

CORRECTED VERSION

(19) World Intellectual Property
Organization

International Bureau

(43) International Publication Date
09 January 2020 (09.01.2020)(10) International Publication Number
WO 2020/009992 A8

(51) International Patent Classification:

A61K 31/282 (2006.01) *C07K 14/705* (2006.01)
A61K 38/16 (2006.01) *C07K 14/495* (2006.01)
A61K 38/17 (2006.01) *C07K 16/28* (2006.01)
A61K 39/395 (2006.01) *C07K 16/46* (2006.01)
C07K 14/71 (2006.01)

(21) International Application Number:

PCT/US2019/040129

(22) International Filing Date:

01 July 2019 (01.07.2019)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

62/693,042 02 July 2018 (02.07.2018) US
 62/801,014 04 February 2019 (04.02.2019) US

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(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DJ, DK, DM, DO,

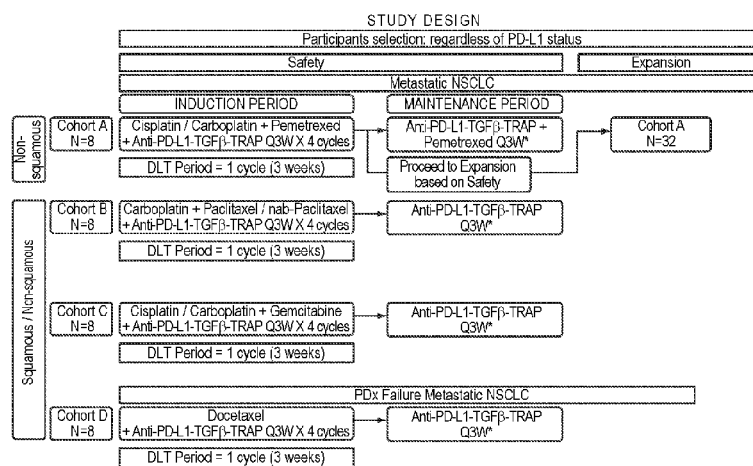
(54) Title: COMBINATION THERAPY WITH TARGETED TGF- β INHIBITION FOR TREATMENT OF ADVANCED NON-SMALL CELL LUNG CANCER

FIG. 8

(57) Abstract: This disclosure relates generally to methods for treating a subject diagnosed with advanced non-small-cell lung cancer (NSCLC), involving targeted TGF- β inhibition with a bi-functional fusion protein, in combination with administration of systemic chemotherapeutic agents, wherein the combination of the bi-functional fusion protein of the present disclosure with systemic chemotherapeutic agents enhances anticancer efficacy over systemic chemotherapeutic agents alone.

[Continued on next page]

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DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JO, JP, KE, KG, KH, KN, KP, KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

- (84) Designated States** (*unless otherwise indicated, for every kind of regional protection available*): ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Published:

- *with international search report (Art. 21(3))*
- *with sequence listing part of description (Rule 5.2(a))*

(48) Date of publication of this corrected version:

13 August 2020 (13.08.2020)

(15) Information about Correction:

see Notice of 13 August 2020 (13.08.2020)

COMBINATION THERAPY WITH TARGETED TGF- β INHIBITION FOR TREATMENT OF ADVANCED NON-SMALL CELL LUNG CANCER

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the benefit of and priority to U.S. Provisional Patent Application No. 62/693,042, filed July 2, 2018; and to U.S. Provisional Patent Application No. 62/801,014, filed February 4, 2019, the entire disclosures of which are incorporated by reference herein.

SEQUENCE LISTING

[0002] The instant application contains a Sequence Listing which has been submitted electronically in ASCII format and is hereby incorporated by reference in its entirety. Said ASCII copy, created on June 19, 2019, is named EMD-011WO_SL_ST25.txt and is 75,851 bytes in size.

FIELD OF THE DISCLOSURE

[0003] The present disclosure relates generally to methods for treating a subject diagnosed with advanced non-small-cell lung cancer (NSCLC), involving targeted TGF- β inhibition with a bi-functional fusion protein, in combination with systemic chemotherapeutic agents.

BACKGROUND

[0004] NSCLC is a heterogeneous group of tumors that can be broadly classified as squamous or non-squamous. Non-squamous NSCLC includes lung adenocarcinomas and large-cell undifferentiated carcinomas.

[0005] Approximately half of all patients with NSCLC present with advanced (stage IV) or metastatic disease at diagnosis. Traditional platinum-based doublet chemotherapy has been shown to improve quality of life and extend survival of advanced NSCLC patients, but this group of patients is considered incurable by currently available treatments and the median survival of patients with advanced NSCLC is 8-10 months. Studies of prolonged treatment with standard chemotherapeutic agent pemetrexed (or “continuation maintenance therapy”) significantly improves survival (*see, e.g., V. Polo and B. Besse, Maintenance Strategies in Stage IV Non-Small-Cell Lung Cancer (NSCLC): In Which Patients, With Which Drugs?* 25 ANNALS

OF ONCOLOGY 1283-1293 (2014); *see also* L.G. Paz-Ares *et al.*, *PARAMOUNT: Final Overall Survival Results of the Phase III Study of Maintenance Pemetrexed Versus Placebo Immediately After Induction Treatment With Pemetrexed Plus Cisplatin for Advanced Nonsquamous Non-Small-Cell Lung Cancer*, 31 JOURNAL OF CLINICAL ONCOLOGY 2895–2902 (2013)). However, 5 early second-line chemotherapy (“switch maintenance therapy”) has not shown any clinical benefit. The addition of a third chemotherapeutic agent to first-line treatment regimens failed to improve efficacy. *See, e.g.*, S. Ramalingam and C. Belani, *Systemic Chemotherapy for Advanced Non-Small Cell Lung Cancer: Recent Advances and Future Directions*, 13 THE ONCOLOGIST 5-13 (suppl 1) (2008). Moreover, toxicity increase was observed with the addition of a third agent. 10 *Id.*

[0006] Recent efforts to improve therapy for advanced NSCLC have focused on adding a targeted agent, such as EGFR inhibitors or bevacizumab, to standard platinum-based chemotherapy regimens. *Id.* However, none of these are curative. *Id.* There is a need, therefore, to improve the outcome of therapy for advanced and metastatic NSCLC over platinum-based 15 doublet chemotherapy.

