSRS-A3-3	GTAEAASASGASGRST NAGPPGLTGPPGSP	7478
VP-1	GSAPASSRGTNAGPAG LTGPATSGSETPGT	7231	VP-1	GTAEAASASGASSRGT NAGPAGLTGPPGSP	7479
RSN-1752	GSAPASSRTTNTGPSTL TGPATSGSETPGT	7232	RSC-1752	GTAEAASASGASSRTTN TGPSTLTGPPGSP	7480
RSN-1512	GSAPAAGRSDNGTPLEL VAPATSGSETPGT	7233	RSC-1512	GTAEAASASGAAGRSD NGTPLELVAPPGSP	7481
RSN-1517	GSAPEAGRSANHEPLGL VATATSGSETPGT	7234	RSC-1517	GTAEAASASGEAGRSA NHEPLGLVATPGSP	7482
VP-2	GSAPASGRGTNAGPAG LTGPATSGSETPGT	7235	VP-2	GTAEAASASGASGRGT NAGPAGLTGPPGSP	7483
RSN-1018	GSAPLFGRNDNHEPLEL GGGATSGSETPGT	7236	RSC-1018	GTAEAASASGLFGRND NHEPLELGGGPGSP	7484
RSN-1053	GSAPTAGRSDNLEPLGL VFGATSGSETPGT	7237	RSC-1053	GTAEAASASGTAGRSD NLEPLGLVFGPGSP	7485
RSN-1059	GSAPLDGRSDNFHPPEL VAGATSGSETPGT	7238	RSC-1059	GTAEAASASGLDGRSD NFHPPELVAGPGSP	7486
RSN-1065	GSAPLEGRSDNEEPENL VAGATSGSETPGT	7239	RSC-1065	GTAEAASASGLEGRSD NEEPENLVAGPGSP	7487

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-1167	GSAPLKGRSDNNAPLA LVAGATSGSETPGT	7240	RSC-1167	GTAEAASASGLKGRSD NNAPLALVAGPGSP	7488
RSN-1201	GSAPVYSRGTNAGPHG LTGRATSGSETPGT	7241	RSC-1201	GTAEAASASGVYSRGT NAGPHGLTGRPGSP	7489
RSN-1218	GSAPANSRGTNKGFAG LIGPATSGSETPGT	7242	RSC-1218	GTAEAASASGANSRGT NKGFAGLIGPPGSP	7490
RSN-1226	GSAPASSRLTNEAPAGL TIPATSGSETPGT	7243	RSC-1226	GTAEAASASGASSRLTN EAPAGLTIPPGSP	7491
RSN-1254	GSAPDQSRGTNAGPEG LTDPATSGSETPGT	7244	RSC-1254	GTAEAASASGDQSRGT NAGPEGLTDPPGSP	7492
RSN-1256	GSAPESSRGTNIGQGGL TGPATSGSETPGT	7245	RSC-1256	GTAEAASASGESSRGTN IGQGGLTGPPGSP	7493
RSN-1261	GSAPSSSRGTNQDPAGL TIPATSGSETPGT	7246	RSC-1261	GTAEAASASGSSSRGTN QDPAGLTIPPGSP	7494
RSN-1293	GSAPASSRGQNHSPMG LTGPATSGSETPGT	7247	RSC-1293	GTAEAASASGASSRGQ NHSPMGLTGPPGSP	7495
RSN-1309	GSAPAYSRGPNAGPAG LEGRATSGSETPGT	7248	RSC-1309	GTAEAASASGAYSRGP NAGPAGLEGRPGSP	7496
RSN-1326	GSAPASERGNNAGPAN LTGFATSGSETPGT	7249	RSC-1326	GTAEAASASGASERGN NAGPANLTGFPGSP	7497
RSN-1345	GSAPASHRGTNPKPAIL TGPATSGSETPGT	7250	RSC-1345	GTAEAASASGASHRGT NPKPAILTGPPGSP	7498
RSN-1354	GSAPMSSRRTNANPAQ LTGPATSGSETPGT	7251	RSC-1354	GTAEAASASGMSSRRT NANPAQLTGPPGSP	7499
RSN-1426	GSAPGAGRTDNHEPLE LGAAATSGSETPGT	7252	RSC-1426	GTAEAASASGGAGRTD NHEPLELGAAPGSP	7500
RSN-1478	GSAPLAGRSENTAPLEL TAGATSGSETPGT	7253	RSC-1478	GTAEAASASGLAGRSE NTAPLELTAGPGSP	7501
RSN-1479	GSAPLEGRPDNHEPLAL VASATSGSETPGT	7254	RSC-1479	GTAEAASASGLEGRPD NHEPLALVASPGSP	7502
RSN-1496	GSAPLSGRSDNEEPLAL PAGATSGSETPGT	7255	RSC-1496	GTAEAASASGLSGRSD NEEPLALPAGPGSP	7503
RSN-1508	GSAPEAGRTDNHEPLEL SAPATSGSETPGT	7256	RSC-1508	GTAEAASASGEAGRTD NHEPLELSAPPGSP	7504
RSN-1513	GSAPEGGRSDNHGPLEL VSGATSGSETPGT	7257	RSC-1513	GTAEAASASGEGGRSD NHGPLELVSGPGSP	7505
RSN-1516	GSAPLSGRSDNEAPLEL EAGATSGSETPGT	7258	RSC-1516	GTAEAASASGLSGRSD NEAPLELEAGPGSP	7506
RSN-1524	GSAPLGGRADNHEPPEL GAGATSGSETPGT	7259	RSC-1524	GTAEAASASGLGGRAD NHEPPELGAGPGSP	7507
RSN-1622	GSAPPPSRGTNAEPAGL TGEATSGSETPGT	7260	RSC-1622	GTAEAASASGPPSRGTN AEPAGLTGEPGSP	7508
RSN-1629	GSAPASTRGENAGPAG LEAPATSGSETPGT	7261	RSC-1629	GTAEAASASGASTRGE NAGPAGLEAPPGSP	7509
RSN-1664	GSAPESSRGTNGAPEGL TGPATSGSETPGT	7262	RSC-1664	GTAEAASASGESSRGTN GAPEGLTGPPGSP	7510
RSN-1667	GSAPASSRATNESPAGL TGEATSGSETPGT	7263	RSC-1667	GTAEAASASGASSRAT NESPAGLTGEPGSP	7511
RSN-1709	GSAPASSRGENPPPGGL TGPATSGSETPGT	7264	RSC-1709	GTAEAASASGASSRGE NPPPGGLTGPPGSP	7512

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-1712	GSAPAASRGTNTGPAEL TGSATSGSETPGT	7265	RSC-1712	GTAEAASASGAASRGT NTGPAELTGSPGSP	7513
RSN-1727	GSAPAGSRTTNAGPGG LEGPATSGSETPGT	7266	RSC-1727	GTAEAASASGAGSRTT NAGPGGLEGPPGSP	7514
RSN-1754	GSAPAPSRGENAGPATL TGAATSGSETPGT	7267	RSC-1754	GTAEAASASGAPSRGE NAGPATLTGAPGSP	7515
RSN-1819	GSAPESGRAANTGPPTL TAPATSGSETPGT	7268	RSC-1819	GTAEAASASGESGRAA NTGPPTLTAPPGSP	7516
RSN-1832	GSAPNPGRAANEGPPG LPGSATSGSETPGT	7269	RSC-1832	GTAEAASASGNPGRAA NEGPPGLPGSPGSP	7517
RSN-1855	GSAPESSRAANLTPPEL TGPATSGSETPGT	7270	RSC-1855	GTAEAASASGESSRAA NLTPPELTGPPGSP	7518
RSN-1911	GSAPASGRAANETPPGL TGAATSGSETPGT	7271	RSC-1911	GTAEAASASGASGRAA NETPPGLTGAPGSP	7519
RSN-1929	GSAPNSGRGENLGAPG LTGTATSGSETPGT	7272	RSC-1929	GTAEAASASGNSGRGE NLGAPGLTGTPGSP	7520
RSN-1951	GSAPTTGRAANLTPAG LTGPATSGSETPGT	7273	RSC-1951	GTAEAASASGTTGRAA NLTPAGLTGPPGSP	7521
RSN-2295	GSAPEAGRSANHTPAG LTGPATSGSETPGT	7274	RSC-2295	GTAEAASASGEAGRSA NHTPAGLTGPPGSP	7522
RSN-2298	GSAPESGRAANTTPAGL TGPATSGSETPGT	7275	RSC-2298	GTAEAASASGESGRAA NTTPAGLTGPPGSP	7523
RSN-2038	GSAPTTGRATEAANLTP AGLTGPATSGSETPGT	7276	RSC-2038	GTAEAASASGTTGRAT EAANLTPAGLTGPPGSP	7524
RSN-2072	GSAPTTGRAEEAANLTP AGLTGPATSGSETPGT	7277	RSC-2072	GTAEAASASGTTGRAE EAANLTPAGLTGPPGSP	7525
RSN-2089	GSAPTTGRAGEAANLT PAGLTGPATSGSETPGT	7278	RSC-2089	GTAEAASASGTTGRAG EAANLTPAGLTGPPGSP	7526
RSN-2302	GSAPTTGRATEAANAT PAGLTGPATSGSETPGT	7279	RSC-2302	GTAEAASASGTTGRAT EAANATPAGLTGPPGSP	7527
RSN-3047	GSAPTTGRAGEAEGAT SAGATGPATSGSETPGT	7280	RSC-3047	GTAEAASASGTTGRAG EAEGATSAGATGPPGSP	7528
RSN-3052	GSAPTTGEAGEAANAT SAGATGPATSGSETPGT	7281	RSC-3052	GTAEAASASGTTGEAG EAANATSAGATGPPGSP	7529
RSN-3043	GSAPTTGEAGEAAGLTP AGLTGPATSGSETPGT	7282	RSC-3043	GTAEAASASGTTGEAG EAAGLTPAGLTGPPGSP	7530
RSN-3041	GSAPTTGAAGEAANAT PAGLTGPATSGSETPGT	7283	RSC-3041	GTAEAASASGTTGAAG EAANATPAGLTGPPGSP	7531
RSN-3044	GSAPTTGRAGEAAGLT PAGLTGPATSGSETPGT	7284	RSC-3044	GTAEAASASGTTGRAG EAAGLTPAGLTGPPGSP	7532
RSN-3057	GSAPTTGRAGEAANAT SAGATGPATSGSETPGT	7285	RSC-3057	GTAEAASASGTTGRAG EAANATSAGATGPPGSP	7533
RSN-3058	GSAPTTGEAGEAAGAT SAGATGPATSGSETPGT	7286	RSC-3058	GTAEAASASGTTGEAG EAAGATSAGATGPPGSP	7534
RSN-2485	GSAPESGRAANTEPPEL GAGATSGSETPGT	7287	RSC-2485	GTAEAASASGESGRAA NTEPPELGAGPGSP	7535
RSN-2486	GSAPESGRAANTAPEGL TGPATSGSETPGT	7288	RSC-2486	GTAEAASASGESGRAA NTAPEGLTGPPGSP	7536
RSN-2488	GSAPEPGRAANHEPSGL TEGATSGSETPGT	7289	RSC-2488	GTAEAASASGEPGRAA NHEPSGLTEGPGSP	7537

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-2599	GSAPESGRAANHTGAP PGGLTGPATSGSETPGT	7290	RSC-2599	GTAEAASASGESGRAA NHTGAPPGGLTGPPGSP	7538
RSN-2706	GSAPTTGRTGEGANAT PGGLTGPATSGSETPGT	7291	RSC-2706	GTAEAASASGTTGRTG EGANATPGGLTGPPGSP	7539
RSN-2707	GSAPRTGRSGEAANETP EGLEGPATSGSETPGT	7292	RSC-2707	GTAEAASASGRTGRSG EAANETPEGLEGPPGSP	7540
RSN-2708	GSAPRTGRTGESANETP AGLGGPATSGSETPGT	7293	RSC-2708	GTAEAASASGRTGRTG ESANETPAGLGGPPGSP	7541
RSN-2709	GSAPSTGRTGEPANETP AGLSGPATSGSETPGT	7294	RSC-2709	GTAEAASASGSTGRTG EPANETPAGLSGPPGSP	7542
RSN-2710	GSAPTTGRAGEPANATP TGLSGPATSGSETPGT	7295	RSC-2710	GTAEAASASGTTGRAG EPANATPTGLSGPPGSP	7543
RSN-2711	GSAPRTGRPGEGANAT PTGLPGPATSGSETPGT	7296	RSC-2711	GTAEAASASGRTGRPG EGANATPTGLPGPPGSP	7544
RSN-2712	GSAPRTGRGGEAANAT PSGLGGPATSGSETPGT	7297	RSC-2712	GTAEAASASGRTGRGG EAANATPSGLGGPPGSP	7545
RSN-2713	GSAPSTGRSGESANATP GGLGGPATSGSETPGT	7298	RSC-2713	GTAEAASASGSTGRSGE SANATPGGLGGPPGSP	7546
RSN-2714	GSAPRTGRTGEEANATP AGLPGPATSGSETPGT	7299	RSC-2714	GTAEAASASGRTGRTG EEANATPAGLPGPPGSP	7547
RSN-2715	GSAPATGRPGEPANTTP EGLEGPATSGSETPGT	7300	RSC-2715	GTAEAASASGATGRPG EPANTTPEGLEGPPGSP	7548
RSN-2716	GSAPSTGRSGEPANATP GGLTGPATSGSETPGT	7301	RSC-2716	GTAEAASASGSTGRSGE PANATPGGLTGPPGSP	7549
RSN-2717	GSAPPTGRGGEGANTTP TGLPGPATSGSETPGT	7302	RSC-2717	GTAEAASASGPTGRGG EGANTTPTGLPGPPGSP	7550
RSN-2718	GSAPPTGRSGEGANATP SGLTGPATSGSETPGT	7303	RSC-2718	GTAEAASASGPTGRSGE GANATPSGLTGPPGSP	7551
RSN-2719	GSAPTTGRASEGANSTP APLTEPATSGSETPGT	7304	RSC-2719	GTAEAASASGTTGRAS EGANSTPAPLTEPPGSP	7552
RSN-2720	GSAPTYGRAAEAANTT PAGLTAPATSGSETPGT	7305	RSC-2720	GTAEAASASGTYGRAA EAANTTPAGLTAPPGSP	7553
RSN-2721	GSAPTTGRATEGANAT PAELTEPATSGSETPGT	7306	RSC-2721	GTAEAASASGTTGRAT EGANATPAELTEPPGSP	7554
RSN-2722	GSAPTVGRASEEANTTP ASLTGPATSGSETPGT	7307	RSC-2722	GTAEAASASGTVGRAS EEANTTPASLTGPPGSP	7555
RSN-2723	GSAPTTGRAPEAANATP APLTGPATSGSETPGT	7308	RSC-2723	GTAEAASASGTTGRAP EAANATPAPLTGPPGSP	7556
RSN-2724	GSAPTWGRATEPANAT PAPLTSPATSGSETPGT	7309	RSC-2724	GTAEAASASGTWGRAT EPANATPAPLTSPPGSP	7557
RSN-2725	GSAPTVGRASESANATP AELTSPATSGSETPGT	7310	RSC-2725	GTAEAASASGTVGRAS ESANATPAELTSPPGSP	7558
RSN-2726	GSAPTVGRAPEGANSTP AGLTGPATSGSETPGT	7311	RSC-2726	GTAEAASASGTVGRAP EGANSTPAGLTGPPGSP	7559
RSN-2727	GSAPTWGRATEAPNLE PATLTTPATSGSETPGT	7312	RSC-2727	GTAEAASASGTWGRAT EAPNLEPATLTTPPGSP	7560
RSN-2728	GSAPTTGRATEAPNLTP APLTEPATSGSETPGT	7313	RSC-2728	GTAEAASASGTTGRAT EAPNLTPAPLTEPPGSP	7561
RSN-2729	GSAPTQGRATEAPNLSP AALTSPATSGSETPGT	7314	RSC-2729	GTAEAASASGTQGRAT EAPNLSPAALTSPPGSP	7562

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-2730	GSAPTQGRAAEAPNLTP ATLTAPATSGSETPGT	7315	RSC-2730	GTAEAASASGTQGRAA EAPNLTPATLTAPPGSP	7563
RSN-2731	GSAPTSGRAPEATNLAP APLTGPATSGSETPGT	7316	RSC-2731	GTAEAASASGTSGRAPE ATNLAPAPLTGPPGSP	7564
RSN-2732	GSAPTQGRAAEAANLT PAGLTEPATSGSETPGT	7317	RSC-2732	GTAEAASASGTQGRAA EAANLTPAGLTEPPGSP	7565
RSN-2733	GSAPTTGRAGSAPNLPP TGLTTPATSGSETPGT	7318	RSC-2733	GTAEAASASGTTGRAG SAPNLPPTGLTTPPGSP	7566
RSN-2734	GSAPTTGRAGGAENLPP EGLTAPATSGSETPGT	7319	RSC-2734	GTAEAASASGTTGRAG GAENLPPEGLTAPPGSP	7567
RSN-2735	GSAPTTSRAGTATNLTP EGLTAPATSGSETPGT	7320	RSC-2735	GTAEAASASGTTSRAG TATNLTPEGLTAPPGSP	7568
RSN-2736	GSAPTTGRAGTATNLPP SGLTTPATSGSETPGT	7321	RSC-2736	GTAEAASASGTTGRAG TATNLPPSGLTTPPGSP	7569
RSN-2737	GSAPTTARAGEAENLSP SGLTAPATSGSETPGT	7322	RSC-2737	GTAEAASASGTTARAG EAENLSPSGLTAPPGSP	7570
RSN-2738	GSAPTTGRAGGAGNLA PGGLTEPATSGSETPGT	7323	RSC-2738	GTAEAASASGTTGRAG GAGNLAPGGLTEPPGSP	7571
RSN-2739	GSAPTTGRAGTATNLPP EGLTGPATSGSETPGT	7324	RSC-2739	GTAEAASASGTTGRAG TATNLPPEGLTGPPGSP	7572
RSN-2740	GSAPTTGRAGGAANLA PTGLTEPATSGSETPGT	7325	RSC-2740	GTAEAASASGTTGRAG GAANLAPTGLTEPPGSP	7573
RSN-2741	GSAPTTGRAGTAENLA PSGLTTPATSGSETPGT	7326	RSC-2741	GTAEAASASGTTGRAG TAENLAPSGLTTPPGSP	7574
RSN-2742	GSAPTTGRAGSATNLGP GGLTGPATSGSETPGT	7327	RSC-2742	GTAEAASASGTTGRAG SATNLGPGGLTGPPGSP	7575
RSN-2743	GSAPTTARAGGAENLT PAGLTEPATSGSETPGT	7328	RSC-2743	GTAEAASASGTTARAG GAENLTPAGLTEPPGSP	7576
RSN-2744	GSAPTTARAGSAENLSP SGLTGPATSGSETPGT	7329	RSC-2744	GTAEAASASGTTARAG SAENLSPSGLTGPPGSP	7577
RSN-2745	GSAPTTARAGGAGNLA PEGLTTPATSGSETPGT	7330	RSC-2745	GTAEAASASGTTARAG GAGNLAPEGLTTPPGSP	7578
RSN-2746	GSAPTTSRAGAAENLTP TGLTGPATSGSETPGT	7331	RSC-2746	GTAEAASASGTTSRAG AAENLTPTGLTGPPGSP	7579
RSN-2747	GSAPTYGRTTTPGNEPP ASLEAEATSGSETPGT	7332	RSC-2747	GTAEAASASGTYGRTT TPGNEPPASLEAEPGSP	7580
RSN-2748	GSAPTYSRGESGPNEPP PGLTGPATSGSETPGT	7333	RSC-2748	GTAEAASASGTYSRGES GPNEPPPGLTGPPGSP	7581
RSN-2749	GSAPAWGRTGASENET PAPLGGEATSGSETPGT	7334	RSC-2749	GTAEAASASGAWGRTG ASENETPAPLGGEPGSP	7582
RSN-2750	GSAPRWGRAETTPNTPP EGLETEATSGSETPGT	7335	RSC-2750	GTAEAASASGRWGRAE TTPNTPPEGLETEPGSP	7583
RSN-2751	GSAPESGRAANHTGAE PPELGAGATSGSETPGT	7336	RSC-2751	GTAEAASASGESGRAA NHTGAEPPELGAGPGSP	7584
RSN-2754	GSAPTTGRAGEAANLT PAGLTESATSGSETPGT	7337	RSC-2754	GTAEAASASGTTGRAG EAANLTPAGLTESPGSP	7585
RSN-2755	GSAPTTGRAGEAANLT PAALTESATSGSETPGT	7338	RSC-2755	GTAEAASASGTTGRAG EAANLTPAALTESPGSP	7586
RSN-2756	GSAPTTGRAGEAANLT PAPLTESATSGSETPGT	7339	RSC-2756	GTAEAASASGTTGRAG EAANLTPAPLTESPGSP	7587

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-2757	GSAPTTGRAGEAANLT PEPLTESATSGSETPGT	7340	RSC-2757	GTAEAASASGTTGRAG EAANLTPEPLTESPGSP	7588
RSN-2758	GSAPTTGRAGEAANLT PAGLTGAATSGSETPGT	7341	RSC-2758	GTAEAASASGTTGRAG EAANLTPAGLTGAPGSP	7589
RSN-2759	GSAPTTGRAGEAANLT PEGLTGAATSGSETPGT	7342	RSC-2759	GTAEAASASGTTGRAG EAANLTPEGLTGAPGSP	7590
RSN-2760	GSAPTTGRAGEAANLT PEPLTGAATSGSETPGT	7343	RSC-2760	GTAEAASASGTTGRAG EAANLTPEPLTGAPGSP	7591
RSN-2761	GSAPTTGRAGEAANLT PAGLTEAATSGSETPGT	7344	RSC-2761	GTAEAASASGTTGRAG EAANLTPAGLTEAPGSP	7592
RSN-2762	GSAPTTGRAGEAANLT PEGLTEAATSGSETPGT	7345	RSC-2762	GTAEAASASGTTGRAG EAANLTPEGLTEAPGSP	7593
RSN-2763	GSAPTTGRAGEAANLT PAPLTEAATSGSETPGT	7346	RSC-2763	GTAEAASASGTTGRAG EAANLTPAPLTEAPGSP	7594
RSN-2764	GSAPTTGRAGEAANLT PEPLTEAATSGSETPGT	7347	RSC-2764	GTAEAASASGTTGRAG EAANLTPEPLTEAPGSP	7595
RSN-2765	GSAPTTGRAGEAANLT PEPLTGPATSGSETPGT	7348	RSC-2765	GTAEAASASGTTGRAG EAANLTPEPLTGPPGSP	7596
RSN-2766	GSAPTTGRAGEAANLT PAGLTGGATSGSETPGT	7349	RSC-2766	GTAEAASASGTTGRAG EAANLTPAGLTGGPGSP	7597
RSN-2767	GSAPTTGRAGEAANLT PEGLTGGATSGSETPGT	7350	RSC-2767	GTAEAASASGTTGRAG EAANLTPEGLTGGPGSP	7598
RSN-2768	GSAPTTGRAGEAANLT PEALTGGATSGSETPGT	7351	RSC-2768	GTAEAASASGTTGRAG EAANLTPEALTGGPGSP	7599
RSN-2769	GSAPTTGRAGEAANLT PEPLTGGATSGSETPGT	7352	RSC-2769	GTAEAASASGTTGRAG EAANLTPEPLTGGPGSP	7600
RSN-2770	GSAPTTGRAGEAANLT PAGLTEGATSGSETPGT	7353	RSC-2770	GTAEAASASGTTGRAG EAANLTPAGLTEGPGSP	7601
RSN-2771	GSAPTTGRAGEAANLT PEGLTEGATSGSETPGT	7354	RSC-2771	GTAEAASASGTTGRAG EAANLTPEGLTEGPGSP	7602
RSN-2772	GSAPTTGRAGEAANLT PAPLTEGATSGSETPGT	7355	RSC-2772	GTAEAASASGTTGRAG EAANLTPAPLTEGPGSP	7603
RSN-2773	GSAPTTGRAGEAANLT PEPLTEGATSGSETPGT	7356	RSC-2773	GTAEAASASGTTGRAG EAANLTPEPLTEGPGSP	7604
RSN-3047	GSAPTTGRAGEAEGAT SAGATGPATSGSETPGT	7357	RSC-3047	GTAEAASASGTTGRAG EAEGATSAGATGPPGSP	7605
RSN-2783	GSAPEAGRSAEATSAG ATGPATSGSETPGT	7358	RSC-2783	GTAEAASASGEAGRSA EATSAGATGPPGSP	7606
RSN-3107	GSAPSASGTYSRGESGP GSPATSGSETPGT	7359	RSC-3107	GTAEAASASGSASGTYS RGESGPGSPPGSP	7607
RSN-3103	GSAPSASGEAGRTDTHP GSPATSGSETPGT	7360	RSC-3103	GTAEAASASGSASGEA GRTDTHPGSPPGSP	7608
RSN-3102	GSAPSASGEPGRAAEHP GSPATSGSETPGT	7361	RSC-3102	GTAEAASASGSASGEPG RAAEHPGSPPGSP	7609
RSN-3119	GSAPSPAGESSRGTTIA GSPATSGSETPGT	7362	RSC-3119	GTAEAASASGSPAGESS RGTTIAGSPPGSP	7610
RSN-3043	GSAPTTGEAGEAAGLTP AGLTGPATSGSETPGT	7363	RSC-3043	GTAEAASASGTTGEAG EAAGLTPAGLTGPPGSP	7611
RSN-2789	GSAPEAGESAGATPAG LTGPATSGSETPGT	7364	RSC-2789	GTAEAASASGEAGESA GATPAGLTGPPGSP	7612