[0007] US patent application publication number US 20150225483 A1, incorporated herein by reference, describes a bi-functional fusion protein that combines an anti-programmed death ligand 1 (PD-L1) antibody with the soluble extracellular domain of tumor growth factor beta receptor type II (TGF β RII) as a TGF β neutralizing “Trap,” into a single molecule. Specifically, 20 the protein is a heterotetramer, consisting of the two immunoglobulin light chains of anti-PD-L1, and two heavy chains comprising the heavy chain of anti-PD-L1 genetically fused via a flexible glycine-serine linker to the extracellular domain of the human TGF β RII (*see* Fig. 1). This anti-PD-L1/TGF β Trap molecule is designed to target two major mechanisms of immunosuppression in the tumor microenvironment. US patent application publication number US 20150225483 A1 25 describes administration of the Trap molecule at doses based on the patient’s weight.

[0008] The present disclosure provides methods for treating a treatment naïve subject or diagnosed with advanced NSCLC or a PDx failure metastatic NSCLC subject, including both squamous and non-squamous NSCLC, with an anti-PD-L1/TGF β Trap molecule in combination with administration of systemic chemotherapeutic agents.

SUMMARY OF THE DISCLOSURE

[0009] For an effective treatment of patients diagnosed with advanced NSCLC, the present disclosure provides a therapeutic regimen that treats advanced NSCLC, and improves disease prognosis and overall survival of advanced NSCLC patients. The advanced NSCLC being
5 treated can be either squamous or non-squamous NSCLC, and is independent of baseline PD-L1 expression levels.

[0010] In one aspect, the present disclosure provides a method of treating advanced NSCLC by administering anti-PD-L1/TGF β Trap in combination with systemic chemotherapeutic agents to induce tumor cell death, while simultaneously targeting two immune suppressive pathways:
10 PD-L1 and TGF- β . In one aspect, the present disclosure provides body weight-independent dosage regimens for targeted TGF- β inhibition with a bi-functional fusion protein in combination with systemic chemotherapy agents for use in a method of treating a treatment naïve subject diagnosed with advanced NSCLC or a PDx failure metastatic NSCLC subject.

[0011] In one aspect, the present disclosure provides a two-step method of treating advanced
15 NSCLC or inhibiting NSCLC tumor growth in a treatment naïve subject or a PDx failure metastatic NSCLC subject in need thereof, in which the first step involves administering to the subject a dose of at least 1200 mg (*e.g.*, 2400 mg) of anti-PD-L1/TGF β Trap, with concurrent systemic chemotherapy, and the second step involves administering at least 1200 mg (*e.g.*, 2400 mg) of the anti-PD-L1/TGF β Trap. The NSCLC can be either squamous or non-squamous
20 NSCLC. The systemic chemotherapy can be platinum-based chemotherapy, for example, including combinations of cisplatin or carboplatin with gemcitabine, docetaxel, or paclitaxel (nanoparticle albumin-bound (nab)-paclitaxel, or albumin unbound paclitaxel).

[0012] In one aspect, the present disclosure provides a two-step method of treating advanced non-squamous NSCLC or inhibiting non-squamous NSCLC tumor growth in a treatment-naïve
25 subject in need thereof, in which the first step involves administering to the subject a dose of at least 1200 mg (*e.g.*, 2400 mg) of anti-PD-L1/TGF β Trap, with concurrent systemic chemotherapy including pemetrexed, and the second step involves administering at least 1200 mg (*e.g.*, 2400 mg) of the anti-PD-L1/TGF β Trap in combination with pemetrexed as the only chemotherapeutic agent. The systemic chemotherapy in the first step can be platinum-based
30 chemotherapy, for example, including combinations of cisplatin or carboplatin with pemetrexed.

[0013] In one aspect, the present disclosure provides a two-step method of treating advanced non-squamous NSCLC or inhibiting non-squamous NSCLC tumor growth in a treatment-naïve subject in need thereof, in which the first step involves administering to the subject a dose of about 2400 mg of anti-PD-L1/TGFβ Trap, with concurrent systemic chemotherapy including
5 pemetrexed, and the second step involves administering about 2400 mg of the anti-PD-L1/TGFβ Trap in combination with pemetrexed. The systemic chemotherapy can be platinum-based chemotherapy, for example, including combinations of cisplatin/carboplatin with pemetrexed.

[0014] In one aspect, the present disclosure provides a method of treating advanced non-small cell lung cancer (NSCLC) or inhibiting NSCLC tumor growth in subject indicated as
10 having metastatic NSCLC disease progression on previous treatment with an immunotherapy in combination with chemotherapy, or on previous treatment with chemotherapy followed by treatment with an immunotherapy, or on previous treatment with an immunotherapy followed by platinum-based chemotherapy, the method comprising a first step of administering to the subject a dose of at least 1800 mg of an anti-PD-L1/ TGFβ Trap protein as provided in the present
15 disclosure, with concurrent systemic chemotherapy comprising docetaxel, and a second step of administering at least 1800 mg of anti-PD-L1/ TGFβ Trap protein.

[0015] In one aspect, the present disclosure provides a method of treating advanced non-small cell lung cancer (NSCLC) or inhibiting NSCLC tumor growth in subject indicated as having metastatic NSCLC disease progression on previous treatment with an immunotherapy in
20 combination with chemotherapy, or on previous treatment with chemotherapy followed by treatment with an immunotherapy, or on previous treatment with an immunotherapy followed by platinum-based chemotherapy, the method comprising a first step of administering to the subject a dose of about 2400 mg of an anti-PD-L1/ TGFβ Trap protein as provided in the present disclosure, with concurrent systemic chemotherapy comprising docetaxel, and a second step of
25 administering about 2400 mg of anti-PD-L1/ TGFβ Trap protein.