Name	Amino Acid Sequence	SEQ ID NO:	Name	Amino Acid Sequence	SEQ ID NO:
RSN-3109	GSAPSASGAPLELEAGP GSPATSGSETPGT	7365	RSC-3109	GTAEAASASGSASGAPL ELEAGPGSPPGSP	7613
RSN-3110	GSAPSASGEPPELGAGP GSPATSGSETPGT	7366	RSC-3110	GTAEAASASGSASGEPP ELGAGPGSPPGSP	7614
RSN-3111	GSAPSASGEPSGLTEGP GSPATSGSETPGT	7367	RSC-3111	GTAEAASASGSASGEPS GLTEGPGSPPGSP	7615
RSN-3112	GSAPSASGTPAPLTEPP GSPATSGSETPGT	7368	RSC-3112	GTAEAASASGSASGTPA PLTEPPGSPPGSP	7616
RSN-3113	GSAPSASGTPAELTEPP GSPATSGSETPGT	7369	RSC-3113	GTAEAASASGSASGTPA ELTEPPGSPPGSP	7617
RSN-3114	GSAPSASGPPPGLTGPP GSPATSGSETPGT	7370	RSC-3114	GTAEAASASGSASGPPP GLTGPPGSPPGSP	7618
RSN-3115	GSAPSASGTPAPLGGEP GSPATSGSETPGT	7371	RSC-3115	GTAEAASASGSASGTPA PLGGEPGSPPGSP	7619
RSN-3125	GSAPSPAGAPEGLTGPA GSPATSGSETPGT	7372	RSC-3125	GTAEAASASGSPAGAPE GLTGPAGSPPGSP	7620
RSN-3126	GSAPSPAGPPEGLETEA GSPATSGSETPGT	7373	RSC-3126	GTAEAASASGSPAGPPE GLETEAGSPPGSP	7621
RSN-3127	GSAPSPTSGQGGLTGPG SEPATSGSETPGT	7374	RSC-3127	GTAEAASASGSPTSGQG GLTGPGSEPPGSP	7622
RSN-3131	GSAPSESAPPEGLETEST EPATSGSETPGT	7375	RSC-3131	GTAEAASASGSESAPPE GLETESTEPPGSP	7623
RSN-3132	GSAPSEGSEPLELGAAS ETPATSGSETPGT	7376	RSC-3132	GTAEAASASGSEGSEPL ELGAASETPPGSP	7624
RSN-3133	GSAPSEGSGPAGLEAPS ETPATSGSETPGT	7377	RSC-3133	GTAEAASASGSEGSGPA GLEAPSETPPGSP	7625
RSN-3138	GSAPSEPTPPASLEAEPG SPATSGSETPGT	7378	RSC-3138	GTAEAASASGSEPTPPA SLEAEPGSPPGSP	7626

[0210] In some embodiments, the RS for incorporation into the subject recombinant polypeptides can be designed to be selectively sensitive in order to have different rates of cleavage and different cleavage efficiencies to the various proteases for which they are substrates. As a given protease may be found in different concentrations in diseased tissues, including but not limited to a tumor, a blood cancer, or an inflammatory tissue or site of inflammation, compared to healthy tissues or in the circulation, the disclosure provides RS that have had the individual amino acid sequences engineered to have a higher or lower cleavage efficiency for a given protease in order to ensure that the recombinant polypeptide is preferentially converted from the prodrug form to the active form (i.e., by the separation and release of the binding moieties and XTEN from the recombinant polypeptide after cleavage of the RS) when in proximity to the target cell or tissue and its co-localized proteases compared to the rate of cleavage of the RS in healthy tissue or the circulation such that the released antibody fragment binding moieties have a greater ability to bind to ligands in the diseased tissues compared to the prodrug form that remains in circulation. By such selective designs, the therapeutic index of

the resulting compositions can be improved, resulting in reduced side effects relative to convention therapeutics that do not incorporate such site-specific activation.

[0211] As used herein cleavage efficiency is defined as the $\log_2$ value of the ratio of the percentage of the test substrate comprising the RS cleaved to the percentage of the control substrate AC1611 cleaved when each is subjected to the protease enzyme in biochemical assays (further detailed in the Examples) in which reaction in conducted wherein the initial substrate concentration is 6 μM, the reactions are incubated at 37°C for 2 hours before being stopped by adding EDTA, with the amount of digestion products and uncleaved substrate analyzed by non-reducing SDS-PAGE to establish the ratio of the percentage cleaved. The cleavage efficiency is calculated as follows: $Log_2\left(\frac{\%\ Cleaved\ for\ substrate\ of\ interest}{\%\ cleaved\ for\ AC1611\ in\ the\ same\ experiment}\right)$. Thus, a cleavage efficiency of -1 means that the amount of test substrate cleaved was 50% compared to that of the control substrate, while a cleavage efficiency of +1 means that the amount of test substrate cleaved was 200% compared to that of the control substrate. A higher rate of cleavage by the test protease relative to the control would result in a higher cleavage efficiency, and a slower rate of cleavage by the test protease relative to the control would result in a lower cleavage efficiency. As detailed in the Examples, a control RS sequence AC1611 (RSR-1517), having the amino acid sequence EAGRSANHEPLGLVAT (SEQ ID NO: 7001), was established as having an appropriate baseline cleavage efficiency by the proteases legumain, MMP-2, MMP-7, MMP-9, MMP-14, uPA, and matriptase, when tested in *in vitro* biochemical assays for rates of cleavage by the individual proteases. By selective substitution of amino acids at individual locations in the RS peptides, libraries of RS were created and evaluated against the panel of the 7 proteases (detailed more fully in the Examples), resulting in profiles that were used to establish guidelines for appropriate amino acid substitutions in order to achieve RS with desired cleavage efficiencies. In making RS with desired cleavage efficiencies, substitutions using the hydrophilic amino acids A, E, G, P, S, and T are preferred, however other L-amino acids can be substituted at given positions in order to adjust the cleavage efficiency so long as the RS retains at least some susceptibility to cleavage by a protease. Conservative substitutions of amino acids in a peptide to retain or effect activity is well within the knowledge and capabilities of a person within skill in the art. In one embodiment, the disclosure provides RS in which the RS is cleaved by a protease including but not limited to legumain, MMP-1, MMP-2, MMP-7, MMP-9, MMP-11, MMP-14, uPA, or matriptase with at least a 0.2 $\log_2$, or 0.4 $\log_2$, or 0.8 $\log_2$, or 1.0 $\log_2$ higher cleavage efficiency in an *in vitro* biochemical competitive assay compared to the cleavage by the same protease of a control sequence RSR-1517 having the sequence EAGRSANHEPLGLVAT (SEQ ID NO. 7001). In some embodiments, the disclosure provides RS in which the RS is cleaved by a protease including but not limited to legumain, MMP-1, MMP-2, MMP-7, MMP-9, MMP-11, MMP-14, uPA, or matriptase with at least a 0.2 $\log_2$, or 0.4 $\log_2$, or 0.8 $\log_2$, or 1.0 $\log_2$ lower cleavage efficiency in an *in vitro* biochemical competitive assay compared to the cleavage by the same protease of a control sequence RSR-1517 having the sequence

EAGRSANHEPLGLVAT (SEQ ID NO. 7001) . In one embodiment, the disclosure provides RS in which the rate of cleavage of the RS by a protease including but not limited to legumain, MMP-1, MMP-2, MMP-7, MMP-9, MMP-11, MMP-14, uPA, or matriptase is at least 2-fold, or at least 4-fold, or at least 8 fold, or at least 16-fold faster compared to the control sequence RSR-1517 having the sequence EAGRSANHEPLGLVAT (SEQ ID NO. 7001). In some embodiments, the disclosure provides RS in which the rate of cleavage of the RS by a protease including but not limited to legumain, MMP-1, MMP-2, MMP-7, MMP-9, MMP-11, MMP-14, uPA, or matriptase is at least 2-fold, or at least 4-fold, or at least 8 fold, or at least 16-fold slower compared to the control sequence RSR-1517 having the sequence EAGRSANHEPLGLVAT (SEQ ID NO. 7001).

[0212] In some embodiments, the disclosure provides AAC comprising multiple RS wherein each RS sequence is identified herein by the group of sequences set forth in **Table 8a** and the RS are linked to each other by 1 to 6 amino acids that are glycine, serine, alanine, and threonine. In one embodiment, the AAC comprises a first RS and a second RS different from the first RS wherein each RS sequence is identified herein by the sequences set forth in **Table 8a** and the RS are linked to each other by 1 to 6 amino acids that are glycine, serine, alanine, and threonine. In some embodiments, the AAC comprises a first RS, a second RS different from the first RS, and a third RS different from the first and the second RS wherein each sequence is identified herein by the sequences set forth in **Table 8a** and the first and the second and the third RS are linked to each other by 1 to 6 amino acids that are glycine, serine, alanine, and threonine. It is specifically intended that the multiple RS of the AAC can be concatenated to form a sequence that can be cleaved by multiple proteases at different rates or efficiency of cleavage. In some embodiments, the disclosure provides AAC comprising an RS1 and an RS2 identified herein by the sequences set forth in **Tables 8a-8b** and an XTEN 1 and XTEN 2, such as those described hereinabove or described elsewhere herein, wherein the RS1 is fused between the XTEN1 and the binding moieties and the RS2 is fused between the XTEN2 and the binding moieties. It is contemplated that such compositions would be more readily cleaved by diseased target tissues that express multiple proteases, compared with healthy tissues or when in the normal circulation, with the result that the resulting fragments bearing the binding moieties would more readily penetrate the target tissue; e.g., a tumor, and have an enhanced ability to bind and link the target cell and the effector cell (or just the target cell in the case of AAC designed with a single binding moiety.

[0213] The RS of the disclosure are useful for inclusion in recombinant polypeptides as therapeutics for treatment of cancers, autoimmune diseases, inflammatory diseases and other conditions where localized activation of the recombinant polypeptide is desirable. The subject compositions address an unmet need and are superior in one or more aspects including enhanced terminal half-life, targeted delivery, and improved therapeutic ratio with reduced toxicity to healthy tissues compared to conventional antibody therapeutics or bispecific antibody therapeutics that are active upon injection.

[0214] In some embodiments, the (fusion) polypeptide comprises a first release segment (RS1) positioned between the (first) XTEN and the biologically active polypeptide. In some embodiments, the polypeptide further comprises a second release segment (RS2) positioned between the biologically active polypeptide and the second XTEN. In some embodiments, RS1 and RS2 are identical in sequence. In some embodiments, RS1 and RS2 are not identical in sequence. In some embodiments, the RS1 comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence identified herein by those in **Tables 8a-8b** or a subset thereof. In some embodiments, the RS2 comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence identified herein by those in **Tables 8a-8b** or a subset thereof. In some embodiments, the RS1 and RS2 are each a substrate for cleavage by multiple proteases at one, two, or three cleavage sites within each release segment sequence.

[0215] Reference Fragment

[0216] In some embodiments, the (fusion) polypeptide further comprises one or more reference fragments releasable from the polypeptide upon digestion by the protease. In some embodiments, the one or more reference fragments each comprise a portion of the biologically active polypeptide. In some embodiments, the one or more reference fragments is a single reference fragment that differs in sequence and molecular weight from all other peptide fragments that are releasable from the polypeptide upon digestion of the polypeptide by the protease.

[0217] Exemplary Polypeptides

[0218] In some embodiments of the compositions of this disclosure, the polypeptide is a recombinant polypeptide comprising an amino acid sequence having at least (about) 80% sequence identity to a sequence set forth in **Table D** (consisting of **SEQ ID NOS: 12-47**) or a subset thereof. The polypeptide can comprise an amino acid sequence having at least (about) 81%, at least (about) 82%, at least (about) 83%, at least (about) 84%, at least (about) 85%, at least (about) 86%, at least (about) 87%, at least (about) 88%, at least (about) 89%, at least (about) 90%, at least (about) 91%, at least (about) 92%, at least (about) 93%, at least (about) 94%, at least (about) 95%, at least (about) 96%, at least (about) 97%, at least (about) 98%, at least (about) 99%, or (about) 100% sequence identity to a sequence set forth in **Table D (SEQ ID NOS: 12-47)** or a subset thereof. The polypeptide can comprise an amino acid sequence having at least (about) 90%, at least (about) 91%, at least (about) 92%, at least (about) 93%, at least (about) 94%, at least (about) 95%, at least (about) 96%, at least (about) 97%, at least (about) 98%, at least (about) 99%, or (about) 100% sequence identity to a sequence set forth in **Table D (SEQ ID NOS: 12-47)** or a subset thereof. The polypeptide can comprise an amino acid sequence identical to a sequence set forth in **Table D (SEQ ID NOS: 12-47)** or a subset thereof. It is specifically contemplated that the compositions of this disclosure can comprise sequence variants of the amino acid

sequences set forth in **Table D**, such as with linker sequence(s) inserted or with purification tag sequence(s) attached thereto, so long as the variants exhibit substantially similar or same bioactivity/bioactivities and/or activation mechanism(s).

Table D: Exemplary amino acid sequences of polypeptides

SEQ ID NO.	Amino Acid Sequences
12	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPLGGRADNHEPPELGAGATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTAVAW YQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWVRQA PGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMDYWG QGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAP GTPARFSGSSLGGSAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGG SAESEPPGEGEVQLQESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATY YADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAAS ASGLGGRADNHEPPELGAGSAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTE EGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTST EEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEG SAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSE GSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEP SEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPA TSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPA GSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSE PATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGT SESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPG TSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSA PGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGST ETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSE GSAPGTATESPEGSAPGTSESATPESGPGTSTEPSEGSAPGTSAESATPESGPGSEPATSGSETPGTSTEP SEGSAPGTSTEPSEGSAPGTSESATPESGPGTESAS
13	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGEEPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPASTRGENAGPAGLEAPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVSIGVAW YQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMDWVRQA PGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRAPG TPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTGGS AESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYY ADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAASA SGASTRGENAGPAGLEAPPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSP AGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGT SESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEG TSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESG

SEQ ID NO.	Amino Acid Sequences
	PGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPES GPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGSEPATSGS ETPGTSESATPESGPGTSTEPSEGSAPGEPEA
14	ASSPAGSPTSTESGTSESATPESGPGTETEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGESPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPEAGRSANHTPAGLTGPATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDVSIGVAW YQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTMDWVRQA PGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRAPG TPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTGGS AESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYY ADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAASA SGEAGRSANHTPAGLTGPTPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPT STEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSES ATPESGPGTSESATPESGPGSEPATSGSETPGSESATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTST EPSEGSAPGSEPATSGSETPGTSESA
15	SPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEG SAPGGSAPLFGRNDNHEPLELGGGATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTAVAWYQ QKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTKVEIKG ATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWVRQAPG KGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMDYWGQG TLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAPGT PARFSGSSLGGSAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGGSA ESEPPGEGEVQLQESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYA DSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAASAS GLFGRNDNHEPLELGGGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTE PSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPA GSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGT SESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPG TSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESG PGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGSEPATSGSE TPGTSESATPESGPGTSTEPSEGSAPG
16	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPRTGRTGESANETPAGLGGPATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDVSIG VAWYQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQGTK VEIKGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTMDWV RQAPGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFYFDY

SEQ ID NO.	Amino Acid Sequences
	WGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKR APGTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETT GGSAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYA TYYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEA ASASGRTGRTGESANETPAGLGGPGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEP SEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSES ATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPA GSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTS TEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGT SESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAP GTSESATPESGPGSEPATSGSTETGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSA PGSPAGSPTSTEEGTSESATPESGPGSEPATS
17	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPGAGRTDNHEPLELGAAATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVSIGVAW YQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMDWVRQA PGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAPG TPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGGS AESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYY ADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAASA SGGAGRTDNHEPLELGAATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPT STEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSES ATPESGPGTSESATPESGPGSEPATSGSETPGSESATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTST EPSEGSAPGSEPATSGSETPGTSESA
18	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSA PGTSESATPESGPGSESATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTE PSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPG SEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPESGRAANTGPPTLTAPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTAV AWYQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTKV EIKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWVR QAPGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMDY WGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKR APGTPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETT GGSAESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYA

SEQ ID NO.	Amino Acid Sequences
	TYYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEA ASASGESGRAANTGPPTLTAPPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEG TSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEE GTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSA PGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGS APGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSE GSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATS GSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGS PTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPA TSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSE SATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTS ESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGS EPATSGSETPGTSESATPESGPGTSTEPSEGSAPG
19	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPASGRSTNAGPPGLTGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTAVAW YQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWVRQA PGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMDYWG QGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAP GTPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGG SAESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATY YADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAAS ASGASGRSTNAGPPGLTGPPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPG TSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEE GTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSET PGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPE SGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPG
20	SAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGSTPAESGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTE PSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSTETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGT SESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEE GTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESG PGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPES GPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATS GSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESA TPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSE SATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTS ESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGT SESATPESGPGTESASLFGRNDNHEPLELGGGATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDV SIGVAWYQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQ

SEQ ID NO.	Amino Acid Sequences
	GTKVEIKGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTM DWVRQAPGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFY FDYWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGT NKRAPGTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPG ETTGGSAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYN NYATYYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGT AEAASASGLFGRNDNHEPLELGGGEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAG SPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTST EPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTS ESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGT SESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPG TSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESASPESGPGTSTEPSEGSAP GSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTE EGTSTEPSEGSAPGTSESATPESGPGTSESAT
21	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSA PATSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESASPESGPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTE PSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPG SEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPTTGRAGEAEGATSAGATGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVN TAVAWYQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQG TKVEIKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIH WVRQAPGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYA MDYWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGT NKRAPGTPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPG ETTGGSAESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYN NYATYYADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGT AEAASASGTTGRAGEAEGATSAGATGPGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTS TEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGT SESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPG SPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSA PGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGS APGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEG SAPGTSESATPESGPGSEPATSGSTETGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSE GSAPGSPAGSPTSTEEGTSESATPESGPGSEPATS
22	ASSPAGSPTSTESGTSESATPESGPGTETEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGESPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPASGRGTNAGPAGLTGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVSIGVAW YQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMDWVRQA PGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRAPG

SEQ ID NO.	Amino Acid Sequences
	TPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTGGS AESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYY ADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAASA SGASGRGTNAGPAGLTGPPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTST EPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTS ESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGT SESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPG TSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSA PGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSE TPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTS TEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESA TPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGSEPA TSGSETPGTSESATPESGPGTSTEPSEGSAPG
23	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGEEPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPGAGRTDNHEPLELGAAATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTAVAW YQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWVRQA PGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMDYWG QGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAP GTPARFSGSSLGGSAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGG SAESEPPGEGEVQLQESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATY YADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAAS ASGGAGRTDNHEPLELGAAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTS TEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGT SESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPG TSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAP GTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESG PGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATS GSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESA TPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGftab TSESATPESGPGSEPATSGPTESGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGEPEA
24	SAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGSTPAESGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTE PSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSTETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGT SESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEE GTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESG PGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPES GPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATS GSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESA

SEQ ID NO.	Amino Acid Sequences
	TPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSE SATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTS ESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGT SESATPESGPGTESASASGRAANETPPGLTGAATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDV SIGVAWYQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQ GTKVEIKGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTM DWVRQAPGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFY FDYWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGT NKRAPGTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPG ETTGGSAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYN NYATYYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGT AEAASASGASGRAANETPPGLTGAGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEP SEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSES ATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPA GSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTS TEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGT SESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAP GTSESATPESGPGSEPATSGSTETGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSA PGSPAGSPTSTEEGTSESATPESGPGSEPATS
25	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSA PATSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESASPESGPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTE PSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPG SEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPASGRSTNAGPPGLTGPATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDVSIGV AWYQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQGTKV EIKGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTMDWVR QAPGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFYFDYW GQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRA PGTPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTG GSAESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYAT YYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAA SASGASGRSTNAGPPGLTGPGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGS APGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPE SGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPT STEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSES ATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSE SATPESGPGSEPATSGSTETGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSP AGSPTSTEEGTSESATPESGPGSEPATS
26	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGEEPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP

SEQ ID NO.	Amino Acid Sequences
	ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPRTGRTGESANETPAGLGGPATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDVSIG VAWYQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQGTK VEIKGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTMDWV RQAPGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFYFDY WGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKR APGTPARFSGSSLGGSAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETT GGSAESEPPGEGEVQLQESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYA TYYADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEA ASASGRTGRTGESANETPAGLGGPPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPES GPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEG SAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSP TSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSE SATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGSE PATSGSETPGTSESATPESGPGTSTEPSEGSAPG
27	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPLFGRNDNHEPLELGGGATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVSIGVAW YQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMDWVRQA PGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAPG TPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGGS AESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYY ADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAASA SGLFGRNDNHEPLELGGGPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTST EPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTS ESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGT SESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPG TSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSA PGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSE TPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTS TEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGftabT SESATPESGPGSEPATSGPTESGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPG SEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGEPEA
28	ASSPAGSPTSTESGTSESATPESGPGTETEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGESPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPTTGRAGEAANATSAGATGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTA VAWYQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTK VEIKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWV RQAPGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMD YWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNK RAPGTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGET

SEQ ID NO.	Amino Acid Sequences
	TGGSAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNY ATYYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAE AASASGTTGRAGEAANATSAGATGPSAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAG SPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPA GSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTS TEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGT STEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPG TSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAP GTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESG PGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTST EEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSG SETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESAT PESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEP SEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTE PSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEP ATSGSTETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGT STEPSEGSAPGTATESPEGSAPGTSESATPESGPGTSTEPSEGSAPGTSAESATPESGPGSEPATSGSETP GTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTESAS
29	SPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEG SAPGGSAPTTGRATEAANATPAGLTGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVSIGVA WYQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQGTKVE IKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMDWVRQ APGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYFDYWG QGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAP GTPARFSGSSLGGSAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGG SAESEPPGEGEVQLQESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATY YADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAAS ASGTTGRATEAANATPAGLTGPTPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATS GSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEP SEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPA GSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGS EPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPG TSESATPESGPGTSESATPESGPGSEPATSGSETPGSESATSGSETPGSPAGSPTSTEEGTSTEPSEGSAP GTSTEPSEGSAPGSEPATSGSETPGTSESA
30	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSA PGTSESATPESGPGSESATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTE PSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPG SEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPTTAPAGSAENLSPSGLTGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVS

SEQ ID NO.	Amino Acid Sequences
	IGVAWYQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQG TKVEIKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMD WVRQAPGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYF DYWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTN KRAPGTPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGE TTGGSAESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNN YATYYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTA EAASASGTTARAGSAENLSPSGLTGPEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSE SATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSP AGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGT STEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPG TSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAP GTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSA PGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESASPESGPGTSTEPSEGS APGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTS TEEGTSTEPSEGSAPGTSESATPESGPGTSESAT
31	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSA PGTSESATPESGPGSESATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTE PSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPG SEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPTTGRAGEAEGATSAGATGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVS IGVAWYQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQG TKVEIKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMD WVRQAPGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYF DYWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTN KRAPGTPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGE TTGGSAESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNN YATYYADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTA EAASASGTTGRAGEAEGATSAGATGPPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPT STEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSP TSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTE PSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSE SATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTS TEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS EPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEG SPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEG SAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG
32	ASSPAGSPTSTESGTSESATPESGPGTETEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGESPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS

SEQ ID NO.	Amino Acid Sequences
	EGSAPGGSAPPTGRSGEGANATPSGLTGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTA VAWYQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTK VEIKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWV RQAPGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMD YWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNK RAPGTPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGET TGGSAESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNY ATYYADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAE AASASGPTGRSGEGANATPSGLTGPPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTS TEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTST EPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSE PATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGS PAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPG SEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESG PGftabTSESATPESGPGSEPATSGPTESGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTP SEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGEPEA
33	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPLGGRADNHEPPELGAGATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTAVAW YQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWVRQA PGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMDYWG QGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRAP GTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTGG SAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATY YADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAAS ASGLGGRADNHEPPELGAGTPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSE TPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEG SAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATP ESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSP TSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESA TPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPA TSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSE SATPESGPGTSESATPESGPGSEPATSGSETPGSESATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTS TEPSEGSAPGSEPATSGSETPGTSESA
34	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPEAGRSANHTPAGLTGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVSIGVAW YQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMDWVRQA PGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRAP GTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTGG SAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATY

SEQ ID NO.	Amino Acid Sequences
	YADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAAS ASGEAGRSANHTPAGLTGPPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPG TSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEE GTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSET PGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPE SGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPGEPEA
35	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPGAGRTDNHEPLELGAAATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDVSIGVAW YQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTMDWVRQA PGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAPG TPARFSGSSLGGSAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGGS AESEPPGEGEVQLQESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYY ADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAASA SGGAGRTDNHEPLELGAAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSP AGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGT SESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEG TSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESG PGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPES GPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGSEPATSGS ETPGTSESATPESGPGTSTEPSEGSAPGEPEA
36	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSA PATSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESASPESGPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSTE PSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPG SEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPASGRAANETPPGLTGAATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDVSIGV AWYQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQGTKV EIKGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTMDWVR QAPGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFYFDYW GQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRA PGTPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTG GSAESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYAT YYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAA SASGASGRAANETPPGLTGASAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTST EEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTS

SEQ ID NO.	Amino Acid Sequences
	TEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSE GSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPS EGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESA TPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTE PSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSP AGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGS EPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPG TSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGP GTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSA PGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGS APGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGS TETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPS EGSAPGTATESPEGSAPGTSESATPESGPGTSTEPSEGSAPGTSAESATPESGPGSEPATSGSETPGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGTESAS
37	SAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGSTPAESGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTE PSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSTETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGT SESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEE GTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESG PGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPES GPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATS GSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESA TPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSE SATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTS ESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGT SESATPESGPGTESASESGRAANTGPPTLTAPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDV SIGVAWYQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQ GTKVEIKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTM DWVRQAPGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFY FDYWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGT NKRAPGTPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPG ETTGGSAESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYN NYATYYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGT AEAASASGESGRAANTGPPTLTAPSAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGS PTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAG SPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTST EPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTS TEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGT SESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPG TSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTE EGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEP SEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPA

SEQ ID NO.	Amino Acid Sequences
	TSGSTETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTS TEPSEGSAPGTATESPEGSAPGTSESATPESGPGTSTEPSEGSAPGTSAESATPESGPGSEPATSGSETPG TSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTESAS
38	ASSPAGSPTSTESGTSESATPESGPGTETEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGESPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPASGRGTNAGPAGLTGPATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDVSIGVAW YQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTMDWVRQA PGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAPG TPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGGS AESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYY ADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAASA SGASGRGTNAGPAGLTGPEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTE EGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPE SGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATP ESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESAT PESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESASPESGPGTSTEPSEGSAPGSPAGS PTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTE PSEGSAPGTSESATPESGPGTSESAT
39	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGEEPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPEAGRSANHTPAGLTGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTAVAW YQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWVRQA PGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMDYWG QGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRAP GTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTGG SAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATY YADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAAS ASGEAGRSANHTPAGLTGPSAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTE EGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTST EEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEG SAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSE GSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEP SEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPA TSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPA GSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSE PATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGT SESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPG TSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAP GSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSA PGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGST ETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSE GSAPGTATESPEGSAPGTSESATPESGPGTSTEPSEGSAPGTSAESATPESGPGSEPATSGSETPGTSTEP SEGSAPGTSTEPSEGSAPGTSESATPESGPGTESAS

SEQ ID NO.	Amino Acid Sequences
40	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPASTRGENAGPAGLEAPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVSIGVAW YQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMDWVRQA PGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRAPG TPARFSGSSLGGSAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTGGS AESEPPGEGEVQLQESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYY ADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAASA SGASTRGENAGPAGLEAPPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTST EPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTS ESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGT SESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPG TSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSA PGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSE TPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTS TEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGFtabT SESATPESGPGSEPATSGPTESGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPG SEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGEPEA
41	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSA PGTSESATPESGPGSESATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEG SAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPS EGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTE PSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGS PAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPG SEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPPTGRSGEGANATPSGLTGPATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDVS IGVAWYQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQG TKVEIKGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTMD WVRQAPGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFYF DYWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTN KRAPGTPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGE TTGGSAESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNN YATYYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTA EAASASGPTGRSGEGANATPSGLTGPPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATP ESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPS EGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAG SPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEP ATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGT

SEQ ID NO.	Amino Acid Sequences
	SESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPG SEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG
42	SPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEG SAPGGSAPRTGRTGESANETPAGLGGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVSIGVA WYQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQGTKVE IKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMDWVRQ APGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYFDYWG QGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRAP GTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTGG SAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATY YADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAAS ASGRTGRTGESANETPAGLGGPPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGP GTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSA PGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSE TPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTS TEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESA TPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGSEPA TSGSETPGTSESATPESGPGTSTEPSEGSAPGEPEA
43	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGS ETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPTTARAGSAENLSPSGLTGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTA VAWYQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTK VEIKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWV RQAPGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMD YWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNK RAPGTPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGET TGGSAESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNY ATYYADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAE AASASGTTARAGSAENLSPSGLTGPPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTS TEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPT STEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTST EPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSE PATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGS PAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPG SEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESG PGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGS APGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG
44	ASSPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGEEPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPLGGRADNHEPPELGAGATSGSETPGTDIQMTQSPSSLSASVGDRVTITCKASQDVSIGVAW

SEQ ID NO.	Amino Acid Sequences
	YQQKPGKAPKLLIYSASYRYTGVPSRFSGSGSGTDFTLTISSLQPEDFATYYCQQYYIYPYTFGQGTKVEI KGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFTFTDYTMDWVRQA PGKGLEWVADVNPNSGGSIYNQRFKGRFTLSVDRSKNTLYLQMNSLRAEDTAVYYCARNLGPSFYFDYWGQ GTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKRAPG TPARFSGSSLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETTGGS AESEPPGEGEVQLQESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYY ADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEAASA SGLGGRADNHEPPELGAGTPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPT STEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSES ATPESGPGTSESATPESGPGSEPATSGSETPGSESATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTST EPSEGSAPGSEPATSGSETPGTSESA
45	SPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGP GSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPES GPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEG SAPGGSAPTTGRAGEAEGATSAGATGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRASQDVNTAVA WYQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPTFGQGTKVE IKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKDTYIHWVRQ APGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGDGFYAMDYW GQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGLIGGTNKRA PGTPARFSGSSLGGSAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAETESPGETTG GSAESEPPGEGEVQLQESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYAT YYADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTVSSGTAEAA SASGTTGRAGEAEGATSAGATGPEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESAT PESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGS PTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTE PSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSE SATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTS ESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGT SESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESASPESGPGTSTEPSEGSAPG SPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEE GTSTEPSEGSAPGTSESATPESGPGTSESAT
46	SAGSPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEP SEGSAPGTSESATPESGPGSTPAESGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTE PSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTST EPSEGSAPGTSESATPESGPGSEPATSGSTETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGT SESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG TSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEE GTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESG PGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPES GPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPT STEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATS GSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESA TPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTE PSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSE SATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTS ESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGT

SEQ ID NO.	Amino Acid Sequences
	SESATPESGPGTESASTTGRAGEAANATSAGATGPATSGSETPGTDIQMTQSPSSLSASVGDRVTITCRAS QDVNTAVAWYQQKPGKAPKLLIYSASFLYSGVPSRFSGSRSGTDFTLTISSLQPEDFATYYCQQHYTTPPT FGQGTKVEIKGATPPETGAETESPGETTGGSAESEPPGEGEVQLVESGGGLVQPGGSLRLSCAASGFNIKD TYIHWVRQAPGKGLEWVARIYPTNGYTRYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCSRWGGD GFYAMDYWGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSTGAVTTSNYANWVQQKPGQAPRGL IGGTNKRAPGTPARFSGSLLGGKAALTLSGVQPEDEAEYYCALWYSNLWVFGGGTKLTVLGATPPETGAET ESPGETTGGSAESEPPGEGEVQLLESGGGLVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIR SKYNNYATYYADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYVSWFAYWGQGTLVTV SSGTAEAASASGTTGRAGEAANATSAGATGPPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSP AGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGS PAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPG TSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSTEPSEGSAP GTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSET PGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGS APGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPT STEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESAT PESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSES ATPESGPGftabTSESATPESGPGSEPATSGPTESGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAP GTESTPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGEPEA
47	ASSPAGSPTSTESGTSESATPESGPGTETEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES GPGSTPAESGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGESPATSGS TPEGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATP ESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPS EGSAPGGSAPTTGRATEAANATPAGLTGPATSGSETPGTEIVLTQSPATLSLSPGERATLSCKASQDVSIG VAWYQQKPGQAPRLLIYSASYRYSGVPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQYYIYPYTFGQGTK VEIKGATPPETGAETESPGETTGGSAESEPPGEGQVQLVQSGVEVKKPGASVKVSCKASGFTFTDYTMDWV RQAPGQGLEWMADVNPNSGGSIYNQRFKGRVTLTTDSSTTTAYMELKSLQFDDTAVYYCARNLGPSFYFDY WGQGTLVTVSSGGGSELVVTQEPSLTVSPGGTVTLTCRSSNGAVTSSNYANWVQQKPGQAPRGLIGGTNKR APGTPARFSGSLLGGKAALTLSGVQPEDEAVYYCALWYPNLWVFGGGTKLTVLGATPPETGAETESPGETT GGSAESEPPGEGEVQLLESGGGIVQPGGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYA TYYADSVKDRFTISRDDSKNTVYLQMNNLKTEDTAVYYCVRHENFGNSYVSWFAHWGQGTLVTVSSGTAEA ASASGTTGRATEAANATPAGLTGPPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPES GPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEG SAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSG SETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSP TSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPAT SGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSES ATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSE SATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTESTPSEGSAPGSE PATSGSETPGTSESATPESGPGTSTEPSEGSAPGEPEA

POLYPEPTIDE MIXTURE

[0219] Disclosed herein includes a mixture comprising a plurality of polypeptides of varying length; the mixture comprising a first set of polypeptides and a second set of polypeptides. In some embodiments, each polypeptide of the first set of polypeptides comprises a barcode fragment that (a) is releasable from the polypeptide by digestion with a protease and (b) has a sequence and molecular weight that differs from the sequence and molecular weight of all other fragments that are releasable from the first set of polypeptides. In

some embodiments, the second set of polypeptides lack the barcode fragment of the first set of polypeptides (e.g., due to truncation). In some embodiments, both the first set of polypeptides and the second set of polypeptides each comprise a reference fragment that (a) is common to the first set of polypeptides and the second set of polypeptides and (b) releasable by digestion with the protease. In some embodiments, the ratio of the first set of polypeptides to polypeptides comprising the reference fragment is greater than 0.70. In some embodiments, the ratio of the first set of polypeptides to polypeptides comprising the reference fragment is greater than 0.80, 0.90, 0.95, or 0.98. In some embodiments, the reference fragment occurs no more than once in each polypeptide of the first set of polypeptides and the second set of polypeptides. In some embodiments, the protease is a protease that cleaves on the C-terminal side of glutamic acid residues. In some embodiments, the protease is a Glu-C protease. In some embodiments, the protease is not trypsin. In some embodiments, the polypeptides of varying lengths comprise polypeptides comprising at least one extended recombinant polypeptide (XTEN), such as any described hereinabove or described anywhere else herein. In some embodiments, the first set of polypeptides comprises a full-length polypeptide, wherein the barcode fragment is a portion of the full-length polypeptide. In some embodiments, the full-length polypeptide is a (fusion) polypeptide, such as any described hereinabove or described anywhere else herein. In some embodiments, the barcode fragment lacks (does not comprise) both the N-terminal amino acid and C-terminal amino acid of the full-length polypeptide. In some embodiments, the mixture of polypeptides of varying lengths differ from one another due to N-terminal truncation, C-terminal truncation, or both N- and C-terminal truncation of a full-length polypeptide. In some embodiments, the first set of polypeptides and the second set of polypeptides may differ in one or more pharmacological properties. Non-limiting exemplary properties include.

METHOD OF POLYPEPTIDE CHARACTERIZATION

[0220] Disclosed herein includes a method for assessing, in a mixture comprising polypeptides of varying length, a relative amount of a first set of polypeptides in the mixture to a second set of polypeptides in the mixture, wherein (1) each polypeptide of the first set of polypeptides shares a barcode fragment that occurs once and only once in the polypeptide and (2) each polypeptide of the second set of polypeptides lacks the barcode fragment that is shared by polypeptides of the first set, wherein individual polypeptides of both the first of polypeptides and the second set of polypeptides each comprises a reference fragment. The method can comprise contacting the mixture with a protease to produce a plurality of proteolytic fragments that result from cleavage of the first set of polypeptides and the second set of polypeptides, wherein the plurality of proteolytic fragments comprise a plurality of reference fragments, and a plurality of barcode fragments. The method can further comprise determining a ratio of the amount of barcode fragments to the amount of reference fragments, thereby assessing the relative amounts of the first set of polypeptides to the second set of polypeptides. In some embodiments, the barcode fragment occurs no more than once in each polypeptide of the first set of polypeptides.

In some embodiments, the reference fragment occurs no more than once in each polypeptide of the first set of polypeptides and the second set of polypeptides. In some embodiments, the plurality of proteolytic fragments comprises a plurality of reference fragments, and a plurality of barcode fragments. In some embodiments, the protease cleaves the first and second sets of polypeptides (or the polypeptides of varying length) on the C-terminal side of glutamic acid residues that are not followed by a proline residue. In some embodiments, the protease is a Glu-C protease. In some embodiments, the protease is not trypsin. In some embodiments, the step of determining a ratio of the amount of barcode fragments to the amount of reference fragments comprises identifying barcode fragments and reference fragments from the mixture after it has been contacted with the protease. In some embodiments, the barcode fragments and the reference fragments are identified based on their respective masses. In some embodiments, the barcode fragments and the reference fragments are identified via mass spectrometry. In some embodiments, the barcode fragments and reference fragments are identified via liquid chromatography-mass spectrometry (LC-MS). In some embodiments, the step of determining a ratio of the barcode fragments to the reference fragments comprises isobaric labeling. In some embodiments, the step of determining a ratio of the barcode fragments to the reference fragments comprises spiking the mixture with one or both of an isotope-labeled reference fragment and an isotope labeled barcode fragment. In some embodiments, the polypeptides of varying lengths comprise polypeptides that comprise at least one extended recombinant polypeptide (XTEN), as described hereinabove or described anywhere else herein. In some embodiments, the XTEN is characterized in that (i) it comprises at least 100, or at least 150 amino acids; (ii) at least 90% of the amino acid residues of the XTEN are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P); and (iii) it comprises at least 4 different types of amino acids that are G, A, S, T, E, or P. In some embodiments, the barcode fragment, when present, is a portion of the XTEN. In some embodiments, the mixture of polypeptides of varying lengths comprises a polypeptide as any described hereinabove or described anywhere else herein. In some embodiments, the polypeptides of varying length comprise a full-length polypeptide and truncated fragments thereof. In some embodiments, the polypeptides of varying length consist essentially of the full-length polypeptide and truncated fragments thereof. In some embodiments, the mixture of polypeptides of varying lengths differ from one another due to N-terminal truncation, C-terminal truncation, or both N- and C-terminal truncation of a full-length polypeptide. In some embodiments, the full-length polypeptide is a polypeptide as described hereinabove or described anywhere else herein. In some embodiments, the ratio of the amount of barcode fragments to reference fragments is greater than 0.50, 0.60, 0.70, 0.80, 0.90, 0.95, 0.98, or 0.99.

Isobaric Labeling-Based Quantification of Peptides

[0221] In some embodiments, isobaric labeling can be used for determining a ratio of the barcode fragments to the reference fragments. One of ordinary skill will understand that isobaric labeling is a mass spectrometry

strategy used in quantitative proteomics, wherein peptides or proteins (or portions thereof) are labeled with various chemical groups that are isobaric (identical in mass) but vary in terms of distribution of heavy isotopes around their structure. These tags, commonly referred to as tandem mass tags, are designed so that the mass tag is cleaved at a specific linker region upon high-energy collision-induced dissociation (CID) during tandem mass spectrometry, thereby yielding reporter ions of different masses. One of ordinary skill will understand that one of the most common isobaric tags are amine-reactive tags.

[0222] The enhanced ability to detect and quantify truncation products (e.g., via isobaric labeling) can generate knowledge than can aid in designing manufacturing processes to include purification steps to minimize the presence of unwanted variants in the purified drug substance/product.

RECOMBINANT PRODUCTION

[0223] The disclosure herein includes a nucleic acid. The nucleic acid can comprise a polynucleotide (or polynucleotide sequence) encoding a (fusion) polypeptide, such as any described hereinabove or described anywhere else herein; or the nucleic acid can comprise the reverse complement of such a polynucleotide (or polynucleotide sequence).

[0224] The disclosure herein includes an expression vector that comprises a polynucleotide sequence, such as any described in the preceding paragraph, and a regulatory sequence operably linked to the polynucleotide sequence.

[0225] The disclosure herein includes a host cell comprising an expression vector, such as described any in the preceding paragraph. In some embodiments, the host cell is a prokaryote. In some embodiments, the host cell is *E. coli*. In some embodiments, the host cell is a mammalian cell.

[0226] In some embodiments, the disclosure provides methods of manufacturing the subject compositions. In one embodiment, the method comprises culturing a host cell comprising a nucleic acid construct that encodes a polypeptide or an XTEN-containing composition of any of the embodiments described herein under conditions that promote the expression of the polypeptide or BPXTEN fusion polypeptide, followed by recovery of the polypeptide or BPXTEN fusion polypeptide using standard purification methods (e.g., column chromatography, HPLC, and the like) wherein the composition is recovered wherein at least 70%, or at least 80%, or at least 90%, or at least 95%, or at least 97%, or at least 99% of the binding fragments of the expressed polypeptide or BPXTEN fusion polypeptide are correctly folded. In some embodiments of the method of making, the expressed polypeptide or BPXTEN fusion polypeptide is recovered in which at least or at least 90%, or at least 95%, or at least 97%, or at least 99% of the polypeptide or BPXTEN fusion polypeptide is recovered in monomeric, soluble form.

[0227] In some embodiments, the disclosure relates to methods of making the polypeptide and BPXTEN fusion polypeptide at high fermentation expression levels of functional protein using an *E. coli* or mammalian host

cell, as well as providing expression vectors encoding the constructs useful in methods to produce the cytotoxically active polypeptide construct compositions at high expression levels. In one embodiment, the method comprises the steps of 1) preparing the polynucleotide encoding the polypeptides of any of the embodiments disclosed herein, 2) cloning the polynucleotide into an expression vector, which can be a plasmid or other vector under control of appropriate transcription and translation sequences for high level protein expression in a biological system, 3) transforming an appropriate host cell with the expression vector, and 4) culturing the host cell in conventional nutrient media under conditions suitable for the expression of the polypeptide composition. Where desired, the host cell is *E. coli*. By the method, the expression of the polypeptide results in fermentation titers of at least 0.05 g/L, or at least 0.1 g/L, or at least 0.2 g/L, or at least 0.3 g/L, or at least 0.5 g/L, or at least 0.6 g/L, or at least 0.7 g/L, or at least 0.8 g/L, or at least 0.9 g/L, or at least 1 g/L, or at least 2 g/L, or at least 3 g/L, or at least 4 g/L, or at least 5 g/L of the expression product of the host cell and wherein at least 70%, or at least 80%, or at least 90%, or at least 95%, or at least 97%, or at least 99% of the expressed protein are correctly folded. As used herein, the term "correctly folded" means that the antigen binding fragments component of the composition have the ability to specifically bind its target ligand. In some embodiments, the disclosure provides a method for producing a polypeptide or BPXTEN fusion polypeptide, the method comprising culturing in a fermentation reaction a host cell that comprises a vector encoding a polypeptide comprising the polypeptide or BPXTEN fusion polypeptide under conditions effective to express the polypeptide product at a concentration of more than about 10 milligrams/gram of dry weight host cell (mg/g), or at least about 250 mg/g, or about 300 mg/g, or about 350 mg/g, or about 400 mg/g, or about 450 mg/g, or about 500 mg/g of the polypeptide when the fermentation reaction reaches an optical density of at least 130 at a wavelength of 600 nm, and wherein the antigen binding fragments of the expressed protein are correctly folded. In some embodiments, the disclosure provides a method for producing a polypeptide or BPXTEN fusion polypeptide, the method comprising culturing in a fermentation reaction a host cell that comprises a vector encoding the composition under conditions effective to express the polypeptide product at a concentration of more than about 10 milligrams/gram of dry weight host cell (mg/g), or at least about 250 mg/g, or about 300 mg/g, or about 350 mg/g, or about 400 mg/g, or about 450 mg/g, or about 500 mg/g of the polypeptide when the fermentation reaction reaches an optical density of at least 130 at a wavelength of 600 nm, and wherein the expressed polypeptide product is soluble.

PHARMACEUTICAL COMPOSITION

[0228] Disclosed herein includes a pharmaceutical composition comprising a (fusion) polypeptide, such as any described hereinabove or described anywhere else herein, and one or more pharmaceutically acceptable excipients. In some embodiments, the pharmaceutical composition is formulated for intradermal, subcutaneous, oral, intravenous, intra-arterial, intraabdominal, intraperitoneal, intravitreal, intrathecal, or intramuscular

administration. In some embodiments, the pharmaceutical composition is in a liquid form or frozen. In some embodiments, the pharmaceutical composition is in a device that is implanted into the eye or another body part. In some embodiments, the pharmaceutical composition is in a pre-filled syringe for a single injection. In some embodiments, the pharmaceutical composition is formulated as a lyophilized powder to be reconstituted prior to administration.

[0229] In some embodiments, the dose is administered intradermally, subcutaneously, orally, intravenously, intravitreally (or otherwise injected into the eye), intra-arterially, intra-abdominally, intraperitoneally, intrathecally, or intramuscularly. In some embodiments, the pharmaceutical composition is administered using a device implanted into the eye or other body part. In some embodiments, the subject is a mouse, rat, monkey, or human.

[0230] The pharmaceutical compositions can be administered for therapy by any suitable route. In addition, the pharmaceutical compositions can also contain other pharmaceutically active compounds or a plurality of compounds of the invention.

[0231] In some embodiments, the pharmaceutical composition can be administered subcutaneously, orally, intramuscularly, or intravenously. In one embodiment, the pharmaceutical composition is administered at a therapeutically effective dose. In some cases of the foregoing, the therapeutically effective dose results in a gain in time spent within a therapeutic window for the fusion protein compared to the corresponding BP of the fusion protein not linked to the XTEN and administered at a comparable dose to a subject. The gain in time spent within the therapeutic window can at least threefold greater than the corresponding BP not linked to the XTEN, or alternatively, at least four- fold, or five-fold, or six-fold, or seven- fold, or eight- fold, or nine-fold, or at least 10-fold, or at least 20-fold greater than the corresponding BP not linked to the XTEN.