[0016] The bifunctional protein of the present disclosure (anti-PD-L1/TGFβ Trap molecule) includes a first and a second polypeptide. The first polypeptide includes: (a) at least a variable region of a heavy chain of an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGFβRII), or a fragment
30 thereof, capable of binding Transforming Growth Factor β (TGFβ) (*e.g.*, a soluble fragment). The second polypeptide includes at least a variable region of a light chain of an antibody that

binds PD-L1, in which the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1 (*e.g.*, any of the antibodies or antibody fragments described herein). Because the bifunctional protein of the present disclosure binds to two targets, (1) PD-L1, which is largely membrane bound, and (2)

- 5 TGF β , which is soluble in blood and interstitium, the BW-independent dosing regimen requires a dose that is effective not only to inhibit PD-L1 at the tumor site but also sufficient to inhibit TGF β .

[0017] The disclosure also features a method of promoting local depletion of TGF β . The method includes administering a protein described above, where the protein binds TGF β in
10 solution, binds PD-L1 on a cell surface, and carries the bound TGF β into the cell (*e.g.*, a cancer cell).

[0018] The disclosure also features a method of inhibiting SMAD3 phosphorylation in a cell (*e.g.*, a cancer cell or an immune cell), the method including exposing the cell in the tumor microenvironment to a protein described above.

15 [0019] Other embodiments and details of the disclosure are presented herein below.

BRIEF DESCRIPTION OF THE DRAWINGS

[0020] **FIG. 1** is a schematic drawing of an anti-PD-L1/TGF β Trap molecule including one anti-PD-L1 antibody fused to two extracellular domains (ECDs) of TGF β Receptor II via a (Gly₄Ser)₄Gly (SEQ ID NO: 11) linker.

20 [0021] **FIG. 2** shows a graph of a two-step ELISA demonstrating that anti-PD-L1/TGF β Trap simultaneously binds to both PD-L1 and TGF β .

[0022] **FIG. 3** is a graph showing anti-PD-L1/TGF β Trap induces a dramatic increase in IL-2 levels.

[0023] **FIG. 4A** is a graph showing *in vivo* depletion of TGF β 1 in response to the anti-PD-L1/TGF β Trap. Line graphs represent naïve, isotype control, and three different doses, as indicated in the legend. **FIG. 4B** is a graph showing *in vivo* depletion of TGF β 2 in response to the anti-PD-L1/TGF β Trap. Line graphs represent naïve, isotype control, and three different doses, as indicated in the legend. **FIG. 4C** is a graph showing *in vivo* depletion of TGF β 3 in
25

response to the anti-PD-L1/TGF β Trap. Line graphs represent naïve, isotype control, and three different doses, as indicated in the legend. **FIG. 4D** is a graph showing that occupancy of PD-L1 by the anti-PD-L1/TGF β Trap supports a receptor binding model in the EMT-6 tumor system.

[0024] **FIG. 5** is a graph showing anti-tumor efficacy of anti-PD-L1/TGF β Trap control (anti-PD-L1(mut)/TGF β) in Detroit 562 xenograft model.

[0025] **FIG. 6A** is a box-plot of C_{avg} distribution for an entire population for a fixed (1200 mg) versus mg/kg based dosing (17.65 mg/kg) in a simulated population of 68 kg median body weight. **FIG. 6B** is a box-plot of exposure AUC distribution for an entire population for a fixed (1200 mg) versus mg/kg based dosing (17.65 mg/kg) in a simulated population of 68 kg median body weight. **FIG. 6C** is a box-plot of C_{trough} distribution for an entire population for a fixed (1200 mg) versus mg/kg based dosing (17.65 mg/kg) in a simulated population of 68 kg median body weight. **FIG. 6D** is a box-plot of C_{max} distribution for an entire population for a fixed (1200 mg) versus mg/kg based dosing (17.65 mg/kg) in a simulated population of 68 kg median body weight.

[0026] **FIG. 6E** is a box-plot of C_{avg} distribution for an entire population for a fixed (500 mg) versus mg/kg based dosing (7.35 mg/kg) in a simulated population of 68 kg median body weight. **FIG. 6F** is a box-plot of exposure AUC distribution for an entire population for a fixed (500 mg) versus mg/kg based dosing (7.35 mg/kg) in a simulated population of 68 kg median body weight. **FIG. 6G** is a box-plot of C_{trough} distribution for an entire population for a fixed (500 mg) versus mg/kg based dosing (7.35 mg/kg) in a simulated population of 68 kg median body weight. **FIG. 6H** is a box-plot of C_{max} distribution for an entire population for a fixed (500 mg) versus mg/kg based dosing (7.35 mg/kg) in a simulated population of 68 kg median body weight.

[0027] **FIGs. 7A–7C** are graphs showing the predicted PK and PD-L1 receptor occupancy (“RO”) of anti-PD-L1/TGF β Trap molecules at doses and schedules associated with tumor stasis in mice. **FIG. 7A** is a graph showing the predicted plasma concentration vs. time. **FIG. 7B** is a graph showing the predicted PD-L1 RO vs. time in PBMC. **FIG. 7C** is a graph showing the predicted PD-L1 RO vs. time in tumor.

[0028] **FIG. 8** is a schematic of the study design described in Example 2. Abbreviations used in the figure: DLT = dose-limiting toxicity, PD = progression disease, NSCLC = non-small cell lung cancer, Q3W= every 3 weeks.

DETAILED DESCRIPTION

[0029] By “TGFβRII” or “TGFβ Receptor II” is meant a polypeptide having the wild-type human TGFβ Receptor Type 2 Isoform A sequence (*e.g.*, the amino acid sequence of NCBI Reference Sequence (RefSeq) Accession No. NP_001020018 (SEQ ID NO: 8)), or a polypeptide
5 having the wild-type human TGFβ Receptor Type 2 Isoform B sequence (*e.g.*, the amino acid sequence of NCBI RefSeq Accession No. NP_003233 (SEQ ID NO: 9)) or having a sequence substantially identical to the amino acid sequence of SEQ ID NO: 8 or of SEQ ID NO: 9. The TGFβRII may retain at least 0.1%, 0.5%, 1%, 5%, 10%, 25%, 35%, 50%, 75%, 90%, 95%, or 99% of the TGFβ-binding activity of the wild-type sequence. The polypeptide of expressed
10 TGFβRII lacks the signal sequence.

[0030] By a “fragment of TGFβRII capable of binding TGFβ” is meant any portion of NCBI RefSeq Accession No. NP_001020018 (SEQ ID NO: 8) or of NCBI RefSeq Accession No. NP_003233 (SEQ ID NO: 9), or a sequence substantially identical to SEQ ID NO: 8 or SEQ ID NO: 9 that is at least 20 (*e.g.*, at least 30, 40, 50, 60, 70, 80, 90, 100, 110, 120, 130, 140, 150,
15 160, 175, or 200) amino acids in length that retains at least some of the TGFβ-binding activity (*e.g.*, at least 0.1%, 0.5%, 1%, 5%, 10%, 25%, 35%, 50%, 75%, 90%, 95%, or 99%) of the wild-type receptor or of the corresponding wild-type fragment. Typically such fragment is a soluble fragment. An exemplary such fragment is a TGFβRII extra-cellular domain having the sequence of SEQ ID NO: 10. Certain other exemplary fragments of human TGFβRII capable of binding
20 TGFβ are represented by the sequence of SEQ ID NOs: 50, 51, 52, 53, or 54.

[0031] “Treatment naïve” refers to subjects or patients who have not received prior systemic treatment for their advanced NSCLC (stage IV) since being diagnosed with the disease.

[0032] “PDx failure metastatic NSCLC” refers to advanced NSCLC in subjects or patients who had metastatic NSCLC disease progression on previous treatment with PD-(L)1 inhibitors
25 (anti-PD-1 or anti-PD-L1 inhibitors (*e.g.*, antibodies)) in combination with chemotherapy, or on previous treatment with chemotherapy followed by treatment with PD-(L)1 inhibitors (anti-PD-1 or anti-PD-L1 inhibitors (*e.g.*, antibodies)), or on previous treatment with PD-(L)1 inhibitor (anti-PD-1 or anti-PD-L1 inhibitors (*e.g.*, antibodies)) followed by platinum-based chemotherapy.

[0033] “PD-L1 positive” or “PD-L1+” indicates $\geq 1\%$ PD-L1 positive tumor cells as determined, for example, by the Dako IHC 22C3 PharmDx assay, or by the VENTANA PD-L1 (SP263) assay.

[0034] “PD-L1 high” or “high PD-L1” refers to $\geq 80\%$ PD-L1 positive tumor cells as
5 determined by the PD-L1 IHC 73-10 assay (Dako), or tumor proportion score (TPS) $\geq 50\%$ as determined by the Dako IHC 22C3 PharmDx assay (TPS is a term of art related to the IHC 22C3 PharmDx assay, which describes the percentage of viable tumor cells with partial or complete membrane staining (*e.g.*, staining for PD-L1)). Both the IHC 73-10 and the IHC 22C3 assays select a similar patient population at their respective cutoffs. In certain embodiments, the
10 VENTANA PD-L1 (SP263) assay, which has high concordance with the 22C3 PharmDx assay (*see* Sughayer et al., *Appl. Immunohistochem. Mol. Morphol.*, (2018)), can also be used for determining PD-L1 high expression level.

[0035] By “substantially identical” is meant a polypeptide exhibiting at least 50%, desirably 60%, 70%, 75%, or 80%, more desirably 85%, 90%, or 95%, and most desirably 99% amino
15 acid sequence identity to a reference amino acid sequence. The length of comparison sequences will generally be at least 10 amino acids, desirably at least 15 contiguous amino acids, more desirably at least 20, 25, 50, 75, 90, 100, 150, 200, 250, 300, or 350 contiguous amino acids, and most desirably the full-length amino acid sequence.

[0036] By “patient” is meant either a human or non-human animal (*e.g.*, a mammal).
20 “Patient,” “subject,” “patient in need thereof,” and “subject in need thereof” are used interchangeably in the present disclosure, and refer to a living organism suffering from or prone to a disease or condition that can be treated by administration using the methods and compositions provided in the present disclosure.

[0037] The terms “treat,” “treating,” or “treatment,” and other grammatical equivalents as
25 used in the present disclosure, include alleviating, abating, ameliorating, or preventing a disease, condition or symptoms, preventing additional symptoms, ameliorating or preventing the underlying metabolic causes of symptoms, inhibiting the disease or condition, *e.g.*, arresting the development of the disease or condition, relieving the disease or condition, causing regression of the disease or condition, relieving a condition caused by the disease or condition, or stopping the
30 symptoms of the disease or condition, and are intended to include prophylaxis. The terms further

include achieving 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 disorder such that an improvement is observed in the patient, notwithstanding that the patient may still be afflicted with the underlying disorder.

[0038] The term “consolidation” in the context of a therapeutic regimen of the present disclosure is used as is commonly understood in the art. For example, according to the National Cancer Institute, the term “consolidation therapy” is a “[t]reatment that is given after cancer has disappeared following the initial therapy. Consolidation therapy is used to kill any cancer cells that may be left in the body. It may include radiation therapy, a stem cell transplant, or treatment with drugs that kill cancer cells. Also called intensification therapy and postremission therapy.” <https://www.cancer.gov/publications/dictionaries/cancer-terms/def/consolidation-therapy>, last visited on June 9, 2018.

[0039] The term “progression-free survival” or PFS is defined as the time from randomization (which can occur 6 or more weeks after treatment initiation) to the date of the first documented event of tumor progression or death in the absence of disease progression. The term “overall survival” is defined as the time from randomization until death from any cause. Progression-free survival is assessed by the investigators, according to RECIST, version 1.1, as a predefined sensitivity analysis.