[0232] In some embodiments, invention provides a method of treating a disease, disorder or condition, comprising administering the pharmaceutical composition described above to a subject using multiple consecutive doses of the pharmaceutical composition administered using a therapeutically effective dose regimen. In one embodiment of the foregoing, the therapeutically effective dose regimen can result in a gain in time of at least three-fold, or alternatively, at least four- fold, or five-fold, or six-fold, or sevenfold, or eight-fold, or nine-fold, or at least 10-fold, or at least 20-fold between at least two consecutive C_{max} peaks and/or C_{min} troughs for blood levels of the fusion protein compared to the corresponding BP of the fusion protein not linked to XTEN(s) and administered at a comparable dose regimen to a subject. In some embodiments of the foregoing, the administration of the fusion protein results in a comparable improvement in at least one measured parameter using less frequent dosing or a lower total dosage in moles of the fusion protein of the pharmaceutical composition compared to the corresponding biologically active protein component(s) not linked to XTEN(s) and administered to a subject using a therapeutically effective regimen to a subject.

[0233] In one embodiment, the pharmaceutical composition is administered subcutaneously. In this embodiment, the composition may be supplied as a lyophilized powder to be reconstituted prior to administration. The composition may also be supplied in a liquid form or frozen, which can be administered directly to a patient. In one embodiment, the composition is supplied as a liquid in a pre-filled syringe such that a patient can easily self-administer the composition.

[0234] Extended release formulations useful in the present invention may be oral formulations comprising a matrix and a coating composition. Suitable matrix materials may include waxes (e.g., camauba, bees wax, paraffin wax, ceresine, shellac wax, fatty acids, and fatty alcohols), oils, hardened oils or fats (e.g., hardened rapeseed oil, castor oil, beef tallow, palm oil, and soya bean oil), and polymers (e.g., hydroxypropyl cellulose, polyvinylpyrrolidone, hydroxypropyl methyl cellulose, and polyethylene glycol). Other suitable matrix tableting materials are microcrystalline cellulose, powdered cellulose, hydroxypropyl cellulose, ethyl cellulose, with other carriers, and fillers. Tablets may also contain granulates, coated powders, or pellets. Tablets may also be multi-layered. Multi-layered tablets are especially preferred when the active ingredients have markedly different pharmacokinetic profiles. Optionally, the finished tablet may be coated or uncoated.

[0235] The coating composition may comprise an insoluble matrix polymer and/or a water-soluble material. Water soluble materials can be polymers such as polyethylene glycol, hydroxypropyl cellulose, hydroxypropyl methyl cellulose, polyvinylpyrrolidone, polyvinyl alcohol, or monomeric materials such as sugars (e.g., lactose, sucrose, fructose, mannitol and the like), salts (*e.g.*, sodium chloride, potassium chloride and the like), organic acids (e.g., fumaric acid, succinic acid, lactic acid, and tartaric acid), and mixtures thereof. Optionally, an enteric polymer may be incorporated into the coating composition. Suitable enteric polymers include hydroxypropyl methyl cellulose, acetate succinate, hydroxypropyl methyl cellulose, phthalate, polyvinyl acetate phthalate, cellulose acetate phthalate, cellulose acetate trimellitate, shellac, zein, and polymethacrylates containing carboxyl groups. The coating composition may be plasticised by adding suitable plasticisers such as, for example, diethyl phthalate, citrate esters, polyethylene glycol, glycerol, acetylated glycerides, acetylated citrate esters, dibutylsebacate, and castor oil. The coating composition may also include a filler, which can be an insoluble material such as silicon dioxide, titanium dioxide, talc, kaolin, alumina, starch, powdered cellulose, MCC, or polacrilin potassium. The coating composition may be applied as a solution or latex in organic solvents or aqueous solvents or mixtures thereof. Solvents such as water, lower alcohol, lower chlorinated hydrocarbons, ketones, or mixtures thereof may be used.

[0236] BPXTEN polypeptides of the present invention can be formulated according to known methods to prepare pharmaceutically useful compositions, whereby the polypeptide is combined in admixture with a pharmaceutically acceptable carrier vehicle, such as aqueous solutions or buffers, pharmaceutically acceptable suspensions and emulsions. Examples of non-aqueous solvents include propyl ethylene glycol, polyethylene

glycol and vegetable oils. Therapeutic formulations are prepared for storage by mixing the active ingredient having the desired degree of purity with optional physiologically acceptable carriers, excipients or stabilizers, as described in Remington's Pharmaceutical Sciences 16th edition, Osol, A. Ed. (1980), in the form of lyophilized formulations or aqueous solutions. The compositions of the invention may be formulated using a variety of excipients. Suitable excipients include microcrystalline cellulose (e.g. Avicel PH 102, Avicel PHIOI), polymethacrylate, poly(ethyl acrylate, methyl methacrylate, trimethylammonioethyl methacrylate chloride) (such as Eudragit RS-30D), hydroxypropyl methylcellulose (Methocel KIOOM, Premium CR Methocel KIOOM, Methocel E5, Opadry®), magnesium stearate, talc, triethyl citrate, aqueous ethylcellulose dispersion (Surelease®), and protamine sulfate. The slow release agent may also comprise a carrier, which can comprise, for example, solvents, dispersion media, coatings, antibacterial and antifungal agents, isotonic and absorption delaying agents. Pharmaceutically acceptable salts can also be used in these slow release agents, for example, mineral salts such as hydrochlorides, hydrobromides, phosphates, or sulfates, as well as the salts of organic acids such as acetates, proprionates, malonates, or benzoates. The composition may also contain liquids, such as water, saline, glycerol, and ethanol, as well as substances such as wetting agents, emulsifying agents, or pH buffering agents. Liposomes may also be used as a carrier.

[0237] In some embodiments, the compositions of the present invention are encapsulated in liposomes, which have demonstrated utility in delivering beneficial active agents in a controlled manner over prolonged periods of time. Liposomes are closed bilayer membranes containing an entrapped aqueous volume. Liposomes may also be unilamellar vesicles possessing a single membrane bilayer or multilamellar vesicles with multiple membrane bilayers, each separated from the next by an aqueous layer. The structure of the resulting membrane bilayer is such that the hydrophobic (non-polar) tails of the lipid are oriented toward the center of the bilayer while the hydrophilic (polar) heads orient towards the aqueous phase. In one embodiment, the liposome may be coated with a flexible water-soluble polymer that avoids uptake by the organs of the mononuclear phagocyte system, primarily the liver and spleen. Suitable hydrophilic polymers for surrounding the liposomes include, without limitation, PEG, polyvinylpyrrolidone, polyvinylmethylether, polymethyloxazoline, polyethyloxazoline, polyhydroxypropyloxazoline, polyhydroxypropylmethacrylamide, polymethacrylamide, polydimethylacrylamide, polyhydroxypropylmethacrylate, polyhydroxethylacrylate, hydroxymethylcellulose hydroxyethylcellulose, polyethyleneglycol, polyaspartamide and hydrophilic peptide sequences as described in U.S. Pat. Nos. 6,316,024; 6,126,966; 6,056,973; 6,043,094, the contents of which are incorporated by reference in their entirety.

[0238] Liposomes may be comprised of any lipid or lipid combination known in the art. For example, the vesicle-forming lipids may be naturally-occurring or synthetic lipids, including phospholipids, such as phosphatidylcholine, phosphatidylethanolamine, phosphatidic acid, phosphatidylserine, phosphatidylglycerol,

phosphatidylinositol, and sphingomyelin as disclosed in U.S. Pat. Nos. 6,056,973 and 5,874,104. The vesicle-forming lipids may also be glycolipids, cerebrosides, or cationic lipids, such as 1,2-dioleyloxy-3-(trimethylamino) propane (DOTAP); N-[l-(2,3,-ditetradecyloxy)propyl]-N,N- dimethyl-N-hydroxyethylammonium bromide (DMRIE); N-[I [(2,3,-dioleyloxy)propyl]-N,N-dimethyl- N-hydroxy ethylammonium bromide (DORIE); N-[I -(2,3 -dioleyloxy)propyl] -N,N,N- trimethylammonium chloride (DOTMA); 3 [N-(N',N'-dimethylaminoethane) carbamoyl] cholesterol (DC-Choi); or dimethyldioctadecylammonium (DDAB) also as disclosed in U.S. Pat. No. 6,056,973. Cholesterol may also be present in the proper range to impart stability to the vesicle as disclosed in U.S. Pat. Nos. 5,916,588 and 5,874,104.

[0239] Additional liposomal technologies are described in U.S. Pat. Nos. 6,759,057; 6,406,713; 6,352,716; 6,316,024; 6,294,191; 6,126,966; 6,056,973; 6,043,094; 5,965,156; 5,916,588; 5,874,104; 5,215,680; and 4,684,479, the contents of which are incorporated herein by reference. These describe liposomes and lipid-coated microbubbles, and methods for their manufacture. Thus, one skilled in the art, considering both the disclosure of this invention and the disclosures of these other patents could produce a liposome for the extended release of the polypeptides of the present invention. For liquid formulations, a desired property is that the formulation be supplied in a form that can pass through a 25-, 28-, 30-, 31-, 32-gauge needle for intravenous, intramuscular, intraarticular, or subcutaneous administration.

[0240] Administration via transdermal formulations can be performed using methods also known in the art, including those described generally in, e.g., U.S. Pat. Nos. 5,186,938 and 6,183,770, 4,861,800, 6,743,211, 6,945,952, 4,284,444, and WO 89/09051, incorporated herein by reference in their entireties. A transdermal patch is a particularly useful embodiment with polypeptides having absorption problems. Patches can be made to control the release of skin-permeable active ingredients over a 12 hour, 24 hour, 3 day, and 7 day period. In one example, a 2-fold daily excess of a polypeptide of the present invention is placed in a non-volatile fluid. The compositions of the invention are provided in the form of a viscous, non-volatile liquid. The penetration through skin of specific formulations may be measures by standard methods in the art (for example, Franz et al., J. Invest. Derm. 64: 194-195 (1975)). Examples of suitable patches are passive transfer skin patches, iontophoretic skin patches, or patches with microneedles such as Nicoderm. In other embodiments, the composition may be delivered via intranasal, buccal, or sublingual routes to the brain to enable transfer of the active agents through the olfactory passages into the CNS and reducing the systemic administration. Devices commonly used for this route of administration are included in U.S. Pat. No. 6,715,485. Compositions delivered via this route may enable increased CNS dosing or reduced total body burden reducing systemic toxicity risks associated with certain drugs. Preparation of a pharmaceutical composition for delivery in a subdermally

implantable device can be performed using methods known in the art, such as those described in, e.g., U.S. Pat. Nos. 3,992,518; 5,660,848; and 5,756,115.

[0241] Osmotic pumps may be used as slow release agents in the form of tablets, pills, capsules or implantable devices. Osmotic pumps are well known in the art and readily available to one of ordinary skill in the art from companies experienced in providing osmotic pumps for extended release drug delivery. Examples are ALZA's DUROS™; ALZA's OROS™; Osmotica Pharmaceutical's Osmodex™ system; Shire Laboratories' EnSoTrol™ system; and Alzet™. Patents that describe osmotic pump technology are U.S. Pat. Nos. 6,890,918; 6,838,093; 6,814,979; 6,713,086; 6,534,090; 6,514,532; 6,361,796; 6,352,721; 6,294,201; 6,284,276; 6,110,498; 5,573,776; 4,200,0984; and 4,088,864, the contents of which are incorporated herein by reference. One skilled in the art, considering both the disclosure of this invention and the disclosures of these other patents could produce an osmotic pump for the extended release of the polypeptides of the present invention.

[0242] Syringe pumps may also be used as slow release agents. Such devices are described in U.S. Pat. Nos. 4,976,696; 4,933,185; 5,017,378; 6,309,370; 6,254,573; 4,435,173; 4,398,908; 6,572,585; 5,298,022; 5,176,502; 5,492,534; 5,318,540; and 4,988,337, the contents of which are incorporated herein by reference. One skilled in the art, considering both the disclosure of this invention and the disclosures of these other patents could produce a syringe pump for the extended release of the compositions of the present invention.

PHARMACEUTICAL KITS

[0243] In some embodiments, the invention provides a kit to facilitate the use of the BPXTEN polypeptides. In one embodiment, the kit comprises, in at least a first container: (a) an amount of a BPXTEN fusion protein composition sufficient to treat a disease, condition or disorder upon administration to a subject in need thereof; and (b) an amount of a pharmaceutically acceptable carrier; together in a formulation ready for injection or for reconstitution with sterile water, buffer, or dextrose; together with a label identifying the BPXTEN drug and storage and handling conditions, and a sheet of the approved indications for the drug, instructions for the reconstitution and/or administration of the BPXTEN drug for the use for the prevention and/or treatment of an approved indication, appropriate dosage and safety information, and information identifying the lot and expiration of the drug. In some embodiments of the foregoing, the kit can comprise a second container that can carry a suitable diluent for the BPXTEN composition, which will provide the user with the appropriate concentration of BPXTEN to be delivered to the subject.

METHOD OF TREATMENT

[0244] Disclosed herein includes use of a polypeptide, such as any described hereinabove or described anywhere else herein, in the preparation of a medicament for the treatment of a disease in a subject. In some embodiments, the particular disease to be treated will depend on the choice of the biologically active proteins.

In some embodiments, the disease is cancer (including any form thereof) In some cases, the cancer or tumor can be characterized by a low, medium, or high level of HER2 expression.

[0245] Disclosed herein includes a method of treating a disease in a subject, the method comprising administering to the subject in need thereof one or more therapeutically effective doses of the pharmaceutical composition, such as any described hereinabove or described anywhere else herein. In some embodiments, the disease is cancer (including any form thereof) In some embodiments, the subject is a mouse, rat, monkey, and human. In some cases, the cancer or tumor can be characterized by a low, medium, or high level of HER2 expression.

[0246] In certain embodiments, the HER-2 targeted bispecific compositions of the present invention (and particularly AMX818) may be advantageously combined with a second therapeutic agent effective for treating or ameliorating the effects of the cancer. The additional therapeutic agent may be selected from the group consisting of an antibody, an antibody fragment, an antibody conjugate, a cytotoxic agent, a toxin, a radionuclide, an immunomodulator, a photoactive therapeutic agent, a radiosensitizing agent, a hormone, an anti-angiogenesis agent, and combinations thereof. Particularly preferred second or additional therapeutic agents include other HER2 targeting agents, chemotherapy agents, radiotherapeutic agents, as well as agents that target HER3 and other targets that are involved in resistance to treatment of HER2-driven cancers.

[0247] Examples of therapeutic antibodies that may be used in the present invention include rituximab (Rituxan), Brentuximab Vedotin (Adcetriz), Ado-trastuzumab emtansine (Kadcyla), Cetuximab (Erbitux), bevacizumab (Avastin), Ibritumomab (Zevalin), vedolizumab (Entyvio), Ipilimumab (Yervoy), Nivolumab (Opdivo), pembrolizumab (Keytruda), Alemtuzamab atezolizumab (Tecentriq), avelumab (Bavencio), durvalumab (Imfinzi), B-701, Ofatumumab, Obinutuzumab (Gazyva) Panitumumab, plozalizumab, BI-754091, OREG-103, COM-701, BI-754111, and combinations thereof.

[0248] According to some embodiments, the antibody, fragment thereof, or conjugate thereof is selected from the group consisting of rituximab (Rituxan), Brentuximab Vedotin (Adcetriz), Ado-trastuzumab emtansine (Kadcyla), Ipilimumab (Yervoy), Nivolumab (Opdivo), pembrolizumab (Keytruda), Alemtuzamab atezolizumab (Tecentriq), durvalumab (Imfinzi), Ofatumumab, Obinutuzumab (Gazyva) Panitumumab, and combinations thereof.

[0249] In other embodiments, the additional agent may be a DNA damaging agent, antimetabolite, anti-microtubule agent, antibiotic agent, etc. DNA damaging agents include alkylating agents, platinum-based agents, intercalating agents, and inhibitors of DNA replication. Non-limiting examples of DNA alkylating agents include cyclophosphamide, mechlorethamine, uramustine, melphalan, chlorambucil, ifosfamide, carmustine, lomustine, streptozocin, busulfan, temozolomide, pharmaceutically acceptable salts thereof, prodrugs, and combinations thereof. Non-limiting examples of platinum-based agents include cisplatin,

carboplatin, oxaliplatin, nedaplatin, satraplatin, triplatin tetranitrate, pharmaceutically acceptable salts thereof, prodrugs, and combinations thereof. Non-limiting examples of intercalating agents include doxorubicin, daunorubicin, idarubicin, mitoxantrone, pharmaceutically acceptable salts thereof, prodrugs, and combinations thereof. Non-limiting examples of inhibitors of DNA replication include irinotecan, topotecan, amsacrine, etoposide, etoposide phosphate, teniposide, pharmaceutically acceptable salts thereof, prodrugs, and combinations thereof. Antimetabolites include folate antagonists such as methotrexate and premetrexed, purine antagonists such as 6-mercaptopurine, dacarbazine, and fludarabine, and pyrimidine antagonists such as 5-fluorouracil, arabinosylcytosine, capecitabine, gemcitabine, decitabine, pharmaceutically acceptable salts thereof, prodrugs, and combinations thereof. Anti-microtubule agents include without limitation vinca alkaloids, paclitaxel (Taxol®), docetaxel (Taxotere®), and ixabepilone (Ixempra®). Antibiotic agents include without limitation actinomycin, anthracyclines, valrubicin, epirubicin, bleomycin, plicamycin, mitomycin, pharmaceutically acceptable salts thereof, prodrugs, and combinations thereof.

[0250] Exemplary cytotoxic agents are know to those of skill in the art, and may, for example, be selected from the group consisting of cyclophosphamide, mechlorethamine, uramustine, melphalan, chlorambucil, ifosfamide, carmustine, lomustine, streptozocin, busulfan, temozolomide, cisplatin, carboplatin, oxaliplatin, nedaplatin, satraplatin, triplatin tetranitrate, doxorubicin, daunorubicin, idarubicin, mitoxantrone, methotrexate, pemetrexed, 6-mercaptopurine, dacarbazine, fludarabine, 5-fluorouracil, arabinosylcytosine, capecitabine, gemcitabine, decitabine, vinca alkaloids, paclitaxel (Taxol), docetaxel (Taxotere), ixabepilone (Ixempra), actinomycin, anthracyclines, valrubicin, epirubicin, bleomycin, plicamycin, mitomycin, pharmaceutically acceptable salts thereof, prodrugs, and combinations thereof.

[0251] Cytotoxic agents according to the present invention also include an inhibitor of the PI3K/Akt pathway. Non-limiting examples of an inhibitor of the PI3K/Akt pathway include A-674563 (CAS #552325-73-2), AGL 2263, AMG-319 (Amgen, Thousand Oaks, Calif.), AS-041164 (5-benzo[1,3]dioxol-5-ylmethylene-thiazolidine-2,4-dione), AS-604850 (5-(2,2-Difluoro-benzo[1,3]dioxol-5-ylmethylene)-thiazolidine-2,4-dione), AS-605240 (5-quinoxilin-6-methylene-1,3-thiazolidine-2,4-dione), AT7867 (CAS #857531-00-1), benzimidazole series, Genentech (Roche Holdings Inc., South San Francisco, Calif.), BML-257 (CAS #32387-96-5), BVD-723, CAL-120 (Gilead Sciences, Foster City, Calif.), CAL-129 (Gilead Sciences), CAL-130 (Gilead Sciences), CAL-253 (Gilead Sciences), CAL-263 (Gilead Sciences), CAS #612847-09-3, CAS #681281-88-9, CAS #75747-14-7, CAS #925681-41-0, CAS #98510-80-6, CCT128930 (CAS #885499-61-6), CH5132799 (CAS #1007207-67-1), CHR-4432 (Chroma Therapeutics, Ltd., Abingdon, UK), FPA 124 (CAS #902779-59-3), GS-1101 (CAL-101) (Gilead Sciences), GSK 690693 (CAS #937174-76-0), H-89 (CAS #127243-85-0), Honokiol, IC87114 (Gilead Science), IPI-145 (Intellikine Inc.), KAR-4139 (Karus Therapeutics, Chilworth, UK), KAR-4141 (Karus Therapeutics), KIN-1 (Karus Therapeutics), KT 5720 (CAS

#108068-98-0), Miltefosine, MK-2206 dihydrochloride (CAS #1032350-13-2), ML-9 (CAS #105637-50-1), Naltrindole Hydrochloride, OXY-111A (NormOxys Inc., Brighton, Mass.), perifosine, PHT-427 (CAS #1191951-57-1), PI3 kinase delta inhibitor, Merck KGaA (Merck & Co., Whitehouse Station, N.J.), PI3 kinase delta inhibitors, Genentech (Roche Holdings Inc.), PI3 kinase delta inhibitors, Incozen (Incozen Therapeutics, Pvt. Ltd., Hydrabad, India), PI3 kinase delta inhibitors-2, Incozen (Incozen Therapeutics), PI3 kinase inhibitor, Roche-4 (Roche Holdings Inc.), PI3 kinase inhibitors, Roche (Roche Holdings Inc.), PI3 kinase inhibitors, Roche-5 (Roche Holdings Inc.), PI3-alpha/delta inhibitors, Pathway Therapeutics (Pathway Therapeutics Ltd., South San Francisco, Calif.), PI3-delta inhibitors, Cellzome (Cellzome AG, Heidelberg, Germany), PI3-delta inhibitors, Intellikine (Intellikine Inc., La Jolla, Calif.), PI3-delta inhibitors, Pathway Therapeutics-1 (Pathway Therapeutics Ltd.), PI3-delta inhibitors, Pathway Therapeutics-2 (Pathway Therapeutics Ltd.), PI3-delta/gamma inhibitors, Cellzome (Cellzome AG), PI3-delta/gamma inhibitors, Cellzome (Cellzome AG), PI3-delta/gamma inhibitors, Intellikine (Intellikine Inc.), PI3-delta/gamma inhibitors, Intellikine (Intellikine Inc.), PI3-delta/gamma inhibitors, Pathway Therapeutics (Pathway Therapeutics Ltd.), PI3-delta/gamma inhibitors, Pathway Therapeutics (Pathway Therapeutics Ltd.), PI3-gamma inhibitor Evotec (Evotec), PI3-gamma inhibitor, Cellzome (Cellzome AG), PI3-gamma inhibitors, Pathway Therapeutics (Pathway Therapeutics Ltd.), PI3K delta/gamma inhibitors, Intellikine-1 (Intellikine Inc.), PI3K delta/gamma inhibitors, Intellikine-1 (Intellikine Inc.), pictilisib (Roche Holdings Inc.), PIK-90 (CAS #677338-12-4), SC-103980 (Pfizer, New York, N.Y.), SF-1126 (Semafore Pharmaceuticals, Indianapolis, Ind.), SH-5, SH-6, Tetrahydro Curcumin, TG100-115 (Targegen Inc., San Diego, Calif.), Triciribine, X-339 (Xcovery, West Palm Beach, Fla.), XL-499 (Evotech, Hamburg, Germany), pharmaceutically acceptable salts thereof, and combinations thereof.

[0252] The additional agent in the combination therapy may be a poison or venom of plant or animal origin. An example is diphtheria toxin or portions thereof. In other examples, the additional agent may be a "radionuclide" i.e., a radioactive substance administered to the patient, e.g., intravenously or orally, after which it penetrates via the patient's normal metabolism into the target organ or tissue, where it delivers local radiation for a short time. Examples of radionuclides include, but are not limited to, I-125, At-211, Lu-177, Cu-67, I-131, Sm-153, Re-186, P-32, Re-188, In-114m, and Y-90.