[0040] By “cancer” is meant locally advanced and/or metastatic non-small cell lung cancer (NSCLC), including squamous or non-squamous NSCLC. Advanced/Stage IV NSCLC is used according to its plain and ordinary meaning, and refers to stages IVA or IVB of NSCLC, characterized by, for example, metastasis to one or more sites. Thus, in various embodiments, the cancer is a metastatic NSCLC.

[0041] The terms “risk,” “at risk,” and “risk factor,” are used here as conventionally understood in the art. For example, a risk factor is any attribute, characteristic or exposure of an individual that increases the likelihood of developing a disease or injury. In certain embodiments, a person at risk of developing a disease, disorder, or condition means that the person is exposed to a risk factor that contributes or enhances the probability of incidence of that disease, disorder, or condition.

[0042] Throughout the description and claims of the present disclosure the word “comprise” and other forms of the word, such as “comprising” and “comprises,” means including but not limited to, and is not intended to exclude, for example, other components.

5 [0043] By “co-administer” it is meant that a composition described herein is administered at the same time, just prior to, or just after the administration of additional therapies. The protein and the composition of the present disclosure can be administered alone or can be co-administered with a second, third, or fourth therapeutic agent(s) to a patient. Co-administration is meant to include simultaneous or sequential administration of the protein or composition individually or in combination (more than one therapeutic agent).

10 [0044] The term “a” is not meant to limit as a singular. In certain embodiments, the term “a” may refer to a plural form. As used throughout the present disclosure, the singular forms “a,” “an,” and “the” include plural reference unless the context clearly dictates otherwise. Thus, for example, a reference to “a composition” includes a plurality of such compositions, as well as a single composition.

15 [0045] A “reconstituted” formulation is one which has been prepared by dissolving a lyophilized formulation in an aqueous carrier such that the bifunctional molecule is dissolved in the reconstituted formulation. The reconstituted formulation is suitable for intravenous administration (IV) to a patient in need thereof.

20 [0046] The term “about” refers to any minimal alteration in the concentration or amount of an agent that does not change the efficacy of the agent in preparation of a formulation and in treatment of a disease or disorder. In embodiments, the term “about” may include $\pm 15\%$ of a specified numerical value or data point.

25 [0047] Ranges can be expressed in the present disclosure as from “about” one particular value, and/or to “about” another particular value. When such a range is expressed, another aspect includes from the one particular value and/or to the other particular value. Similarly, when values are expressed as approximations, by use of the antecedent “about,” it is understood that the particular value forms another aspect. It is further understood that the endpoints of each of the ranges are significant both in relation to the other endpoint, and independently of the other endpoint. It is also understood that there are a number of values disclosed in the present
30 disclosure, and that each value is also disclosed as “about” that particular value in addition to the

value itself. It is also understood that throughout the application, data are provided in a number of different formats and that the data represent endpoints and starting points and ranges for any combination of the data points. For example, if a particular data point “10” and a particular data point “15” are disclosed, it is understood that greater than, greater than or equal to, less than, less than or equal to, and equal to 10 and 15 are considered disclosed as well as between 10 and 15. It is also understood that each unit between two particular units are also disclosed. For example, if 10 and 15 are disclosed, then 11, 12, 13, and 14 are also disclosed.

[0048] An “isotonic” formulation is one which has essentially the same osmotic pressure as human blood. Isotonic formulations will generally have an osmotic pressure from about 250 to 350 mOsmol/kgH₂O. The term “hypertonic” is used to describe a formulation with an osmotic pressure above that of human blood. Isotonicity can be measured using a vapor pressure or ice-freezing type osmometer, for example.

[0049] The term “buffering agent” refers to one or more components that when added to an aqueous solution is able to protect the solution against variations in pH when adding acid or alkali, or upon dilution with a solvent. In addition to phosphate buffers, there can be used glycinate, carbonate, citrate buffers and the like, in which case, sodium, potassium or ammonium ions can serve as counterion.

[0050] An “acid” is a substance that yields hydrogen ions in aqueous solution. A “pharmaceutically acceptable acid” includes inorganic and organic acids which are nontoxic at the concentration and manner in which they are formulated.

[0051] A “base” is a substance that yields hydroxyl ions in aqueous solution. “Pharmaceutically acceptable bases” include inorganic and organic bases which are non-toxic at the concentration and manner in which they are formulated.

[0052] A “lyoprotectant” is a molecule which, when combined with a protein of interest, prevents or reduces chemical and/or physical instability of the protein upon lyophilization and subsequent storage.

[0053] A “preservative” is an agent that reduces bacterial action and may be optionally added to the formulations herein. The addition of a preservative may, for example, facilitate the production of a multi-use (multiple-dose) formulation. Examples of potential preservatives

include octadecyldimethylbenzyl ammonium chloride, hexamethonium chloride, benzalkonium chloride (a mixture of alkylbenzyldimethylammonium chlorides in which the alkyl groups are long-chain compounds), and benzethonium chloride. Other types of preservatives include aromatic alcohols such as phenol, butyl and benzyl alcohol, alkyl parabens such as methyl or propyl paraben, catechol, resorcinol, cyclohexanol, 3-pentanol, and m-cresol.

[0054] A “surfactant” is a surface active molecule containing both a hydrophobic portion (*e.g.*, alkyl chain) and a hydrophilic portion (*e.g.*, carboxyl and carboxylate groups). Surfactant may be added to the formulations of the invention. Surfactants suitable for use in the formulations of the present invention include, but are not limited to, polysorbates (*e.g.* polysorbates 20 or 80); poloxamers (*e.g.* poloxamer 188); sorbitan esters and derivatives; Triton; sodium laurel sulfate; sodium octyl glycoside; lauryl-, myristyl-, linoleyl-, or stearyl-sulfobetaine; lauryl-, myristyl-, linoleyl- or stearyl-sarcosine; linoleyl-, myristyl-, or cetyl-betaine; lauramidopropyl-cocamidopropyl-, linoleamidopropyl-, myristamidopropyl-, palmidopropyl-, or isostearamidopropylbetaine (*e.g.*, lauroamidopropyl); myristamidopropyl-, palmidopropyl-, or isostearamidopropyl-dimethylamine; sodium methyl cocoyl-, or disodium methyl oleyl-aurate; and the MONAQUAT™ series (Mona Industries, Inc., Paterson, N.J.), polyethylene glycol, polypropyl glycol, and copolymers of ethylene and propylene glycol (*e.g.*, Pluronic, PF68 etc.).

Body Weight-Independent Dosing Regimen

[0055] Body weight-independent dosing regimens involving the administration to treatment naïve patients of at least 1200 mg of the bifunctional anti-PD-L1/TGFβ Trap molecules described herein have been developed, informed by the results of a variety of pre-clinical and clinical assessments of the molecules. Two studies investigated the safety, tolerability, and pharmacokinetics of the molecules, and included assessments of PD-L1 target occupancy on peripheral blood mononuclear cells obtained from the blood of treated patients and measurements of the concentrations of TGFβ1, TGFβ2, and TGFβ3. These assessments were based on data from a total of 350 subjects (dose escalation cohorts of 1, 3, 10 and 20 mg/kg in solid tumors, and expansion cohorts of 3 mg/kg, 10 mg/kg, 500 mg, and 1200 mg in selected tumor types).

PK/Efficacy Model (Mouse Model)

[0056] Experiments were also conducted to determine the efficacy of the anti-PD-L1/TGF β Trap molecule in a tumor model. Efficacy results from EMT-6 xenografts were used to establish the PK/Efficacy model. The established PK model in mice was used to simulate anti-PD-

5 L1/TGF β Trap plasma exposure for the efficacy experiment settings. The estimated parameters are reported in Table 1. The estimated KC₅₀ value was 55.3 μ g/mL, which represents the average plasma concentrations for which 50% of the maximal anti-tumor activity of the anti-PD-L1/TGF β Trap molecule could be achieved.

[0057] Basic diagnostics plots of the model revealed no model misspecification. The model
10 predictions are able to capture the tumor volume distributions. Conditional weighted residuals are normally distributed with a 0 mean and 1 variance without a trend. The PK/Efficacy model was then used to simulate tumor growth inhibition (TGI) using the human predicted concentration-time profiles at different doses.

[0058] **Table 1:** Mouse PK/Efficacy model parameters for anti-PD-L1/TGF β Trap molecule
15 in EMT-6 xenograft mice

Parameters	Estimate	Std	CV%	%IV
K_g (h^{-1})	0.068	0.0005	0.82	40
K_e (h^{-1})	0.055	0.0024	4.4	76
KC ₅₀ (ng/mL)	55324.6	522.3	4.4	232
K_{max}	2	0.09	1	93
Baseline (mm^3)	88.3	0.87	1	47

Response Analysis Based on PD-L1 Occupancy (in a Mouse Model)

[0059] Using the efficacy experiments, responses in mice have been analyzed and sorted by either tumor regression or tumor stasis, and PK and PD-L1 receptor occupancy (RO) have been
20 predicted based on the integrated PK/RO model. The approach demonstrated that an anti-PD-L1/TGF β Trap molecule plasma concentration between 40 and 100 μ g/mL associated with a PD-L1 RO above 95% in tumor is required to reach tumor regression. The plasma concentration of

anti-PD-L1/TGF β Trap molecule between 10 and 40 μ g/mL associated with a PD-L1 RO above 95% in periphery is required to reach tumor stasis.

[0060] Response analysis and predicted PK/RO in mice lead to **FIGs. 7A-7C**, which summarize the PK/RO/Efficacy for the anti-PD-L1/TGF β Trap molecule in mice. 95% of PD-L1 RO is achieved at a plasma concentration of 40 μ g/mL with an expected/estimate TGI of only about 65%. Increasing the concentration above 40 μ g/mL results in an additional increase in tumor growth inhibition. 95% of tumor growth inhibition is achieved at average plasma concentration of about 100 μ g/mL.

[0061] Based on the population PK model described below, a flat dose of at least 500 mg administered once every two weeks is required to maintain an average concentration of about 100 μ g/mL, while a flat dose of about 1200 mg administered once every two weeks is required to maintain a C_{trough} of about 100 μ g/mL. In certain embodiments about 1200 mg to about 3000 mg (*e.g.*, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, etc.) of a protein product of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap) is administered to a subject. In certain embodiments, about 1200 mg of anti-PD-L1/TGF β Trap molecule is administered to a subject once every two weeks. In certain embodiments, about 2400 mg of anti-PD-L1/TGF β Trap molecule is administered to a subject once every three weeks.

[0062] In embodiments, about 1200 mg to about 3000 mg (*e.g.*, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, etc.) of the protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1 is administered to a subject. In certain embodiments, about 1200 mg to about 3000 mg (*e.g.*, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, etc.) of the protein product with a first polypeptide that includes a first polypeptide comprising the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide comprising the amino acid sequences of SEQ ID NOs: 38, 39, and 40 is administered to a subject.

[0063] In certain embodiments, about 1200 mg of the protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1 is administered to a subject once every two weeks. In certain embodiments, about 1800 mg of the protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1 is administered to a subject once every three weeks. In certain embodiments, about 2100 mg of the protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1 is administered to a subject once every three weeks. In certain
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embodiments, about 2400 mg of the protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1 is administered to a subject once every three weeks.

[0064] In certain embodiments, about 1200 mg of the protein product that includes a first polypeptide comprising the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide comprising the amino acid sequences of SEQ ID NOs: 38, 39, and 40 is administered to a subject once every two weeks. In certain embodiments, about 1800 mg of the protein product that includes a first polypeptide comprising the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide comprising the amino acid sequences of SEQ ID NOs: 38, 39, and 40 is administered to a subject once every three weeks. In certain
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embodiments, about 2100 mg of the protein product that includes a first polypeptide comprising the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide comprising the amino acid sequences of SEQ ID NOs: 38, 39, and 40 is administered to a subject once every three weeks. In certain embodiments, about 2400 mg of the protein product that includes a first polypeptide comprising the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second
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polypeptide comprising the amino acid sequences of SEQ ID NOs: 38, 39, and 40 is administered to a subject once every three weeks.