[0253] The term "immunomodulator" means a substance that alters the immune response by augmenting or reducing the ability of the immune system to produce antibodies or sensitized cells that recognize and react with the antigen that initiated their production. Immunomodulators may be recombinant, synthetic, or natural preparations and include cytokines, corticosteroids, cytotoxic agents, thymosin, and immunoglobulins. Some immunomodulators are naturally present in the body, and certain of these are available in pharmacologic preparations. Examples of immunomodulators include, but are not limited to, granulocyte colony-stimulating factor (G-CSF), LAG-3, IMP-321, JCAR-014, ASLAN-002 (BMS-777607), interferons, imiquimod and

cellular membrane fractions from bacteria, IL-2, IL-7, IL-12, CCL3, CCL26, CXCL7, synthetic cytosine phosphate-guanosine (CpG), immune-checkpoint inhibitors, and combinations thereof. Targeted cytokine therapy, particularly targeting to lymphotoxin and LiGHT may also be useful in combination with the compositions of the present invention.

[0254] In some combination treatments, the additional agent may be a "radiosensitizing agent" that makes tumor cells more sensitive to radiation therapy. Examples of radiosensitizing agents include misonidazole, metronidazole, tirapazamine, and trans sodium crocetinate, and combination thereof.

[0255] In still other embodiments, the additional agent is an "anti-angiogenesis" agent that reduces or inhibits the growth of new blood vessels, such as, e.g., an inhibitor of vascular endothelial growth factor (VEGF) and an inhibitor of endothelial cell migration. Anti-angiogenesis agents include without limitation 2-methoxyestradiol, angiostatin, bevacizumab, cartilage-derived angiogenesis inhibitory factor, endostatin, IFN-α, IL-12, itraconazole, linomide, platelet factor-4, prolactin, SU5416, suramin, tasquinimod, tecogalan, tetrathiomolybdate, thalidomide, thrombospondin, thrombospondin, TNP-470, ziv-aflibercept, pharmaceutically acceptable salts thereof, prodrugs, and combinations thereof.

[0256] In particularly preferred embodiments, the HER2-targeted bispecific compositions of the present invention may be combined with checkpoint inhibitors, such as anti-PD-1/anti-PD-L1 compositions, CTLA-4, OX40 and the like. In particular embodiments of such combination therapy, the compositions of the present invention can be combined with antagonists of the cell surface receptor programmed cell death protein 1, also known as PD-1 as well as, antagonists of PD-L1.

[0257] PD-1 plays an important role in down-regulating the immune system and promoting self-tolerance by suppressing T cell inflammatory activity. Binding of the PD-1 ligands, PD-L1 and PD-L2 to the PD-1 receptor found in T cells inhibits T-cell proliferation and cytokine production. Upregulation of PD-1 ligands occurs in some tumors and signaling through this pathway can contribute to inhibition of active T-cell immune surveillance of tumors. Anti-PD-1 antibodies bind to the PD-1 receptor and block its interaction with PD-L1 and PD-L3, releasing PD-1 pathway-mediated inhibition of the immune response, including the anti-tumor immune response.

[0258] Those of skill in the art are aware of various anti-PD-1 antibodies that may be used. In some embodiments, an exemplary anti-PD-1 antibody used in combination with the compounds of the present invention is Pembrolizumab (Keytruda®). In other embodiments, the anti-PD-1 antibody used in combination with the compound described above is Nivolumab (Opdivo®). In other embodiments, the anti-PD-1 antibody used in combination with the compound described above is Pidilizumab (Medivation).

[0259] Additional PD-1 antibodies known to those of skill in the art, include AGEN-2034 (Agenus), AMP-224 (Medimmune), BCD-100 (Biocad), BGBA-317 (Beigene), BI-754091 (Boehringer Ingelheim), CBT-501

(Genor Biopharma), CC-90006 (Celgene), cemiplimab (Regeneron Pharmaceuticals), durvalumab+MEDI-0680 (Medimmune), GLS-010 (Harbin Gloria Pharmaceuticals), IBI-308 (Eli Lilly), JNJ-3283 (Johnson & Johnson), JS-001 (Shanghai Junshi Bioscience Co.), MEDI-0680 (Medimmune), MGA-012 (MacroGenics), MGD-013 (Marcogenics), pazopanib hydrochloride+pembrolizumab (Novartis), PDR-001 (Novartis), PF-06801591 (Pfizer), REGN-2810 (Regeneron), SHR-1210 (Jiangsu Hengrui Medicine Co.), TSR-042 (Tesaro Inc.), LZM-009 (Livzon Pharmaceutical Group Inc) and ABBV-181 (AbbVie Inc). Each possibility represents a separate embodiment of the present invention.

[0260] In one preferred embodiment for combination therapy of the present invention, the anti-PD-1 antibody is pembrolizumab (Keytruda®).

[0261] In other embodiments, the compositions of the present invention are combined with an anti-PD-L1 antibody. Exemplary such anti-PD-L1 antibodies used in the combinations of the present invention may be selected from the group consisting of Durvalumab (MedImmune LLC), Atezolizumab (Hoffmann-La Roche Ltd, Chugai Pharmaceutical Co Ltd), Avelumab (Merck KGaA), CX-072 (CytomX Therapeutics Inc), BMS-936559 (ViiV Healthcare Ltd), SHR-1316 (Jiangsu Hengrui Medicine Co Ltd), M-7824 (Merck KGaA), LY-3300054 (Eli Lilly and Co), FAZ-053 (Novartis AG), KN-035 (AlphaMab Co Ltd), CA-170 (Curis Inc), CK-301 (TG Therapeutics Inc), CS-1001 (CStone Pharmaceuticals Co Ltd), HLX-10 (Shanghai Henlius Biotech Co Ltd), MCLA-145 (Merus NV), MSB-2311 (MabSpace Biosciences (Suzhou) Co Ltd) and MEDI-4736 (Medimmune).

[0262] In certain other embodiments, the combination therapies of the present invention may include other HER2-direct therapies. For example, it may be advantageous to combine the compositions of the present invention with trastuzumab and related HER2 based (i.e., those that target the trastuzumab epitope) therapies such as Trastuzumab, Trastuzumab based ADCs, Kadcyla, Enhertu, HER2 tyrosine kinase inhibitors such as lapatinib, neratinib, tucatinib, HER2 targeting immunoconjugates, such as TLRs, and cytokines. Other immunotherapies and checkpoint inhibitor-based therapies that may be useful in combination with the compositions of the present invention include CTLA4, TIGIT, OX40, PD1, PDL1, TIM3-based therapies. The compositions of the invention may further be combined with CAR-T, NK, or T-cell based therapies, as well as with immunotherapy vaccines. Combination with cytokines and therapies that are cytokine targeting also are contemplated. Of particular interest may be use of TGFb, VEGF and VEGFR1-3 and anti-angiogenesis targeting therapies such as Avastin, Ramucirumab, or tyrosine kinase inhibitors such as axitinib, lenvatinib, cabozantinib, regorafenib, sunitib, sorafenib. In other embodiments, CDK4/6 inhibitors such as palbociclib and the like may be used. EGFR inhibitors such as cetuximab, panitumumab, erlotinib, and Osimertinib also may be of use in combination therapies.

[0263] The following are examples of compositions and evaluations of compositions of the disclosure. It is understood that various other embodiments may be practiced, given the general description provided above.

EXAMPLES

Example 1. Design of barcoded XTEN by minimal mutations from general-purpose XTEN

[0264] This example illustrates an exemplary design approach to barcoded XTEN by making minimal mutation(s) of the amino acid sequence of a general-purpose XTEN (such as one of Table 3b hereinabove). The relevant criteria for performing minimal mutation(s) include one or more of the following: (a) to minimize the sequence change of the corresponding XTEN; (b) to minimize the amino acid composition change in the corresponding XTEN; (c) to substantially maintain the net charge in the corresponding XTEN; (d) to substantially maintain the low immunogenicity of the corresponding XTEN; (e) to substantially maintain the pharmacokinetic properties afforded by the XTEN.

[0265] For example, barcoded XTENs were constructed by performing one or more mutations comprising deletion of a glutamic acid residue, insertion of a glutamic acid residue, substitution of a glutamic acid residue, or substitution for a glutamic acid residue, or any combination thereof to the general-purpose XTENs in Table 9.

Table 9. Four general-purpose XTENs used for engineering of barcoded XTEN

SEQ ID NO.	XTEN Name	Amino Acid Sequence
676	AE144	GSEPATSGSETPGTSESATPESGPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEG SAPGSEPATSGSETPGSEPATSGSETPGSEPATSGSETPGTSTEPSEGSAPGTSESA TPESGPGSEPATSGSETPGTSTEPSEGSAP
677	AE288_1	GTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPE SGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESA TPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTS ESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEE GTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEG SAP
678	AE576	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEG SAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGS PTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTS TEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPE SGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEP SEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTS TEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPE SGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEP SEGSAP
679	AE864	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEG SAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGS PTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTS TEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGP GSEPATSGSETPGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSESATPE SGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEP

SEQ ID NO.	XTEN Name	Amino Acid Sequence
		SEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTS TEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPE SGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEP SEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTS ESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETP GTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPE SGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGS PTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGSEPATSGSETPGTSESATPESGPGTS TEPSEGSAP

Example 2. Sequence analysis of barcoded XTEN and selection thereof for fusion to a biologically-active polypeptide ("BP")

[0266] This example illustrates the design and selection of barcoded XTEN (and the assembly of more than one barcoded XTENs into a set) for fusion to a biologically-active polypeptide. Depending on the location of the barcode fragment(s) within the XTEN(s) and the manner in which the XTEN(s) is/are fused to a biologically-active protein to form an XTEN-containing construct (e.g., an XTENylated protease-activated T-cell engager (XPAT)), the barcode fragment(s) can indicate truncation(s) of the XTEN.

[0267] *In silico* GluC digestion analysis was performed on two exemplary XTENs (XTEN864 and XTEN288_1) to identify the releasable peptide fragments upon complete GluC digestion of the XTEN. The *in silico* analysis will take into consideration that, with respect to the XTEN having consecutive glutamic acid residues (e.g., "EE"), GluC may cleave after either one of the glutamic acid residues. As shown in the results summarized below in **Table 10**, a 10-mer peptide sequence "TPGTSTEPSE" (SEQ ID NO: 96) and a 14-mer peptide sequence "GSAPGSEPATSGSE" (SEQ ID NO: 97) each occur once and only once in the longer XTEN864, while all other peptide sequences occur two or more times in XTEN864. And the 14-mer peptide sequence "GSAPGSEPATSGSE" (SEQ ID NO: 97) also occurs once and only once in the shorter XTEN288_1.

[0268] The uniqueness of a candidate barcode is assessed in relation to all other peptide fragments releasable from the XTEN-containing construct. Accordingly, a barcode sequence in one XTEN cannot occur anywhere else in the XTEN-containing construct, including any other XTEN contained therewithin, any biologically-active protein contained therewithin, or any connection between neighboring components thereof. For example, **Table 11** shows a peptide "uniqueness" table for the set of two XTEN. Due to its presence in both XTEN864 and XTEN288, the 14-mer peptide sequence "GSAPGSEPATSGSE" (SEQ ID NO: 97) is not unique to the set of XTEN comprising both XTEN864 and XTEN288 and, thus, may not be used as a barcode for detecting truncations in polypeptide products that contain both of the two XTEN sequences.

[0269] The selection of a barcode (or a set of barcodes) may further involve identifying and determining the proper location(s) or position(s) of the candidate barcode(s) within the XTEN. The location or position of a

candidate barcode can be associated with pharmacologically relevant information of the XTEN (and the XTEN-containing construct as a whole), such as truncation of the XTEN beyond a critical length and/or deletion(s) in the XTEN sequence. The 10mer peptide "TPGTSTEPSE" (SEQ ID NO: 96) could serve as a suitable barcode fragment if XTEN864 is placed at the N-terminus of the XTEN-containing product and if the truncation of 238 amino acids from the N-terminus of the product does not significantly impact the pharmacological properties of the product.

Table 10. Representative XTEN sequences for *in silico* GluC digestion analysis

Exemplary XTEN	Amino acid sequence
XTEN864	GSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTE PSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPES GPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTS ESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSE**TPGTSTEPSE**GSAPGTSTEPS EGSAPGTSESATPESGPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETP GTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTE PSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSE TPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSP AGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATP ESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPG SPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGS PTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSET PGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSE**GSAPGSEPATSGSE**TPGTS ESATPESGPGTSTEPSEGSAP (SEQ ID NO: 98)
AE288_1	GTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTE PSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTST EEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSE PATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSE**GSAPGSEPATS GSE**TPGTSESATPESGPGTSTEPSEGSAP (SEQ ID NO: 99)

Table 11. Peptide "uniqueness" analysis

Peptide fragments	SEQ ID NO:	XTEN AE288_1	XTEN864	Both
SATPE	125	9	23	32
SGPGSEPATSGSE	126	4	9	13
GSAPGTSTEPSE	127	1	10	11
TPGTSE	128	4	7	11
SGPGTSTEPSE	129	2	8	10
GSAPGTSE	130	1	8	9
GTSTEPSE	131	2	5	7
GSAPGSPAGSPTSTEE	132	1	4	5
GSPAGSPTSTEE	133	1	4	5
SGPGTSE	134	2	3	5
SGPGSPAGSPTSTEE	135	1	3	4
TPGSEPATSGSE	136	1	2	3
TPGSPAGSPTSTEE	137	1	2	3

Peptide fragments	SEQ ID NO:	XTEN AE288_1	XTEN864	Both
GSAPGSEPATSGSE	138	1	1	2
TPGTSTEPSE	139		1	1

All underlined sequences produce unique GluC peptides
Non-XTEN core underlined and italic
Barcode peptides are bold

[0270] Exemplary barcode peptide sequences are illustrated below in **Table 12**. These barcode sequences should be flanked according to the structural formula (I):

AAA–Glu–Barcode Peptide–BBB,

wherein "AAA" represents Gly, Ala, Ser, Thr or Pro and "BBB" represent Gly, Ala, Ser, or Thr configured to facilitate efficient release of the barcode peptide by GluC digestion. Notably, the insertion of each barcode peptide in the XTEN may result in additional unique sequences directly preceding or following the inserted barcode peptides.

Table 12. List of non-limiting examples of barcode peptides

Candidate Barcode Peptide(s)	SEQ ID NO:
SPATSGSTPE	140
GSAPATSE	141
GSAPGTATE	142
GSAPGTE	143
PATSGPTE	144
SASPE	145
PATSGSTE	146
GSAPGTSAE	147
SATSGSE	148
SGPGSTPAE	149

Example 3: Design and selection of XTEN(s) in full sequence XTENylated polypeptide constructs

[0271] This example illustrates the design of a full-sequence polypeptide construct, containing two XTENs, one at the N-terminus and the other at the C-terminus.

[0272] **Table 13** below illustrates XTEN sequences used in a representative barcoded BPXTEN (containing barcoded XTENs at both the N- and C-termini) and a reference BPXTEN (containing general-purpose XTENs at both the N- and C-termini). In the representative barcoded BPXTEN, a barcoded XTEN (**SEQ ID No. 8014**) is fused at the N-terminus of the BP, and another barcoded XTEN (**SEQ ID No. 8015**) is fused at the C-terminus of the BP. In the reference BPXTEN, a "Ref-N" XTEN is fused at the N-terminus of the BP, and a "Ref-C" XTEN is fused at the C-terminus of the BP. The "Ref-N" XTEN is comparable in length to the barcoded XTEN **SEQ ID No. 8014**; and the "Ref-C" XTEN is comparable in length to the barcoded XTEN **SEQ ID No. 8015**. The barcoded and reference BPXTENs each contain a reference sequence in the BP component. The reference sequence is unique and differs in molecular weight from all other peptide fragments that are releasable from the corresponding BPXTEN upon complete digestion by GluC protease (e.g., according to **Example 5**). The

uniqueness of the reference sequence is assessed in relation to all other peptide fragments releasable from the BPXTEN construct.

Table 13. Representative sets of N- and C-terminal XTENs used in full-length BPXTEN constructs

*Barcode peptides are bold
All underlined sequences produce unique GluC peptides;

SEQ ID NO.	XTEN Type	Amino Acid Sequence	Total # of AAs
8014 (from **Table 3a**)	N-terminal XTEN	SPAGSPTSTESGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPES**SGPGSTPAESGSE**TPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSAP	292
8015 (from **Table 3a**)	C-terminal XTEN	PGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSE**GSAPGTE**STPSEGSAPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGEPEA	582
8021 Ref-N	N-terminal XTEN	SPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSTEPSEGSAPGTSESATPESGPGTSESATPESGPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGGSAP	292
8022 Ref-C	C-terminal XTEN	PGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGSEPATSGSETPGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGTSTEPSEGSAPGSPAGSPTSTEEGTSTEPSEGSAPGTSTEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSEPATSGSETPGTSEPSEGSAPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGTSESATPESGPGSEPATSGSETPGTSESATPESGPGSEPATSGSETPGTSESATPESGPGTSTEPSEGSAPGTSESATPESGPGSPAGSPTSTEEGSPAGSPTSTEEGTSESATPESGPGTSTEPSEGSAPGAAEPEA	584

Example 4: Recombinant construction and production of barcoded XTENylated fusion polypeptides

[0273] Example 4 illustrates recombinant construction, production, and purification of an XTENylated fusion polypeptide containing a barcoded XTEN at the C-terminus and another barcoded XTEN at the N-terminus using the methods disclosed herein.

[0274] EXPRESSION: A construct encoding an XTENylated fusion polypeptide that contains an anti-CD3 sequence (e.g., set forth in **Tables 6a-6e**), an anti-HER2 sequence (e.g., set forth in **Table 6f**), a barcoded XTEN (e.g., set forth in **Table 3a**) at the C-terminus, and a barcoded XTEN (e.g., set forth in **Table 3a**) at the N-terminus is expressed in a proprietary *E. coli* AmE098 strain and partitioned into the periplasm via an N-terminal secretory leader sequence (MKKNIAFLLASMFVFSIATNAYA) (SEQ ID NO: 100),, which is cleaved during translocation. Fermentation cultures are grown with animal-free complex medium at 37°C and temperature shifted to 26°C prior to phosphate depletion. During harvest, fermentation whole broth is centrifuged to pellet the cells. At harvest, the total volume and the wet cell weight (WCW; ratio of pellet to supernatant) are recorded, and the pelleted cells are collected and frozen at -80°C.

[0275] RECOVERY: The frozen cell pellet is resuspended in Lysis Buffer (100 mM citric acid) targeting 30% wet cell weight. The resuspension is allowed to equilibrate at pH 4.4 then homogenized at 17,000±200 bar while output temperature is monitored and maintained at 15±5°C. The pH of the homogenate is confirmed to be within the specified range (pH 4.4±0.1).

[0276] CLARIFICATION: To reduce endotoxin and host cell impurities, the homogenate is allowed to undergo low-temperature (10±5°C), acidic (pH 4.4±0.1) flocculation overnight (15-20 hours). To remove the insoluble fraction, the flocculated homogenate is centrifuged for 40 minutes at 8,000 RCF and 2-8°C, and the supernatant is retained. To remove nucleic acid, lipids, and endotoxin and to act as a filter aid, the supernatant is adjusted to 0.1% (m/m) diatomaceous earth. To keep the filter aid suspended, the supernatant is mixed via impeller and allowed to equilibrate for 30 minutes. A filter train, consisting of a depth filter followed by a 0.22 µm filter, is assembled then flushed with MQ. The supernatant is pumped through the filter train while modulating flow to maintain a pressure drop of 25±5 psig.

[0277] PURIFICATION

[0278] Protein-L Capture: To remove host cell proteins, endotoxin, and nucleic acid, Protein-L is used to capture the kappa domain present within the aHER2 scFv of the BPXTEN molecule. The Protein-L stationary phase (Tosoh TP AF-rProtein L-650F), Protein-L mobile phase A (11.5 mM citric acid, 24.5 mM Na_2HPO_4, 125 mM NaCl, 0.005% polysorbate 80, pH 5.0), and Protein-L mobile phase B (11 mM phosphoric acid, 0.005% polysorbate 80, pH 2.0) are used herein. The column is equilibrated with Protein-L mobile phase A. The filtrate is adjusted to pH 5.5±0.2 and loaded onto the Protein-L column targeting 2-4 g/L-resin then chased with Protein-L mobile phase A until absorbance at 280 nm (A280) returns to (local) baseline. Bound material is eluted with mobile phase B and collected as a 2 CV fraction pre-spiked with 0.4 CV of 0.5 M Na_2HPO_4 and is analyzed by SDS-PAGE.

[0279] C-tag Intermediate Purification: To ensure C-terminal integrity, C-tag Affinity Chromatography is used to capture the C-terminal-EPEA tag. The C-tag stationary phase (Thermo C-tagXL), C-tag mobile phase A (50

mM histidine, 200 mM NaCl, 0.005% polysorbate 80, pH 6.5), and C-tag mobile phase B (20 mM Tris, 0.6 M $MgCl_2$, 0.005% polysorbate 80, pH 7.0) are used herein. The column is equilibrated with C-tag mobile phase A. The IMAC Elution is loaded onto the C-tag column targeting 2 g/L-resin and chased with C-tag mobile phase A until absorbance at 280 nm (A280) returned to (local) baseline. Bound material is eluted with a C-tag mobile phase B. The C-tag Elution is collected as a 2 CV fraction and is analyzed by SDS-PAGE.

[0280] AEX Polishing: To separate dimer and aggregate from monomeric product Anion Exchange (AEX) chromatography is utilized to capture the electronegative N- and C-terminal XTEN domains. The AEX1 stationary phase (BIA QA-80), AEX1 mobile phase A (50 mM histidine, 200 mM NaCl, 0.005% polysorbate 80, pH 6.5), and AEX1 mobile phase B (50 mM histidine, 500 mM NaCl, 0.005% polysorbate 80, pH 6.5) are used herein. The column is equilibrated with AEX mobile phase A. The C-tag elution is diluted to 10 mS/cm with MQ, loaded targeting 2 g/L-resin, and then chased with AEX mobile phase A until absorbance at 280 nm returned to (local) baseline. Bound material is eluted with a gradient from 0% B to 100% B over 60 CV. Fractions are collected in 1 CV aliquots while A280 $\geq$ 2 mAU above (local) baseline. Elution fractions are analyzed by SDS-PAGE and SE-HPLC, and fractions found to be $\geq$ 98% monomer are pooled (AEX Pool) for further processing.

[0281] FORMULATION: To exchange the product into formulation buffer and to bring the product to the target concentration (0.5 g/L), Ultrafiltration/Diafiltration (UF/DF) is used. Using a 10 kDa membrane with an area of 0.1 m^2 and a TMP target of 15 psi, the AEX pool is concentrated to 0.5 g/L, then diluted 10-fold with Formulation Buffer (50 mM histidine, 200 mM NaCl, 0.005% polysorbate 80, pH 6.5). The AEX pool is concentrated 10-fold and diluted 10-fold two more times. The recovered Formulated product is 0.22 µm filtered within a BSC, aliquoted, labeled, and stored at -80°C as Bulk Drug Substance (BDS). The BDS is confirmed by various analytical methods to meet all Lot Release criteria. Overall quality is analyzed by SDS-PAGE, the ratio of monomer to dimer and aggregate is analyzed by SE-HPLC, and N-terminal quality and product homogeneity is analyzed by HI-HPLC. Identity is confirmed by ESI-MS.

Example 5. Release of barcode peptides by protease digest

[0282] This example illustrates the release of barcode fragment(s) and reference fragment(s) from a polypeptide mixture that contain varying lengths or truncated forms of the XTEN-containing construct using the methods disclosed herein.

[0283] A sample of XTEN-containing construct is reduced and alkylated via incubation in DTT and then iodoacetamide, sequentially. The samples are then buffer exchanged and desalted using a size-exclusion spin cartridge. Glu-C protease is added to the samples at an enzyme to substrate ratio of 1:5 and the samples are incubated at 37°C for digestion. Samples are then moved to 4°C to halt the proteolytic reaction and placed in autosampler vials for analysis.

Example 6. Detection and quantification of barcode peptide(s) and reference peptide(s)

[0284] This example illustrates mass spectrometry methods used to generate quantitative measurements of individual barcode peptides. An LC- Parallel Reaction Monitoring (PRM) method is programmed into a high-resolution accurate mass (HRAM) mass spectrometer. Unlike traditional Data-Dependent Acquisition (DDA) mass spectrometry methods, PRM methods focus on a specific set of 15-30 peptides in one run, sequencing each by MS-MS once per duty cycle. As such, this method generates eXtracted Ion Chromatograms (XICs) for the unfragmented precursor ions of the intact peptide, as well as for each fragment ion of the peptide to confirm its sequence. Fragment ion XICs are often more sensitive and selectively quantitative than the precursor ion fragments. The LC-PRM method used includes the light and heavy versions of seven barcode peptides. Chromatographic peak areas of all fragment ions of these 14 peptides are measured post-acquisition and the strongest fragment ion is used for quantitative measurement. Peak area ratios of the XTEN barcode peptides to the PAT barcode peptides are then calculated for relative XTEN:PAT abundance at various points across the XTEN molecules.

Example 7. Stable isotope labeling to quantify the peptides by mass spectrometry (MS)

[0285] This example illustrates the stable-isotope labeling schema to enable absolute (rather than relative) quantitation of barcode peptides from XTEN-containing polypeptides. A standard Heavy labeled Amino acid quantitative schema will be employed wherein synthetic analogues of barcode peptides in which the C-terminal Glutamic Acid is replaced with the $(^{13}C)_5H_7(^{15}N)O_3$ heavy labeled analogue are procured from a specialized vendor. A calibration curve will be prepared where a known amount of XTEN barcode containing polypeptide is serially diluted into a matrix where the heavy-labeled synthetic peptide is held at a constant concentration. Accurate quantitation can be performed by calibrating chromatographic peak area heavy:light ratios from the curve against research samples containing the same spike-level of heavy labeled peptide.

Example 8. Quantification of truncation of XTEN-containing polypeptide

[0286] This example illustrates the quantification of length variants or truncation variants in a mixture of XTEN-containing polypeptides.

[0287] For example, a barcode peptide "SGPGSTPAESGSE" (SEQ ID NO: 150), is positioned 76 amino acids into the representative barcoded BPXTEN sequence described in and obtained from **Example 3** to indicate a severe truncation of the XTEN at the N-terminal end of the BPXTEN. Also consider a potential barcode fragment "SPAGSPTSTESGTSE" (SEQ ID NO: 151), is positioned at the N-terminus. The abundance measurement ratio of each barcode peptide relative to a unique reference peptide sequence from the biologically active protein (e.g., an scFv fragment) sequence following the procedure of **Example 6** indicates the total amount of the full-length polypeptides and the variants having truncations that could affect pharmacological efficacy in the sample mixture. The abundance measurement of at least one reference fragment is used to

indicate the total amount of all variants of the polypeptide in the sample mixture. Accordingly, differential abundance between the reference fragment and the barcode fragments informs the amount of truncated polypeptide variants. The LC-MS data are analyzed to determine the ratio of the amount of the barcode fragment to the reference fragment, indicating the relative amount of pharmacologically-efficacious variants in the polypeptide mixture.

[0288] A set of two (or three) barcodes are used to indicate different levels of truncation of the polypeptide. The LC-MS data are used to determine the ratio of the amount of each barcode fragment to the amount of the reference fragment, thereby quantifying the distribution of truncation variants in the polypeptide mixture.

Example 9. *In vitro* cytotoxicity of XTENylated (masked) and de-XTENylated (unmasked, activated) XPATs (Protease-Activated T cell engager) against target cells

[0289] This example illustrates masking by XTENs on XTENylated Protease-Activated T-cell engagers ("XPATs") in general, and on HER2-XPATs in particular. This example illustrates the differential in cytotoxicity of XTENylated (masked) HER2-XPATs (e.g., as set forth in Table D) and the corresponding de-XTENylated (unmasked) HER2-PATs.

[0290] The cytotoxicity of an XTENylated PAT (as set forth in Table D) and the corresponding de-XTENylated PAT (protease-treated) was determined using an *in vitro* cytotoxicity assay which utilized the amount of ATP present in wells of lysed target cells post treatment as a proxy for measuring cell viability. HER2-expressing target cells were seeded on white, opaque bottom plates at varying densities (BT474: 20k cells/well, and SKOV3: 10k cells/well) and allowed to incubate at 37°C, 5% CO_2 overnight (18-24 hours). Prior to the end of the overnight incubation, peripheral blood mononuclear cells (PBMCs) were thawed and incubated at 37°C, 5% CO_2 overnight. The PBMCs were isolated from screened, healthy donors by Ficoll density gradient centrifugation from either whole blood or from lymphocyte-enriched buffy coat preparations obtained from BioIVT. 10X XTENylated and de-XTENylated PAT titrations were prepared using a 9-point, 3-fold titration (10th point is non-treatment) with a starting concentration of 2400 nM for the XTENylated PAT and 10 nM for the de-XTENylated PAT. The PBMCs were seeded in the wells at 1:1 Effector:Target ratio. 10X XTENylated and de-XTENylated PAT titrations were diluted 10-fold into the wells for starting concentrations of 240 nM and 1 nM, respectively. The plates were incubated at 37°C, 5% CO_2 for 48 hours. After the 48-hour incubation, the plates were washed 3X with 1X PBS, and 100 µL of 1X PBS was added to all wells. 100 µL of CellTiter-Glo® luminescent substrate solution was added to all wells, and the plates were allowed to incubate at room temperature for 1-5 minute(s). The plates were then shaken on a plate shaker at 300-500 rpm for 30-60 seconds to mix the contents of the wells and then read in a luminometer using an integration time of 100 ms. The intensity of signal produced correlates to the amount of viable cells present in the wells. The mean of the signal from all non-treatment wells was calculated and used to determine % Live cells from

treatment wells ((Treatment Signal/Mean of Non-Treatment Signal)*100 = % Live). The % Live was plotted by concentration, and half maximal response (EC50) values were derived with a 4-parameter logistic regression equation using GraphPad Prism software.

[0291] The XTENylated PAT and the corresponding de-XTENylated PAT displayed large differences in cytotoxicity on all HER2-expressing cell lines tested, confirming XTENylation results in reduction of cytolytic activity of the masked bispecific antibody (inactivated state). As shown in **Figures 5A-5B**, cytotoxicity of de-XTENylated (unmasked, activated) PAT on BT-474 and SK-OV-3 cells was observed in a dose-dependent manner, with maximal killing of ~80% observed at 0.3 nM. As shown in **Figures 5A-5B**, the de-XTENylated PAT exhibited EC50 values of 4.8 pM (BT474) and 3.4 pM (SKOV3), while the XTENylated PAT exhibited EC50 values of 49,370 pM (BT474) and 44,474 pM (SKOV3). The observations corroborate that de-XTENylated HER2-PAT has cytotoxic activity on cell lines with HER2 expression, and XTENylation masks (or shields) the ability to form an immune synapse, resulting in a reduction of cytotoxicity.

Example 10. *In vitro* cytotoxicity of XTENylated (masked) and de-XTENylated (unmasked, activated) XPATs against target cells that express low levels of target antigen

[0292] Example 10a:

[0293] Normal human cardiomyocytes express low levels of HER2 and as a result, rare cases of cardiac toxicity have been observed in patients treated with some HER2-targeted therapies. Given the known potential cardiotoxicity of HER2-targeted agents, AMX-818 toxicity against human cardiomyocytes which express HER2 was assessed. Consistent with the lack of cardiotoxicity in non-human primate (NHP) preclinical studies, AMX-818 showed limited cytotoxicity against human cardiomyocytes *in vitro*, even at micromolar doses. In contrast, the unmasked TCE counterpart demonstrated significantly greater cytotoxicity activity (EC50 ~100pM)

[0294] This example illustrates the sensitivity of primary cardiomyocytes to T cell-directed cytotoxicity in response to increasing concentrations of a HER2-XPAT (e.g., set forth in Table D) and its active proteolytic product, the corresponding HER2-PAT.

[0295] Normal human cardiomyocytes purchased from FujiFilm Cellular Dynamics were used. Cardiomyocytes (iCell cardiomyocytes, Cellular Dynamics International) were revived from liquid nitrogen and plated at 20,000 cells per 96-well for 7 days and treated as per the manufacturer's instructions. Human peripheral blood lymphocytes were added onto the iCell cardiomyocytes at a 1:1 Effector:Target ratio with increasing 3-fold concentrations of the XTENylated HER2-XPAT or the corresponding de-XTENylated HER2-PAT and incubated for 48 hours at 37°C, 5% CO_2. The assay was performed in RPMI (Roswell Park Memorial Institute) medium and 10% heat-inactivated fetal bovine serum. Cardiomyocyte cell viability was determined via ATP quantification and was performed with the Cell Titer-Glo Luminescent Cell Viability Assay System (Promega). Cell supernatant was aspirated, and cells were washed twice with phosphate buffered saline (PBS),

aspirated and followed by addition of PBS (100µl per well). Automated plate washing was carried out using an LS405 microplate washer dispenser (BioTek). Cell Titer-Glo reagent was added (100µl per well), and assay plates were incubated for 5 minutes at room temperature. Luminescence was quantified with a multi label reader (Molecular Devices) with a luminescence detector. For analysis of cytotoxicity, % viable cells was calculated from relative luminescence units (RLU). %live= (Test well RLU/Target cell only RLU)*100. For EC_{50} determination, data were transformed in Microsoft Excel and analyzed with Graph Pad Prism 8.3.1 software 'log(agonist) vs.response-variable slope (four parameters).

[0296] As shown in **Figure 6A**, the cardiomyocytes were killed by T cell-directed cytolysis in response to the de-XTENylated HER2-PAT at approximate EC50 concentration of 64 pM, while the cardiomyocytes remained refractory to killing by XTENylated HER2-XPAT at concentrations as high as 1 µM, illustrating that the XTENylated PAT has less activity in the cardiomyocytes as compared to the cancer cell lines.

[0297] **Example 10b:** This example illustrates the sensitivity of another cell line with relatively low level of HER2 expression, MCF-7, to T cell-directed cytotoxicity in response to increasing concentrations of a HER2-XPAT (e.g., set forth in Table D) and its active proteolytic product, the corresponding HER2-PAT.

[0298] MCF-7 cells were cultured according to established protocols. The concentration-response curves and EC50 values were measured and analyzed according to the process outlined in Example 10a.

[0299] As shown in **Figure 6B**, the MCF-7 cells were effectively killed by T cell-directed cytolysis in response to the proteolytically de-XTENylated HER2-XPAT (solid circles) at an EC50 concentration between 0.01 nM and 0.1 nM. XTENylation on HER2-XPAT (e.g., set forth in Table D) reduced T cell-mediated cytotoxicity by at least 10^4-fold, as indicated by a right-ward shift of the dose-response curve (solid squares).

[0300] Example 10c:

[0301] This example illustrates sensitivity of another cell line that expresses a medium level of HER2, MDA-MB-453, to T cell-directed cytotoxicity in response to increasing concentrations of a HER2-XPAT (e.g., set forth in Table D) and its active proteolytic product, the corresponding HER2-PAT. MDA-MB-453 is a breast cancer cell line with medium HER2 expression.

[0302] The cytotoxicity of an XTENylated PAT (as set forth in Table D) and the corresponding de-XTENylated PAT (protease-treated) was determined using an *in vitro* cytotoxicity assay which utilized the amount of ATP present in wells of lysed target cells post treatment as a proxy for measuring cell viability. HER2-expressing target cells (MDA-MB-453 line) were seeded on white, opaque bottom plates at 10k cells/well and allowed to incubate at 37°C, 5% CO_2 overnight (18-24 hours). Prior to the end of the overnight incubation, peripheral blood mononuclear cells (PBMCs) were thawed and incubated at 37°C, 5% CO_2 for 3-4 hours. The PBMCs were isolated from screened, healthy donors by Ficoll density gradient centrifugation from either whole blood or from lymphocyte-enriched buffy coat preparations obtained from

BioIVT. 10X XTENylated and de-XTENylated PAT titrations were prepared using a 7-point, 5-fold titration with a starting concentration of 3000 nM for the XTENylated PAT and 10 nM for the de-XTENylated PAT. The PBMCs were seeded in the wells at 10:1 Effector: Target ratio. 10X XTENylated and de-XTENylated PAT titrations were diluted 10-fold into the wells for starting concentrations of 300 nM and 1 nM, respectively. The plates were incubated at 37°C, 5% CO_2 for 48 hours. After the 48-hour incubation, the plates were washed 3X with 1X PBS, and 100 µL of 1X PBS was added to all wells. 100 µL of CellTiter-Glo® luminescent substrate solution was added to all wells, and the plates were allowed to incubate at room temperature for 1-5 minute(s). The plates were then shaken on a plate shaker at 300-500 rpm for 30-60 seconds to mix the contents of the wells and then read in a luminometer using an integration time of 100 ms. The intensity of signal produced correlates to the amount of viable cells present in the wells. The mean of the signal from all non-treatment wells was calculated and used to determine % Live cells from treatment wells ((Treatment Signal/Mean of Non-Treatment Signal)*100 = % Live). The % Live was plotted by concentration, and half maximal response (EC50) values were derived with a 4-parameter logistic regression equation using GraphPad Prism software.

[0303] The concentration-response curves for the de-XTENylated HER2-XPAT (solid circles) and the XTENylated HER2-PAT (solid squares) are shown, respectively, in **Figure 6C**.

[0304] HER2 expression levels of the cell lines tested herein are known in the art. For example, Hendriks *et al.* (Mol. Cancer Ther. September 1, 2013;12(9):1816-1828), which is incorporated by reference herein in its entirety, details mean HER2 level of various cells in Figure 1E (X-axis indicates number of HER2 receptors per cell). BT-474 can have a relatively high HER2 level (an immunohistochemistry test score) of "3+"; cardiomyocytes can have a HER2 level of "2+"; and MCF-7 can have a relatively low HER2 level of "1+/0."

Example 11. Tumor regression induced in subjects by administration of XTENylated XPATs

[0305] This example illustrates tumor regression induced in subjects by administration of vehicle, non-cleavable XTENylated HER2-PAT, cleavable XTENylated HER2-PAT, and unmasked HER2-PAT. The non-cleavable constructs used in the experiments are identical to the corresponding cleavable constructs, but the release site has been replaced with a non-cleavable sequence of similar length made from GASTEP amino acids (glycine, alanine, serine, threonine, glutamate, and/or proline).

[0306] **Example 11a:** Mice were implanted with estrogen pellets (17β-estradiol, 60 day release) at the right flank one day before the tumor inoculation. BT-474 tumor cells were inoculated subcutaneously at the right flank region of each mouse at a concentration of 2×10^7 BT-474/200ul RPMI 1640 with Matrigel (1:1)/mouse for tumor development. The date of tumor cell inoculation was denoted as Day 0 (D0).

[0307] When tumors become palpable (mean tumor volume (MTV) = 117 mm^3) on D8, PBMCs were intravenous implanted at 1 x 10^7 PBMC/200ul RPMI 1640/mouse. One group of mice was not injected with

PBMCs (the "without hPBMCs" group) and underwent the remainder of experimental treatments in parallel with the PBMC-injected mice. When mean tumor volume reached 147 mm^3 (D10), the "without hPBMCs" group of mice was administered with vehicle (diluent) (solid square in **Figure 7A**); and the PBMC-injected mice were randomized into four groups and administered with vehicle (diluent) (solid circle in **Figure 7A**), unmasked bispecific (solid triangle in **Figure 7A**), cleavable XTENylated bispecific (hexagon in **Figure 7A**), and non-cleavable XTENylated bispecific (diamond in **Figure 7A**), respectively. All five groups received an equimolar dose at 15nmol/Kg.

[0308] As illustrated in **Figure 7A**, by the end of study, D35, the cleavable XTENylated HER2-PAT (e.g., set forth in Table D) and the unmasked bispecific both demonstrated significant anti-tumor efficacy when compared to controls (vehicle with or without hPBMC) and the non-cleavable XTENylated bispecific counterpart. The lack of any tumor growth inhibition of the non-cleavable XTENylated bispecific construct supports that the anti-tumor efficacy observed on the XTENylated HER2 bispecific is driven by proteolytic cleavage.

[0309] **Example 11b:** The experimental setup implemented was similar to that described in Example 11a. Mice were implanted with estrogen pellets (17β-estradiol, 60 day release) at the right flank one day before the tumor inoculation. BT-474 tumor cells were inoculated subcutaneously at the right flank region of each mouse at a concentration of 2×10^7 BT-474/200ul RPMI 1640 with Matrigel (1:1)/mouse for tumor development. The date of tumor cell inoculation was denoted as Day 0 (D0). When tumors become palpable (MTV = 94 mm^3) on D8, PBMCs were intravenous implanted at 1×10^7 PBMC/200ul RPMI 1640/mouse. One group of mice was not injected with PBMCs (the "without hPBMCs" group) and underwent the remainder of experimental treatments in parallel with the PBMC-injected mice. When MTV reached 444 mm^3 (D20), the PBMC injected mice were randomized, and vehicle (diluent) and test article treatments were delivered as a single dose for all groups on D21.

[0310] **Figure 7B** illustrates significant anti-tumor efficacy of cleavable XTENylated HER2-XPAT (e.g., set forth in Table D) even in large established tumors (e.g., mean tumor volume (MTV) > 400 mm^3) after a single dose, when compared to controls and the non-cleavable XTENylated bispecific counterpart. The lack of appreciable level of tumor growth inhibition achieved by the non-cleavable XTENylated HER2-PAT supports that the anti-tumor efficacy of the cleavable XTENylated HER2-PAT is driven by proteolytic cleavage.

[0311] **Example 11c:** Examples 11a and 11b used xenografts generated from a breast cancer cell line that expresses relatively high levels of HER2, in the present example, the tumor regression studies were performed using a colorectal xenograft model that is characterized by low levels of HER2 expression. Mice were inoculated subcutaneously at the right flank region of each mouse at a concentration of 5×10^6 HT-55/200ul

RPMI 1640 with Matrigel (1:1)/mouse for tumor development. The date of tumor cell inoculation was denoted as Day 0 (D0).

[0312] When tumors become palpable (mean tumor volume (MTV) = 82 mm^3) on D6, PBMCs were intravenously implanted at 1 x 10^7 PBMC/200ul RPMI 1640/mouse. One group of mice was not injected with PBMCs (the "without hPBMCs" group) and underwent the remainder of experimental treatments in parallel with the PBMC-injected mice. When mean tumor volume reached 130 mm^3 (D10), the "without hPBMCs" group of mice was administered with vehicle (diluent) (solid square in **Figure 7C**); and the PBMC-injected mice were randomized into five groups and administered with vehicle (diluent) (solid circle in **Figure 7C**), unmasked bispecific at 15nmol/Kg (solid triangle in **Figure 7C**), cleavable XTENylated bispecific at 15nmol/Kg (triangle in **Figure 7C**) cleavable XTENylated bispecific at 36nmol/Kg (hexagon in **Figure 7C**), and non-cleavable XTENylated bispecific. (diamond in **Figure 7A**)

[0313] As illustrated in **Figure 7C**, by the end of the study the cleavable XTENylated HER2-PAT (e.g., set forth in Table D) and the unmasked bispecific (at both 15nmol/Kg and 36nmol/Kg) demonstrated significant anti-tumor efficacy when compared to controls (vehicle with or without hPBMC) and the non-cleavable XTENylated bispecific counterpart. The lack of any tumor growth inhibition of the non-cleavable XTENylated bispecific construct supports that the anti-tumor efficacy observed on the XTENylated HER2 bispecific is driven by proteolytic cleavage.

[0314] In additional *in vivo* experiments, it was demonstrated that HER2-XPAT (i.e., XTENylated or masked HER2-PAT) is preferentially unmasked in tumor tissue with significantly greater cleavage seen in tumors as compared to normal tissue. BT-474 tumors were established in NSG mice. The mice were injected with HER2-XPAT that was site-specifically labelled with an IR dye. After a period of 2 days, the tissue (tumor, brain, heart and liver) was harvested, homogenized and the presence of unmasked HER2-PAT in the various tissues of the BT-474 was assessed. Further, in order to determine whether the cleavage/unmasking of the HER2-XPAT occurred in vivo in the tumor or whether it was an artefact of the homogenization of the tissue once the tissue had been harvested, the samples were spiked with a differently labeled HER2-XPAT prior to same preparation. **Figure 7D** demonstrates that there is preferential cleavage of HER2-XPAT to the unmasked PAT in the tumor tissue as compared to the cleavage seen in the brain, heart, and liver tissue. Further, **Figure 7D** also shows that the cleavage occurred in vivo in the tissue as opposed to ex vivo as a result of homogenization or other handling. In addition, there is strong evidence of XPAT cleavage in fresh tumor samples.

[0315] Examples 11a-11c demonstrate that cleavable XTENylated HER2-PAT induces robust tumor regression in tumor models that express low levels of HER2 as well as tumors that are characterized with high HER2 expression. As such tumor regression is not observed in the test subjects treated with non-cleavable XPAT. Moreover, the data presented herein show that HER2-XPAT is preferentially unmasked in vivo, in the

tumor microenvironment. The experimental design for Examples 11a-11c are further discussed below in Example 17.

Example 12. Lymphocyte margination induced in subjects by administration of XTENylated XPATs

[0316] This example illustrates lymphocyte margination induced in subjects (e.g., female and male monkeys) by administration of an XTENylated HER2-PAT (e.g., as set forth in Table 6), for example, a single-dose intravenous (IV) infusion (e.g., at 25 mg/kg at a dose volume of 10 ml/kg).

[0317] Test article dosing formulations were prepared by diluting the XPAT-of-interest according to established protocols to meet dose level requirements. The test article (the HER2-XPAT of interest) and the appropriate diluent were provided as single-use aliquots and stored in a freezer set to maintain -80°C until use. Once thawed, aliquots were stored at 2°C-8°C and were not refrozen. The dosing formulations were prepared on the day of dosing. On the day of use, an aliquot of XPAT stock solution and the appropriate diluent was thawed at room temperature and an appropriate amount of test article was diluted to obtain the proper concentration. The dosing formulation was mixed by inverting the tube gently several times. The IV route of exposure was selected as a candidate route of human exposure.

[0318] As shown in **Figure 8**, the HER2-XPAT-related changes in hematology parameters following a single dose (25 mg/kg), including a decrease of lymphocytes (summarized in the table immediately below), were observed starting at 6 hours post-dose (0.5-0.2x of the pre-dose level) and sustained at 24 hours post-dose (0.27-0.1x of the pre-dose level) and at 48h (0.09-0.36x of the pre-dose level). At 72 hr post-dose, the lymphocyte level remained low (0.3-0.1x of the pre-dose level).

Table 14. Hematology: lymphocyte changes associated with HER2-XPAT administration

Parameter	Time Point	Dose (25 mg/kg)	
		M	F
Lymphocyte count	Pre-dose	7.22	7.31
	6 hours post-dose	1.49	3.87
	24 hours post-dose	0.7	1.98
	48 hours post-dose	0.67	2.63
	72 hours post-dose	0.84	2.27

Example 13. Evaluation of toxicity and pharmacokinetics of XTENylated PATs (XPATs) in subjects

[0319] Toxicity and pharmacokinetics of XTENylated HER2-PAT (e.g., set forth in Table D) were assessed in cynomolgus monkeys. Animals were dosed with 25 mg/kg of a cleavable XTENylated HER2-PAT or a corresponding non-cleavable XTENylated counterpart over 1 hour. Blood for plasma was collected at pre-dose and post end of infusion at: 5 min, 2 hrs, 4 hrs, 6 hrs, 24 hrs, 48 hrs, 96 hrs, 144 hrs, 168 hrs, and 240 hrs. All blood was collected on time +/- 2 minutes. Plasma samples were analyzed for the concentration of the cleavable and non-cleavable XTENylated bispecific constructs. The plasma drug concentrations were measured by

ECLIA (electrochemiluminescent immunoassay) using recombinant HER2 as capture and an antibody directed against the XTEN mask for detection. As shown in **Figure 9**, in cynomolgus monkeys, the HER2-XPAT (e.g., set forth in Table D) showed a pharmacokinetic profile analogous to the corresponding non-cleavable version due to its stability in peripheral tissues. The non-cleavable constructs used in the experiments are identical to the corresponding cleavable constructs, but for the fact that the release site has been replaced with a non-cleavable sequence of similar length made from GASTEP amino acids (glycine, alanine, serine, threonine, glutamate, and/or proline). The presence of proteolytic metabolites in circulation after two separate doses was also monitored and shown in **Figure 9B** as a percentage of the total HER2-XPAT. As shown in **Figure 9B** the presence of the 1x-metabolites is less than 4% of the total HER2-XPAT.