Establishing Body Weight-Independent Dosing Regimen

[0065] Informed by the clinical and pre-clinical data, a new, body weight-independent dosing regimen for the administration of anti-PD-L1/TGFβ Trap molecules has been created to achieve
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less variability in exposure, reduce dosing errors, reduce the time necessary for dose preparation, and reduce drug wastage compared to the mg/kg dosing, thus facilitating favorable treatment

outcomes. According to one embodiment, a flat dose of at least 500 mg can be administered, regardless of the patient's body weight. According to another embodiment, a flat dose of at least 1200 mg can be administered, regardless of the patient's body weight. According to another embodiment, a flat dose of 1800 mg can be administered, regardless of the patient's body weight. Typically, such doses would be administered repeatedly, such as once every two weeks or once every 3 weeks, for example. For example, a flat dose of 1200 mg can be administered once every two weeks, or a flat dose of 1800 mg, 2100 mg, or 2400 mg can be administered once every three weeks.

Pharmacokinetic (PK) Analysis Sampling in Humans

10 [0066] An example of pharmacokinetic analysis to determine the optimal flat dose of the anti-PD-L1/TGF β Trap is provided by the experiments described below.

[0067] Serum samples for pharmacokinetic (PK) data analysis were collected before the start of the first dose and at the following time points after the first dose: on Day 1 immediately after the infusion and 4 hours after the start of the infusion; on Day 2 at least 24 hours after the Day 1 end of infusion; and on Days 8 and 15. At selected subsequent dosing occasions pre-dose, end-of-infusion and 2 to 8 hours after the end of infusion samples were collected on days 15, 29, 43. For later time points on days 57, 71 and 85, pre-dose samples were or were to be collected followed by once every 6 weeks PK sampling until 12 weeks, then once every 12 weeks PK sampling. In the expansion phase sparse PK sampling was conducted.

20 [0068] The PK data described above were used to produce a population PK model and to perform simulations of possible dosing regimens. A modeling method, known as the full approach model, described in Gastonguay, M., *Full Covariate Models as an Alternative to Methods Relying on Statistical Significance for Inferences about Covariate Effects: A Review of Methodology and 42 Case Studies*, (2011) p. 20, Abstract 2229, was applied to the population model data obtained from the simulations to obtain parameters having the following features: 2-compartment PK model with linear elimination, IIV on CL, V1, and V2, combined additive and proportional residual error, full covariate model on CL and V1. The following baseline covariates were included in the final model: age, weight, sex, race, albumin, CRP, platelet count, eGFR, hepatic impairment, ECOG score, tumor size, tumor type, and previous treatment with biologics. The following estimates of typical parameter estimates of pharmacokinetics of the

protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap) were obtained: clearance (CL) 0.0177 L/h (6.2%), central volume of distribution (V1) 3.64 L (8.81%), peripheral volume of distribution (V2) 0.513 L (25.1%), and inter-compartmental clearance (Q) 0.00219 L/h (17.8%). The inter-patient variability was 22% for CL, 20% for V1, and 135% for V2. Body weight was a
5 relevant covariate on both CL and V1. To support the flat dosing approach, the impact of the dosing strategy on the exposure variability of the protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap) was explored. Specifically, simulations were performed to compare the exposure distribution using a flat dosing approach of 1200 mg once every two weeks versus a BW-
10 adjusted dosing approach of either 17.65 mg/kg once every two weeks (corresponding to 1200 mg once every two weeks for a 68 kg subject or 15 mg/kg once every two weeks (corresponding to 1200 mg for a 80 kg subject). Further simulations were performed to compare the exposure distribution using a flat dosing approach of 500 mg once every two weeks versus a BW-adjusted dosing approach of 7.35 mg/kg once every two weeks (corresponding to 500 mg once every two weeks for a 68 kg subject). In addition, simulations were performed to assess the following flat
15 doses at once every three weeks: 1200 mg, 1400 mg, 1600 mg, 1800 mg, 2000 mg, 2200 mg, 2400 mg, 2600 mg, 2800 mg, 3000 mg.

[0069] The following methodology for simulations was used: N=200 sets of parameter estimates were drawn from multivariate normal distribution of parameter estimates, using the final PK model variance-covariance matrix. For each parameter estimate, 200 IIV estimates were
20 drawn from \$OMEGA multivariate normal distribution, resulting in total 40000 (200 x 200) subjects. The original dataset (N=380) was resampled with replacement to generate 40000 sets of matched covariates and steady-state exposure metrics (AUC, C_{avg}, C_{trough} and C_{max}) were generated for each dosing regimen.

[0070] Simulations showed that across a wide BW spectrum, variability in exposure is
25 slightly higher for BW-based dosing in comparison with fixed dosing. An example of exposure distribution at 17.65 mg/kg and 1200 mg flat dose, or 7.35 mg/kg and 500 mg flat dose for a median body weight of 68 kg is shown in FIGs. 6A and 6E, respectively. Simulations also showed the opposite trend in exposure distributions across weight quartiles across the patient population: low-weight patients have higher exposure with fixed dosing, whereas high-weight
30 patients have higher exposure with BW-adjusted dosing.

Establishing Efficacious Dose/Dosing Regimen in Humans: preliminary dose-response in 2nd Line Non-Small-Cell Lung Cancer (2L NSCLC) following once every 2 weeks (q2w) dosing of anti-PD-L1/TGFβ Trap

[0071] An example of the therapeutic efficacy of the anti-PD-L1/TGFβ Trap is established by the clinical study described below.