Example 14. XTEN Masking Expands Safety Margin of HER2-XPAT vs. HER2-PAT

[0320] Additional data shows that HER2-XPAT is well-tolerated in cynomolgus monkeys and the presence of the provides a 450-fold higher tolerated Cmax as compared to that seen with unmasked PAT. HER2-XPAT was administered IV, single dose/animal (doses 2.5-42mpk) and weekly x2 at 50mpk. *At doses 21mpk and above, a variant of HER2-XPAT with a shorter C-terminal XTEN mask was used. HER2-PAT was administered by continuous infusion due to its short half-life. Plasma concentrations of HER2-XPAT were measured by ECLIA* using recombinant HER2 capture and an antibody directed against the XTEN mask for detection. The Cmax values for HER2 PAT were determined by ECLIA utilizing an a-idiotypic Ab directed against the a-CD3 scFv as capture and recombinant HER2 as detection. *ECLIA = Electrochemiluminescent Immunoassay. **Figure 10A** shows the maximum tolerated dose of 42 mg/kg, indeed no overt CRS was seen even at 50 mg/kg. Conversely, in **Figure 10B** the maximum tolerated dose of the unmasked HER2-PAT is 0.2mg/kg. **Figure 10C** shows the plasma concentrations of masked HER2-PAT (at a 450-fold higher tolerated Cmax) as compared to unmasked HER2-PAT.

[0321] In **Figure 13A-Figure 13B** there is additional evidence supporting the safety profile of a preferred HER2-XPAT of the present invention. HER2-XPAT or its non-cleavable counterpart, HER2-XPAT-NoClvSite were administered as a single IV dose of 25 or 42 mg/kg. Dose normalized plasma drug concentrations were measured by ECLIA* using recombinant HER2 as capture and an antibody directed against the XTEN mask for detection. *ECLIA = Electrochemiluminescent Immunoassay. The data in Figure 12A shows a comparable PK between HER2-XPAT and the non-cleavable HER2-PAT formats, demonstrating that the protease release site remains largely stable in circulation of cynomolgus monkeys even at high doses. In Figure 12B, concentrations of singly-cleaved HER2-XPAT(1X-C) and HER2-XPAT(1X-N) were measured in plasma from cynomolgus monkeys administered HER2-XPAT. Quantitative Western was performed using an antibody recognizing the anti-HER2 scFv with standards prepared with recombinant versions of the singly-cleaved

molecules. Results are expressed as a percentage of the total XTENylated species as measured by ECLIA as described above. Figure 12B shows that even at high dose of HER2 XPAT there is very limited systemic accumulation of metabolites lacking one or both XTEN masks.

Example 15. HER2-XPAT Does Not Induce Cytokine Release Syndrome

[0322] Consistent with the tolerability of HER2-XPAT, the present Example demonstrates that HER2-XPAT in pilot NHP toxicology studies led to minimal T cell activation in circulation in NHPs, while its unmasked counterpart led to significant T cell activation even at much lower doses. Consistent with this masking of T cell activation, cytokine release upon administration of HER2-XPAT was also robustly attenuated relative to what was observed with its unmasked counterpart. There was no overt Cytokine Release Syndrome (CRS) with HER2-XPAT, even at the MTD of 42 mg/kg, while its unmasked TCE counterpart caused death due to CRS at doses as low as 0.3 mg/kg. Overall, AMX-818 was well-tolerated in NHP studies at doses that would be significantly higher than anticipated clinically relevant doses, and there were no gross or microscopic findings in HER2-expressing tissues such as the heart, where other approved HER2-targeted agents have demonstrated clinical toxicity.

[0323] Thus, even at 50mg/kg, administration of HER2-XPAT did not induce cytokine release or systemic T cell activation. This supports that there is minimal CRS risk for XPATs vs standard TCEs. Only 1-3% of singly-cleaved XPAT metabolites were detected in plasma from NHP administered high doses of HER2-XPAT (25 & 42mg/kg). For this study, peripheral T cell activation (%$CD25^+$) was evaluated by flow cytometry 24 hours post-HER2-XPAT treatment. Figure 11A and 11B show $CD25^+CD4^+$ T cell (**Figure 11A**) and $CD25^+CD8^+$ (**Figure 11B**) T cell in the presence of HER2-XPAT vs. HER2-PAT.

[0324] Cytokine analysis was performed with a Luminex® suspension array system on plasma samples. Figures 11C-11E show the IL-6 (**Figure 11C**), TNF-a (**Figure 11D**) and IFN-g (**Figure 11E**) levels in plasma in response to HER2-XPAT vs HER2-PAT. Data presented are maximal values measured between 6-24 hours at each evaluated dose. In additional GLP toxicology studies (data not shown), NHPs were dosed weekly with AMX-818 at 3 dose levels (0.5mg/kg, 2 mg/kg and 6mg/kg) over a 31-day period. The results from these GLP toxicology studies are consistent with the prior pilot NHP studies and there were no AMX-818 related adverse findings based on standard toxicology endpoints, including histopathologic examination, and all clinical pathological findings were reversible.

[0325] Also consistent with the safety of AMX-818 in NHPs up to 42mg/kg, AMX-818 is largely stable in circulation where protease inhibitors are abundant, as evidenced by the limited systemic accumulation of active metabolites. Because healthy NHPs were dosed and subjects to be treated with AMX-818 also may have systemic inflammation, the stability of AMX-818 was analyzed ex vivo in plasma samples taken from patients

with cancer and inflammatory disease, as well as NHPs in which we induced systemic inflammation. As shown in **Figure 12**, similar AMX-818 stability was observed in plasma samples from healthy NHPs, inflamed NHPs, healthy humans, and human patients with cancer or inflammatory disease in a stability study in which we incubated AMX-818 with plasma samples for seven days at 37°C.

[0326] For this experiment, AMX-818 with a fluorescent label (DyL650) attached to the tandem scFv was incubated in the indicated plasma samples for seven days at 37°C. Samples were then run on a gel and metabolites similar in size to the unmasked, active form of AMX-818 were quantified using a LI-COR detector. Inflammatory disease human samples were derived from patients with rheumatoid arthritis, lupus, inflammatory bowel disease, and multiple sclerosis. Cancer human samples were derived from patients with lung, breast, and colon tumors. Sample sizes: Healthy NHP N = 4, Healthy Human N = 4, "Inflamed" NHP N = 6, "Inflamed" Human N = 27, Cancer Human N = 11.

[0327] Across all four plasma groups, the range of products similar in size to the active, unmasked HER2 TCE was only approximately two to four percent of the total AMX-818 species present. This small percentage reflects an overestimate of potential accumulation of unmasked TCE *in vivo*, as renal filtration will rapidly eliminate such products from circulation (half-life of the unmasked form in NHP is approximately seven hours). These data suggest limited risk of significant systemic accumulation of the active, unmasked form of AMX-818 in cancer patients.

Example 16. AMX-818 and 818-PAT Induce Surface Expression of PD-1 and CD69 on T cells in Response to SKOV3 Tumor Cells

[0328] Surface PD-1 expression was evaluated on CD4+ and CD8+ T cells by flow cytometry following a 48 hour co-incubation of PBMCs and SKOV3 cells at a 5:1 Effector:Target ratio with test articles at the indicated concentrations. On both CD4 and CD8 T cell subsets, 818-PAT and AMX-818 increased the frequency of T cells expressing surface PD-1, with the AMX-818 curve shifted approximately 2.5-logs relative to 818-PAT, demonstrating significant masking by the prodrug relative to its activated counterpart. Maximal activation of the T cells by AMX-818(PAT) was achieved at 625pM concentrations, while maximal activation by AMX-818 was achieved at 150-600nM concentrations.

[0329] Histogram plots comparing the surface levels of PD-1 on CD4 and CD8 T cells at the indicated drug concentrations are shown in **Figure 14A-14D**.

[0330] AMX-818(PAT) and prodrug AMX-818 induced CD69 and PD-1 expression on the surface both CD4+ and CD8+ T-cell subsets to comparable degrees. However, the dose–response curve for AMX-818 was shifted on average >400-fold higher relative to that of the activated AMX-818(PAT), demonstrating effective functional masking of AMX-818 (data shown for CD8 T cells in **Figure 14A** and **Figure 14C**). Maximal

upregulation of activation marker CD69 and inhibitory receptor PD-1 by AMX-818(PAT) was observed at 0.62 nM concentrations, while comparable induction by AMX-818 required 150–600 nM concentrations (**Figure 14B and 14D**).

[0331] AMX-818(PAT) also potently induced surface expression of PD-L1 on SKOV3 tumor cells with an EC_{50} value of 19.7 pM, while XTENylated AMX-818 was 650-fold less potent in inducing comparable frequency and levels of PD-L1 (**see Figure 15A-15C**). The induction of PD-1 on T cells and PD-L1 on target tumor cells by AMX-818 is expected to dampen its activity and provides rationale for combining a PD-1 blocking therapy to AMX-818 in future clinical investigations.

[0332] AMX-818(PAT) and AMX-818 induced PD-1 to comparable levels and maximal percentages within both human $CD4^+$ and $CD8^+$ T-cell subsets. However, AMX-818 was on average >400-fold less potent than AMX-818(PAT). AMX-818(PAT) potently induced surface expression of PD-L1 on HER2-high expressing SKOV3 tumor cells with an EC_{50} value of 19.7 pM, while AMX-818 was > 650-fold less potent for comparable induction. The induction of PD-1 on T cells and PD-L1 on target tumor cells by AMX-818 is expected to dampen its activity and provides the rationale for a combination of PD-1 blocking therapy with AMX-818.

Example 17. XPATs Are Unmasked To TCEs in Human Tumors Implanted in Living Mice, with Minimal Cleavage Observed in Healthy Tissues

[0333] The present example summarizes the potent in vivo efficacy of the HER2-XPATS of the present invention. To assess anti-tumor activity, immunocompromised mice were injected with tumor cells expressing the target of interest. When the tumors were sufficiently large and well-established (over $100mm^3$), human PBMCs were injected and intravenous dosing of the HER2-XPATs and a variety of control molecules was initiated. These studies showed that the HER2 targeting XPATs drove tumor regressions and a number of complete responses. An AMX-818 variant lacking the protease-cleavable linker had no activity, indicating that protease cleavage of the linker drove activity.

[0334] Moreover, the HER2-XPAT expand the safety margin *in vivo*. HER2 has relatively limited expression in healthy tissues. When NHPs were injected with the HER2-XPATs, as well as their unmasked TCE counterparts, and tolerability was assessed to determine the maximum tolerated dose of each molecule. XTEN masks on the XPATs significantly increased the tolerability of the TCEs, enabling an over 400-fold higher Cmax at MTD relative to what is observed with the unmasked HER2 TCE. To provide evidence that the protease-cleavable linker can be cleaved across a broad range of human tumors, XPAT cleavage was analyzed in fresh human tumor samples, as well as patient-derived and xenograft tumors. In fresh human tumors explants,

an XPAT induced protease-cleavable linker-dependent release of cytokines from tumor resident T cells, similar to the cytokine release induced by a positive control (anti-CD3 antibody) that directly activated T cells.

[0335] To quantify the degree of preferential unmasking of XPATs that occurs in tumors, XPAT cleavage was assessed in living mice. A fluorophore labeled XPAT was injected into mice, and two days later harvested tumors and healthy tissues to measure levels of XPAT and cleavage products. To specifically quantify cleavage occurring in the living mouse, and control for any cleavage occurring after tissues were harvested, we also analyzed cleavage of a control XPAT added during tissue processing. Across 31 mice with nine different tumor types, on average, 20% of the XPAT in tumors of living mice was cleaved to the unmasked TCE form, while minimal cleavage of XPATs was observed in healthy tissues. The demonstration that there was limited- to-no cleavage of the control XPAT confirms that cleavage of the injected XPAT occurred in the living mice, not during tissue processing. The study design and data are shown in **Figure 16A-C**.

[0336] While preferred embodiments of the present invention have been shown and described herein, it will be obvious to those skilled in the art that such embodiments are provided by way of example only. It is not intended that the invention be limited by the specific examples provided within the specification. While the invention has been described with reference to the aforementioned specification, the descriptions and illustrations of the embodiments herein are not meant to be construed in a limiting sense. Numerous variations, changes, and substitutions will now occur to those skilled in the art without departing from the invention. Furthermore, it shall be understood that all aspects of the invention are not limited to the specific depictions, configurations or relative proportions set forth herein which depend upon a variety of conditions and variables. It should be understood that various alternatives to the embodiments of the invention described herein may be employed in practicing the invention. It is therefore contemplated that the invention shall also cover any such alternatives, modifications, variations or equivalents. It is intended that the following claims define the scope of the invention and that methods and structures within the scope of these claims and their equivalents be covered thereby.

CLAIMS

WHAT IS CLAIMED IS:

1. A polypeptide having an N-terminal amino acid and a C-terminal amino acid, the polypeptide comprising:

(a) an extended recombinant polypeptide (**XTEN**), comprising:

a barcode fragment (**BAR**) releasable from said polypeptide upon digestion by a protease;

(b) a bispecific antibody construct (**BsAb**), comprising:

a first antigen binding fragment (**AF1**) that specifically binds to cluster of differentiation 3 T cell receptor (CD3), which AF1 comprises light chain complementarity-determining regions 1 (CDR-L1), 2 (CDR-L2), and 3 (CDR-L3) and heavy chain complementarity-determining regions 1 (CDR-H1), 2 (CDR-H2), and 3 (CDR-H3), wherein said CDR-H3 comprises an amino acid sequence of **SEQ ID NO:10**; and

a second antigen binding fragment (**AF2**) that specifically binds to human epidermal growth factor receptor 2 (HER2); and

(c) a release segment (**RS**) positioned between said XTEN and said bispecific antibody construct,

wherein said XTEN is characterized in that:

(i) it comprises at least 100, or at least 150 amino acids;

(ii) at least 90% of its amino acid residues are identified herein by glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P);

(iii) it comprises at least 4 different types of amino acids identified herein by G, A, S, T, E, or P; and

(iv) said XTEN is formed from a plurality of non-overlapping sequence motifs that are each from 9 to 14 amino acids in length,

wherein said plurality of non-overlapping sequence motifs comprise:

(1) a set of non-overlapping sequence motifs, wherein each non-overlapping sequence motif of said set of non-overlapping sequence motifs is repeated at least two times in said XTEN; and

(2) a non-overlapping sequence motif that occurs only once within said XTEN;

wherein said barcode fragment (**BAR**) includes at least part of said non-overlapping sequence motif that occurs only once within said XTEN;

wherein said barcode fragment (**BAR**) differs in sequence and molecular weight from all other peptides fragments that are releasable from said polypeptide upon complete digestion of said polypeptide by said protease; and

wherein said barcode fragment (**BAR**) does not include said N-terminal amino acid or said C-terminal amino acid of said polypeptide.

2. The polypeptide of claim 1, wherein said set of non-overlapping sequence motifs are each independently identified herein by **SEQ ID NOS: 179-200 and 1715-1722**.

3. The polypeptide of claim 1 or 2, wherein said set of non-overlapping sequence motifs are each independently identified herein by **SEQ ID NOS: 186-189**.

4. The polypeptide of claim 3, wherein said set of non-overlapping sequence motifs comprise at least two, at least three, or all four of the sequence motifs **SEQ ID NOS: 186-189**.

5. The polypeptide of any one of claims 1-4, wherein said XTEN comprises a length of from 100 to 3,000, from 150 to 3,000, from 100 to 1,000, or from 150 to 1,000 amino acid residues.

6. The polypeptide of any one of claims 1-5, wherein said XTEN comprises a length of at least 200, at least 250, at least 300, at least 350, at least 400, at least 450, or at least 500 amino acid residues.

7. The polypeptide of any one of claims 1-6, wherein at least 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% of the amino acid residues of said XTEN are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P).

8. The polypeptide of any one of claims 1-7, wherein said XTEN has at least 90%, at least 92%, at least 95%, at least 98%, at least 99% or 100% sequence identity to a sequence set forth in **Table 3a**.

9. The polypeptide of any one of claims 1-8, wherein said barcode fragment (**BAR**) does not include a glutamic acid that is immediately adjacent to another glutamic acid, if present, in said XTEN.

10. The polypeptide of any one of claims 1-9, wherein said barcode fragment (**BAR**) has a glutamic acid at its C-terminus.

11. The polypeptide of any one of claims 1-10, wherein said barcode fragment (**BAR**) has an N-terminal amino acid that is immediately preceded by a glutamic acid residue.

12. The polypeptide of any one of claims 1-11, wherein said barcode fragment (**BAR**) is positioned a distance from either said N-terminus of said polypeptide or said C-terminus of said polypeptide, wherein said distance is from 10 to 150 amino acids, or from 10 to 125 amino acids in length.

13. The polypeptide of any one of claims 1-12, wherein said barcode fragment (**BAR**) is characterized in that:

(i) it does not include a glutamic acid that is immediately adjacent to another glutamic acid, if present, in said XTEN;

(ii) it has a glutamic acid at its C-terminus;

(iii) it has an N-terminal amino acid that is immediately preceded by a glutamic acid residue; and

(iv) it is positioned a distance from either said N-terminus of said polypeptide or said C-terminus of said polypeptide, wherein said distance is from 10 to 150 amino acids, or from 10 to 125 amino acids in length.

14. The polypeptide of any one of claims 11-13, wherein said glutamic acid residue that precedes said N-terminal amino acid of said barcode fragment (**BAR**) is not immediately adjacent to another glutamic acid residue.

15. The polypeptide of any one of claims 1-14, wherein said barcode fragment (**BAR**) does not include a second glutamic acid residue at a position other than the C-terminus of said barcode fragment unless said second glutamic acid is immediately followed by a proline.

16. The polypeptide of any one of claims 1-15, wherein said XTEN is positioned N-terminal of said bispecific antibody construct (**BsAb**), and wherein said barcode fragment (**BAR**) is positioned within 200, within 150, within 100, or within 50 amino acids of said N-terminus of said polypeptide.

17. The polypeptide of any one of claims 1-16, wherein said XTEN is positioned N-terminal of said bispecific antibody construct (**BsAb**), and wherein said barcode fragment (**BAR1**) is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from said N-terminus of said protein.

18. The polypeptide of any one of claims 1-15, wherein said XTEN is positioned C-terminal of said bispecific antibody construct (**BsAb**), and wherein said barcode fragment (**BAR**) is positioned within 200, within 150, within 100, or within 50 amino acids of said C-terminus of said polypeptide.

19. The polypeptide of any one of claims 1-15 and 18, wherein said XTEN is positioned C-terminal of said bispecific antibody construct (**BsAb**), and said barcode fragment (**BAR**) is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from said C-terminus of said protein.

20. The polypeptide of any one of claims 1-19, wherein said barcode fragment (**BAR**) is at least 4 amino acids in length.

21. The polypeptide of claim 20, wherein said barcode fragment (**BAR**) is between 4 and 20, between 5 and 15, between 6 and 12, or between 7 and 10 amino acids in length.

22. The polypeptide of any one of claims 1-21, wherein said barcode fragment (**BAR**) comprises an amino acid sequence set forth in **Table 2**.

23. The polypeptide of any one of claims 1-22, wherein said XTEN has a length defined by a proximal end and a distal end, wherein (1) said proximal end is positioned, relative to said distal end, closer to said bispecific antibody construct (**BsAb**), and wherein (2) said barcode fragment (**BAR**) is positioned within

a region of said XTEN that extends, as measured from said distal end, between 5% and 50%, between 7% and 40%, or between 10% and 30% of said length of said XTEN.

24. The polypeptide of any one of claims 1-23, wherein said XTEN further comprises additional one or more barcode fragments, wherein said additional one or more barcode fragments each differ in sequence and molecular weight from all other peptides fragments that are releasable from said polypeptide upon complete digestion of said polypeptide by said protease.

25. The polypeptide of any one of claims 1-24, wherein said release segment (**RS**) comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence identified herein by **SEQ ID NOS: 7001-7626**.

26. The polypeptide of any one of claims 1-25, wherein said protease cleaves on the C-terminal side of glutamic acid residues that are not followed by proline.

27. The polypeptide of claim 26, wherein said protease is a Glu-C protease.

28. The polypeptide of any one of claims 1-27 expressed as a fusion protein, wherein said fusion protein, in an uncleaved state, has a structural arrangement from N-terminus to C-terminus identified herein by AF1-AF2-RS-XTEN, AF2-AF1-RS-XTEN, XTEN-RS-AF1-AF2, or XTEN-RS-AF2-AF1.

29. The polypeptide of claim 1-28, wherein said release segment (**RS**) is fused to said bispecific antibody construct (**BsAb**) by a spacer.

30. The polypeptide of claim 29, wherein said spacer comprises at least 4 types of amino acids are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P).

31. The polypeptide of claim 29, wherein said spacer comprises an amino acid sequence having at least 80%, 90%, or 100% sequence identity to a sequence set forth in **Table C**.

32. The polypeptide of any one of claims 1-31, wherein said CDR-H1 and said CDR-H2 of said first antigen binding fragment (**AF1**) comprise amino acid sequences of **SEQ ID NOS: 8 and 9**, respectively.

33. The polypeptide of any one of claims 1-32, wherein:
said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:1 or 2**;
said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:4 or 5**; and
said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**.

34. The polypeptide of any one of claims 1-32, wherein:
said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:1**;
said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:4 or 5**; and
said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**.

35. The polypeptide of any one of claims 1-32, wherein:
said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:2**;

said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:4 or 5**; and
said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**.

36. The polypeptide of any one of claims 1-32, wherein:
said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:1**;
said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:4**; and
said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**.

37. The polypeptide of any one of claims 1-32, wherein:
said CDR-L1 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:2**;
said CDR-L2 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:5**; and
said CDR-L3 of said **AF1** comprises an amino acid sequence of **SEQ ID NO:6**.

38. The polypeptide of any one of claims 1-37, wherein said first antigen binding fragment (**AF1**) comprises four chain variable domain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3), and 4 (FR-H4), each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to an amino acid sequence of **SEQ ID NOS: 60, 64, 65, and 67**, respectively.

39. The polypeptide of any one of claims 1-37, wherein said first antigen binding fragment (**AF1**) comprises four chain variable domain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3), and 4 (FR-H4), each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to an amino acid sequence of **SEQ ID NOS: 61, 64, 65, and 67**, respectively.

40. The polypeptide of any one of claims 1-39, wherein said first antigen binding fragment further comprises four light chain variable domain framework regions (FR-L): FR-L1, FR-L2, FR-L3, and FR-L4, each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to amino acid sequences of **SEQ ID NOS: 51, 52, 53, and 59**, respectively.

41. The polypeptide of any one of claims 1-39, wherein said first antigen binding fragment further comprises four light chain variable domain framework regions (FR-L): FR-L1, FR-L2, FR-L3, and FR-L4, each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to amino acid sequences of **SEQ ID NOS: 51, 52, 54, and 59**, respectively.

42. The polypeptide of any one of claims 1-39, wherein said first antigen binding fragment further comprises four light chain variable domain framework regions (FR-L): FR-L1, FR-L2, FR-L3, and FR-L4, each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or

99% sequence identity or being identical to amino acid sequences of **SEQ ID NOS: 51, 52, 55, and 59**, respectively.

43. The polypeptide of any one of claims 1-39, wherein said first antigen binding fragment further comprises four light chain variable domain framework regions (FR-L): FR-L1, FR-L2, FR-L3, and FR-L4, each exhibiting at least 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity or being identical to amino acid sequences of **SEQ ID NOS: 51, 52, 56, and 59**, respectively.

44. The polypeptide of any one of claims 1-39, wherein said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein:

said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**;

said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**;

said FR-L3 comprises an amino acid sequence of **SEQ ID NO:53, 54, 55, or 56**;

said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**;

said FR-H1 comprises an amino acid sequence of **SEQ ID NO:60 or 61**;

said FR-H2 comprises an amino acid sequence of **SEQ ID NO:64**;

said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and

said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

45. The polypeptide of any one of claims 1-39, wherein said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein:

said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**;

said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**;

said FR-L3 comprises an amino acid sequence of **SEQ ID NO:53**;

said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**;

said FR-H1 comprises an amino acid sequence of **SEQ ID NO:60**;

said FR-H2 comprises an amino acid sequence of **SEQ ID NO:64**;

said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and

said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

46. The polypeptide of any one of claims 1-39, wherein said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein:

said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**;

said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**;

said FR-L3 comprises an amino acid sequence of **SEQ ID NO:54**;

said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**;

said FR-H1 comprises an amino acid sequence of **SEQ ID NO:61**;

said FR-H2 comprises an amino acid sequence of **SEQ ID NO:64**;

said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and

said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

47. The polypeptide of any one of claims 1-39, wherein said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein:

said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**;

said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**;

said FR-L3 comprises an amino acid sequence of **SEQ ID NO:55**;

said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**;

said FR-H1 comprises an amino acid sequence of **SEQ ID NO:61**;

said FR-H2 comprises an amino acid sequence of **SEQ ID NO:64**;

said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and

said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

48. The polypeptide of any one of claims 1-39, wherein said first antigen binding fragment (**AF1**) further comprises light chain framework regions 1 (FR-L1), 2 (FR-L2), 3 (FR-L3), and 4 (FR-L4) and heavy chain framework regions 1 (FR-H1), 2 (FR-H2), 3 (FR-H3) and 4 (FR-H4), wherein:

said FR-L1 comprises an amino acid sequence of **SEQ ID NO:51**;

said FR-L2 comprises an amino acid sequence of **SEQ ID NO:52**;

said FR-L3 comprises an amino acid sequence of **SEQ ID NO:56**;

said FR-L4 comprises an amino acid sequence of **SEQ ID NO:59**;

said FR-H1 comprises an amino acid sequence of **SEQ ID NO:61**;

said FR-H2 comprises an amino acid sequence of **SEQ ID NO:64**;

said FR-H3 comprises an amino acid sequence of **SEQ ID NO:65**; and

said FR-H4 comprises an amino acid sequence of **SEQ ID NO:67**.

49. The polypeptide of any one of claims 1-48, wherein said first antigen binding fragment (**AF1**) exhibits a higher thermal stability than an anti-CD3 binding fragment consisting of a sequence set forth in **SEQ ID NO: 206**, as evidenced in an *in vitro* assay by:

a higher melting temperature (T_m) of said first antigen binding fragment relative to that of said anti-CD3 binding fragment; or

upon incorporating said first antigen binding fragment into a test bispecific antigen binding construct, a higher T_m of said test bispecific antigen binding construct relative to that of a control bispecific antigen binding construct, wherein said test bispecific antigen binding construct comprises said first antigen binding fragment and a reference antigen binding fragment that binds to an antigen other than CD3; and wherein said control bispecific antigen binding construct consists of said anti-CD3 binding fragment consisting of said sequence of **SEQ ID NO:206** and said reference antigen binding fragment.

50. The polypeptide of claim 49, wherein said T_m of said first antigen binding fragment is at least 2 °C greater, or at least 3°C greater, or at least 4°C greater, or at least 5°C greater than said T_m of said anti-CD3 binding fragment consisting of said sequence of **SEQ ID NO:206**.

51. The polypeptide of any one of claims 1-50, wherein said first antigen binding fragment (**AF1**) comprises a heavy chain variable region (VH_1), wherein said VH_1 comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or being identical to an amino acid sequence of **SEQ ID NO:102 or 105**.

52. The polypeptide of any one of claims 1-51, wherein said first antigen binding fragment (**AF1**) comprises a light chain variable region (VL_1), wherein said VL_1 comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% sequence identity or is identical to an amino acid sequence of any one of **SEQ ID NOS: 101, 103, 104, 106, or 107**.

53. The polypeptide of claim 51 or 52, wherein said VH_1 and said VL_1 is linked by a linker comprising an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table A**.

54. The polypeptide of any one of claims 1-53, wherein said first antigen binding fragment (**AF1**) comprises an amino acid sequence having at least 95%, 96%, 97%, 98%, 99% sequence identity or being identical to an amino acid sequence of any one of **SEQ ID NOS: 201-205**.

55. The polypeptide of any one of claims 1-53, wherein said first antigen binding fragment (**AF1**) specifically binds human or cynomolgus monkey (cyno) CD3.

56. The polypeptide of claim 55, wherein said first antigen binding fragment (**AF1**) specifically binds human CD3.

57. The polypeptide of any one of claims1-56, wherein said first antigen binding fragment (**AF1**) binds a CD3 complex subunit identified herein by CD3 epsilon, CD3 delta, CD3 gamma, or CD3 zeta unit of CD3.

58. The polypeptide of claim 57, wherein said first antigen binding fragment (**AF1**) binds a CD3 epsilon fragment of CD3.

59. The polypeptide of any one of claims 1-58, wherein said first antigen binding fragment (**AF1**) exhibits an isoelectric point (pI) that is less than or equal to 6.6.

60. The polypeptide of claim 59, wherein said first antigen binding fragment (**AF1**) exhibits an isoelectric point (pI) that is between 6.0 and 6.6, inclusive.

61. The polypeptide of any one of claims 1-60, wherein said first antigen binding fragment (**AF1**) exhibits an isoelectric point (pI) that is at least 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, or 1.0 pH units lower than the pI of a reference antigen binding fragment consisting of a sequence shown in **SEQ ID NO: 206**.

62. The polypeptide of any one of claims 1-61, wherein said first antigen binding fragment (**AF1**) specifically binds human or cyno CD3 with a dissociation constant (K_d) constant between about between about 10 nM and about 400 nM, as determined in an *in vitro* antigen-binding assay comprising a human or cyno CD3 antigen.

63. The polypeptide of any one of claims 1-62, wherein said first antigen binding fragment (**AF1**) specifically binds human or cyno CD3 with a dissociation constant (K_d) of less than about 10 nM, or less than about 50 nM, or less than about 100 nM, or less than about 150 nM, or less than about 200 nM, or less than about 250 nM, or less than about 300 nM, or less than about 350 nM, or less than about 400 nM, as determined in an *in vitro* antigen-binding assay.

64. The polypeptide of any one of claims 1-63, wherein said first antigen binding fragment (**AF1**) exhibits a binding affinity to CD3 that is at least 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, or at least 10-fold weaker relative to that of an antigen binding fragment consisting of an amino acid sequence of **SEQ ID NO:206**, as determined by the respective dissociation constants (K_d) in an *in vitro* antigen-binding assay.

65. The polypeptide of any one of claims 1-64, wherein said first antigen binding fragment (**AF1**) is a chimeric or a humanized antigen binding fragment.

66. The polypeptide of any one of claims 1-65, wherein said first antigen binding fragment (**AF1**) is an Fv, Fab, Fab', Fab'-SH, linear antibody, or single-chain variable fragment (scFv).

67. The polypeptide of any one of claims 1-66, wherein said second antigen binding fragment (**AF2**) is an Fv, Fab, Fab', Fab'-SH, linear antibody, a single domain antibody, or single-chain variable fragment (scFv).

68. The polypeptide of any one of claims 1-66, wherein said first and second antigen binding fragments are configured as an (Fab')2 or a single chain diabody.

69. The polypeptide of any one of claims 1-68, wherein said second antigen binding fragment (**AF2**) comprises:

a heavy chain variable region (VH_H) comprising an amino acid sequence identified herein by **SEQ ID NOS: 778-783**, and

a light chain variable region (VL_H) comprising an amino acid sequence identified herein by **SEQ ID NOS: 878-883**.

70. The polypeptide of claim 69, wherein said VH_H and said VL_H is linked by a linker comprising an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table A**.

71. The polypeptide of any one of claims 1-70, wherein said first and second antigen binding fragments are fused together by a peptide linker.

72. The polypeptide of claim 70, wherein said peptide linker comprises 2 or 3 types of amino acids are glycine, serine, or proline.

73. The polypeptide of claim 70 or 72, wherein said peptide linker comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table B**.

74. The polypeptide of any one of claims 1-73, wherein:

said **XTEN** is a first extended recombinant polypeptide (**XTEN1**);

said plurality of non-overlapping sequence motifs, from which said **XTEN1** is formed, is a first plurality of non-overlapping sequence motifs;

said **BAR** is a first barcode fragment (**BAR1**); and

said **RS** is a first release segment (**RS1**); and

the polypeptide further comprising:

(d) a second extended recombinant polypeptide (**XTEN2**), comprising:

a second barcode fragment (**BAR2**) releasable from said polypeptide upon digestion by said protease; and

(e) a second release segment (**RS2**) positioned between said second XTEN (**XTEN2**) and said bispecific antibody construct (**BsAb**),

wherein said **XTEN2** is characterized in that:

(i) it comprises at least 100, or at least 150 amino acids;

(ii) at least 90% of its amino acid residues are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P); and

(iii) it comprises at least 4 different types of amino acids are G, A, S, T, E, or P;

wherein said second barcode fragment (**BAR2**) differs in sequence and molecular weight from all other peptides fragments that are releasable from said polypeptide upon complete digestion of said polypeptide by said protease; and

wherein said second barcode fragment (**BAR2**) does not include said N-terminal amino acid or said C-terminal amino acid of said polypeptide.

75. The polypeptide of claim 74, wherein said **XTEN1** is positioned N-terminal of said bispecific antibody construct and said **XTEN2** is positioned C-terminal of said bispecific antibody construct.

76. The polypeptide of claim 74, wherein said **XTEN1** is positioned C-terminal of said bispecific antibody construct and said **XTEN2** is positioned N-terminal of said bispecific antibody construct.

77. The polypeptide of any one of claims 74-76, wherein:

(iv) said **XTEN2** is formed from a second plurality of non-overlapping sequence motifs that are each from 9 to 14 amino acids in length, wherein said second plurality of non-overlapping sequence motifs comprise:

(1) a second set of non-overlapping sequence motifs, wherein each non-overlapping sequence motif of said second set of non-overlapping sequence motifs is repeated at least two times in said second XTEN; and

(2) a non-overlapping sequence motif that occurs only once within said second XTEN; and

wherein said second barcode fragment (**BAR2**) includes at least part of said non-overlapping sequence motif that occurs only once within said second XTEN.

78. The polypeptide of claim 77, wherein said second set of non-overlapping sequence motifs are each independently identified herein by **SEQ ID NOS: 179-200 and 1715-1722**.

79. The polypeptide of claim 78, wherein said second set of non-overlapping sequence motifs are each independently identified herein by **SEQ ID NOS: 186-189**.

80. The polypeptide of claim 79, wherein said second set of non-overlapping sequence motifs comprise at least two, at least three, or all four of the sequence motifs **SEQ ID NOS: 186-189**.

81. The polypeptide of any one of claims 74-80, wherein said XTEN2 comprises a length of from 100 to 3,000, from 150 to 3,000, from 100 to 1,000, or from 150 to 1,000 amino acid residues.

82. The polypeptide of any one of claims 74-81, wherein said XTEN2 comprises a length of at least 200, at least 250, at least 300, at least 350, at least 400, at least 450, or at least 500 amino acid residues.

83. The polypeptide of any one of claims 74-82, wherein at least 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% of the amino acid residues of said XTEN2 are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P).

84. The polypeptide of any one of claims 74-83, wherein said XTEN2 has at least 90%, at least 92%, at least 95%, at least 98%, at least 99% or 100% sequence identity to a sequence set forth in Table 3a.

85. The polypeptide of any one of claims 74-84, wherein said second barcode fragment (**BAR2**) does not include a glutamic acid that is immediately adjacent to another glutamic acid, if present, in said XTEN2.

86. The polypeptide of any one of claims 74-85, wherein said second barcode fragment (**BAR2**) has a glutamic acid at its C-terminus.

87. The polypeptide of any one of claims 74-86, wherein said second barcode fragment (**BAR2**) has an N-terminal amino acid that is immediately preceded by a glutamic acid residue.

88. The polypeptide of any one of claims 74-87, wherein said second barcode fragment (**BAR2**) is positioned a distance from either said N-terminus of said polypeptide or said C-terminus of said polypeptide, wherein said distance is from 10 to 150 amino acids, or from 10 to 125 amino acids in length.

89. The polypeptide of any one of claims 74-88, wherein said second barcode fragment (**BAR2**) is characterized in that:

(i) it does not include a glutamic acid that is immediately adjacent to another glutamic acid, if present, in said XTEN2;

(ii) it has a glutamic acid at its C-terminus;

(iii) it has an N-terminal amino acid that is immediately preceded by a glutamic acid residue; and

(iv) it is positioned a distance from either said N-terminus of said polypeptide or said C-terminus of said polypeptide, wherein said distance is from 10 to 150 amino acids, or from 10 to 125 amino acids in length.

90. The polypeptide of any one of claims 87-89, wherein said glutamic acid residue that precedes said N-terminal amino acid of said **BAR2** is not immediately adjacent to another glutamic acid residue.

91. The polypeptide of any one of claims 74-90, wherein said second barcode fragment (**BAR2**) does not include a second glutamic acid residue at a position other than the C-terminus of said second barcode fragment (**BAR2**) unless said second glutamic acid is immediately followed by a proline.

92. The polypeptide of any one of claims 74-91, wherein said **XTEN2** is positioned N-terminal of said bispecific antibody construct (**BsAb**), and wherein said second barcode fragment (**BAR2**) is positioned within 200, within 150, within 100, or within 50 amino acids of said N-terminus of said polypeptide.

93. The polypeptide of any one of claims 74-92, wherein said **XTEN2** is positioned N-terminal of said bispecific antibody construct (**BsAb**), and wherein said second barcode fragment (**BAR2**) is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from said N-terminus of said protein.

94. The polypeptide of any one of claims 74-93, wherein said **XTEN2** is positioned C-terminal of said bispecific antibody construct (**BsAb**), and wherein said second barcode fragment (**BAR2**) is positioned within 200, within 150, within 100, or within 50 amino acids of said C-terminus of said polypeptide.

95. The polypeptide of any one of claims 74-94, wherein said **XTEN2** is positioned C-terminal of said bispecific antibody construct (**BsAb**), and said second barcode fragment (**BAR2**) is positioned at a location that is between 10 and 200, between 30 and 200, between 40 and 150, or between 50 and 100 amino acids from said C-terminus of said protein.

96. The polypeptide of any one of claims 74-95, wherein said second barcode fragment (**BAR2**) is at least 4 amino acids in length.

97. The polypeptide of claim 96, wherein said second barcode fragment (**BAR2**) is between 4 and 20, between 5 and 15, between 6 and 12, or between 7 and 10 amino acids in length.

98. The polypeptide of any one of claims 74-97, wherein said second barcode fragment (**BAR2**) comprises an amino acid sequence set forth in Table 2.

99. The polypeptide of any one of claims 74-98, wherein said **XTEN2** has a length defined by a proximal end and a distal end, wherein (1) said proximal end of said **XTEN2** is positioned, relative to said distal end, closer to said bispecific antibody construct (**BsAb**), and wherein (2) said second barcode fragment (**BAR2**) is positioned within a region of said **XTEN2** that extends, as measured from said distal end of said **XTEN2**, between 5% and 50%, between 7% and 40%, or between 10% and 30% of said length of said **XTEN2**.

100. The polypeptide of any one of claims 74-99, wherein said **XTEN2** further comprises additional one or more barcode fragments, wherein said additional one or more barcode fragments of said **XTEN2** each differ in sequence and molecular weight from all other peptides fragments that are releasable from said polypeptide upon complete digestion of said polypeptide by said protease.

101. The polypeptide of any one of claims 74-100, wherein said first release segment (**RS1**) and said second release segment (**RS2**) are identical in sequence.

102. The polypeptide of any one of claims 74-100, wherein said first release segment (**RS1**) and said second release segment (**RS2**) are not identical in sequence.

103. The polypeptide of any one of claims 74-102, wherein said second release segment (**RS2**) comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence identified herein by **SEQ ID NOS:7001-7626**.

104. The polypeptide of any one of claims 74-103, wherein said first release segment (**RS1**) and said second release segment (**RS2**) are each a substrate for cleavage by multiple proteases at one, two, or three cleavage sites within each release segment sequence.

105. The polypeptide of any one of claims 74-104 expressed as a fusion protein, wherein said fusion protein, in an uncleaved state, has a structural arrangement from N-terminus to C-terminus identified herein by XTEN1-RS1-AF1-AF2-RS2-XTEN2, XTEN1-RS1-AF2-AF1-RS2-XTEN2, XTEN2-RS2-AF1-AF2-RS1-XTEN1, XTEN2-RS2-AF2-AF1-RS1-XTEN1, XTEN1-RS1-diabody-RS2-XTEN2, or XTEN2-RS2-diabody-RS1-XTEN1, wherein

said diabody comprises a light chain variable region (VL_I) of said AF1, a heavy chain variable region (VH_I) of said AF1, a light chain variable region (VL_{II}) of said AF2, and a heavy chain variable region (VH_{II}) of said AF2.

106. The polypeptide of claim 74-105, wherein said spacer of said first release segment (**RS1**) is a first spacer, and wherein said second release segment (**RS2**) is fused to said bispecific antibody construct (**BsAb**) by a second spacer.

107. The polypeptide of claim 106, wherein said second spacer comprises at least 4 types of amino acids are glycine (G), alanine (A), serine (S), threonine (T), glutamate (E) or proline (P).

108. The polypeptide of claim 106, wherein said second spacer comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table C**.

109. The polypeptide of claim 74, wherein:

(a) said **XTEN1** comprises an amino acid sequence having at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence set forth in **Table 3a**;

(b) said **BsAb** comprises:

(I) said **AF1** comprising light chain complementarity-determining regions 1 (CDR-L1), 2 (CDR-L2), and 3 (CDR-L3) and heavy chain complementarity-determining regions 1 (CDR-H1), 2 (CDR-H2), and 3 (CDR-H3), wherein said CDR-H1, said CDR-H2, and said CDR-H3 comprise amino acid sequences of **SEQ ID NOS: 8, 9, and 10**, respectively; and

(II) said **AF2** comprising a light chain variable region (VL_{II}) identified herein by **SEQ ID NOS: 778-783** and a heavy chain variable region (VH_{II}) identified herein by **SEQ ID NOS: 878-883**;

(c) said **RS1** comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence identified herein by **SEQ ID NOS: 7001-7626**;

(d) said **XTEN2** comprises an amino acid sequence having at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence set forth in **Table 3a**; and

(e) said **RS2** comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to a sequence identified herein by **SEQ ID NOS: 7001-7626**,

wherein said polypeptide has a structural arrangement from N-terminus to C-terminus identified herein by: XTEN1-RS1-AF2-AF1-RS2-XTEN2, XTEN1-RS1-AF1-AF2-RS2-XTEN2, XTEN2-RS2-AF2-AF1-RS1-XTEN1, or XTEN2-RS2-AF1-AF2-RS1-XTEN1.

110. The polypeptide of any one of claims 1-109, wherein the polypeptide has a terminal half-life that is at least two-fold longer compared to the bispecific antibody construct not linked to any XTEN.

111. The polypeptide of any one of claims 1-110, wherein the polypeptide is less immunogenic compared to the bispecific antibody construct not linked to any XTEN, as ascertained by measuring production of IgG antibodies that selectively bind to said bispecific antibody construct after administration of comparable doses to a subject.

112. The polypeptide of any one of claims 1-111, wherein the polypeptide exhibits an apparent molecular weight factor under physiological conditions that is greater than about 3, greater than about 4, greater than about 5, or greater than about 6.

113. The polypeptide of claim 1, wherein the polypeptide comprises an amino acid sequence having at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or 100% sequence identity to a sequence set forth in **Table D**.

114. A pharmaceutical composition comprising a polypeptide of any one of claims 1-113 and one or more pharmaceutically suitable excipients.

115. The pharmaceutical composition of claim 114, wherein said pharmaceutical composition is formulated for intradermal, subcutaneous, oral, intravenous, intra-arterial, intraabdominal, intraperitoneal, intrathecal, or intramuscular administration.

116. The pharmaceutical composition of claim 114 or 115, wherein said pharmaceutical composition is in a liquid form or frozen.

117. The pharmaceutical composition of any one of claims 114-116, wherein said pharmaceutical composition is in a pre-filled syringe for a single injection.

118. The pharmaceutical composition of claim 117, wherein said pharmaceutical composition is formulated as a lyophilized powder to be reconstituted prior to administration.

119. The pharmaceutical composition of claim 114, wherein said composition is in a pharmaceutical combination at least one additional therapeutic agent selected from the group consisting of an antibody, an antibody fragment, an antibody conjugate, a cytotoxic agent, a toxin, a radionuclide, an immunomodulator, a photoactive therapeutic agent, a radiosensitizing agent, a hormone, an anti-angiogenesis agent, and combinations thereof.

120. The pharmaceutical composition of claim 119, wherein said additional therapeutic agent is a PD-1/PD-L1(2) inhibitor.

121. The pharmaceutical combination of claim 120, wherein the PD-1/PD-L1(2) inhibitor is an anti-PD-1 antibody or an anti-PD-L1 antibody or an anti-PD-L2 antibody.

122. The pharmaceutical combination of claim 121, wherein the PD-1/PD-L1(2) inhibitor is an anti-PD-1 antibody selected from the group comprising nivolumab (Opdivo, BMS-936558, MDX1106), pembrolizumab (Keytruda, MK-3475, lambrolizumab), pidilizumab (CT-011), PDR-001, JS001, STI-A1110, AMP-224 and AMP-514 (MEDI0680).

123. The pharmaceutical combination of claim 121, wherein the PD-1/PD-L1(2) inhibitor is an anti-PD-L1 antibody selected from the group comprising atezolizumab (Tecentriq, MPDL3280A), durvalumab (MEDI4736), avelumab (MSB0010718C), BMS-936559 (MDX1105) and LY3300054.

124. The pharmaceutical combination of claim 121, wherein the PD-1/PD-L1(2) inhibitor is an anti-PD-L2 antibody.

125. The pharmaceutical combination of claim 119, wherein the combination is a combination pack containing the components separate from one another.

126. The pharmaceutical combination of claim 119, wherein the components are administered in separate dosage forms simultaneously or sequentially for use in the treatment of the same disease.

127. The pharmaceutical combination of claim 119 for use as medicament for treating hyper-proliferative disorders.

128. The pharmaceutical combination of claim 127, wherein the hyper-proliferative disorders are selected from the group consisting of cancers of the breast, respiratory tract, brain, reproductive organs, digestive tract, urinary tract, eye, liver, skin, head and neck, thyroid, parathyroid and their distant metastases.

129. Use of a polypeptide of any one of claims 1-112 or a pharmaceutical combination of any of claims 119-128 in the preparation of a medicament for the treatment of a disease in a subject.

130. The use of claim 129, wherein said disease is cancer.

131. A method of treating a disease in a subject, comprising administering to said subject in need thereof one or more therapeutically effective doses of a pharmaceutical composition of any one of claims 114-118 or a pharmaceutical combination of any one of claims 119-128.

132. The method of claim 131, wherein said disease is cancer.

133. The method of claim 132, wherein said cancer is selected from the group consisting of glioblastoma, melanoma, cholangio carcinoma, small cell lung cancer, colorectal cancer, prostate cancer, vaginal cancer, angiosarcoma, non-small cell lung cancer, appendiceal cancer, squamous cell cancer, salivary duct carcinoma, adenoid cystic carcinoma, small intestine cancer, and gallbladder cancer.

134. The method of any one of claims 131-133, wherein the pharmaceutical composition is administered to the subject as one or more therapeutically effective doses.

135. The method of any one of claims 131-134, wherein the pharmaceutical composition is administered to the subject as one or more therapeutically effective doses over an effective dosing period.

136. The method of any one of claims 131-134, wherein the subject is a mouse, rat, monkey, or human.

137. A nucleic acid comprising (a) a polynucleotide sequence encoding a polypeptide of any one of claims 1-112; or (b) a reverse complement of said polynucleotide sequence of (a).

138. An expression vector comprising said polynucleotide sequence of claim 1-112 and a recombinant regulatory sequence operably linked to said polynucleotide sequence.

139. A host cell, comprising said expression vector of claim 138.

140. The host cell of claim 139, wherein the host cell is a prokaryote.

141. The host cell of claim 140, wherein the host cell is *E. coli*.

142. The host cell of claim 139, wherein the host cell is a mammalian cell.