[0072] Patients with advanced NSCLC unselected for PD-L1 who progressed following 1st line standard treatment (no prior immunotherapy) were randomized to receive the anti-PD-L1/TGFβ Trap of the present disclosure at 500 mg or 1200 mg (n=40 per cohort) once every two weeks (q2w), until disease progression, unacceptable toxicity, or trial withdrawal. The primary objective was to assess best overall response (BOR) per Response Evaluation Criteria in Solid Tumors version 1.1 (RECIST v1.1). Other objectives included dose exploration and safety/tolerability assessment. Tumor cell PD-L1 expression levels (Ab clone 73-10 (Dako) [$>80\% = >50\%$ with Ab clone 22C3 (Dako)]) were characterized as PD-L1 $<1\%$, $\geq 1\%$ (PD-L1+), or $\geq 80\%$ (PD-L1-high). Tumor cell PD-L1 expression was evaluable in 75 patients.

[0073] As of data cut-off at the time of analysis, 80 patients received anti-PD-L1/TGFβ Trap for a median of 11.9 weeks (range, 2-66.1), with a median follow-up of 51.1 weeks. Ten patients remain on treatment. Investigator-assessed confirmed overall response rate (ORR) was 23.8% (500 mg ORR, 20.0%; 1200 mg ORR, 27.5%), with 18 partial responses (PR) seen across both dose levels, and 1 complete response (CR) seen at 1200 mg. As shown in Table 2, clinical activity was observed across PD-L1 expression levels: ORR was 37.0% in PD-L1+ and 85.7% in PD-L1-high patients at 1200 mg. The most common treatment-related adverse events (TRAEs) were pruritus (20.0%), maculopapular rash (18.8%), and decreased appetite (12.5%). Grade 3 TRAEs occurred in 23 patients (28.8%), and Grade 4 TRAEs occurred in 2 patients. Eight patients (500 mg, n=2; 1200 mg, n=6) discontinued treatment due to TRAEs. No treatment-related deaths occurred.

[0074] **Table 2:** Observed response rate in 2L NSCLC patients treated with either 500 mg or 1200 mg of anti-PD-L1/TGFβ Trap once every 2 weeks

ORR	500 mg	1200 mg	Total
All, n, %	8/40, 20.0	11/40, 27.5	19/80, 23.8
PD-L1+ ($\geq 1\%$) pts, n, %	6/31, 19.4	11/27, 40.7	17/58, 29.3
PD-L1 high ($\geq 80\%$) pts, n, %	2/6, 33.3	6/7, 85.7	8/13, 62.0

[0075] These results demonstrate that anti-PD-L1/TGF β Trap monotherapy was well tolerated and showed efficacy across PD-L1 subgroups, with an ORR at 1200 mg of 37.0% and 85.7% in PD-L1+ and PD-L1-high patients, respectively. Given the response rates significantly improved at higher PD-L1 tumor cell expression (*e.g.*, patients treated at 1200 mg), this promising activity of anti-PD-L1/TGF β Trap observed as a 2L treatment is expected to translate or increase as a first line (1L) therapy in treatment naïve PD-L1-high or PD-L1-independent NSCLC patients.

Establishing Dosing Regimen with Various Dosing Frequencies

[0076] Data regimens with various dosing frequencies have been created to allow less frequent administration and/or to allow coordination of dosing schedules with concomitant medications. Specifically, the preliminary population PK modeling and simulation methodology described above has been used to simulate exposures for various dosing regimens and to compare regimens based on exposure.

[0077] Based on these simulations, a flat dose of at least 500 mg administered once every two weeks is required to maintain an average concentration of about 100 $\mu\text{g/mL}$ for a typical subject, while a flat dose of about 1200 mg administered once every two weeks is required to maintain a C_{trough} of about 100 $\mu\text{g/mL}$.

[0078] Based on simulations for C_{avg} , 1200 mg once every two weeks is equivalent to 1800 mg once every three weeks, while for C_{trough} , 1200 mg once every two weeks is equivalent to 2400 mg once every three weeks. And for C_{avg} , 500 mg once every two weeks is equivalent to 750 mg once every three weeks; for C_{trough} 500 mg once every two weeks is equivalent to 1,167 mg once every three weeks.

[0079] For concurrent administration of anti-PD-L1/TGF β Trap with systemic chemotherapies, which are frequently administered on a once every three weeks schedule, 2400 mg once every three weeks of anti-PD-L1/TGF β Trap is selected as a phase Ib/II dose. For the selection of once every three weeks dose, $C_{\text{trough,ss}}$ and average concentration over the dosing interval at steady-state should be similar or higher to that achieved with 1200 mg once every two week dosing, and most patients should have $C_{\text{trough,ss}}$ above the target concentration of 50 $\mu\text{g/mL}$. The median steady state concentration over the dosing interval with 2400 mg once every three weeks dosing is expected to be approximately 328 $\mu\text{g/mL}$. The median steady state concentration

over the dosing interval with 1200 mg once every two weeks dosing is expected to be approximately 246 µg/mL.

TGFβ as a Cancer Target

[0080] The current disclosure permits localized reduction in TGFβ in a tumor
5 microenvironment by capturing the TGFβ using a soluble cytokine receptor (TGFβRII) tethered to an antibody moiety targeting a cellular immune checkpoint receptor found on the exterior surface of certain tumor cells or immune cells. An example of an antibody moiety of the disclosure to an immune checkpoint protein is anti-PD-L1. The bifunctional molecule of the present disclosure, sometimes referred to herein as an “antibody-cytokine Trap,” is effective
10 precisely because the anti-receptor antibody and cytokine Trap are physically linked. The resulting advantage (over, for example, administration of the antibody and the receptor as separate molecules) is partly because cytokines function predominantly in the local environment through autocrine and paracrine functions. The antibody moiety directs the cytokine Trap to the tumor microenvironment where it can be most effective, by neutralizing the local
15 immunosuppressive autocrine or paracrine effects. Furthermore, in cases where the target of the antibody is internalized upon antibody binding, an effective mechanism for clearance of the cytokine/cytokine receptor complex is provided. Antibody-mediated target internalization was shown for PD-L1, and anti-PD-L1/TGFβ Trap was shown to have a similar internalization rate as anti-PD-L1. This is a distinct advantage over using an anti-TGFβ antibody because first, an
20 anti-TGFβ antibody might not be completely neutralizing; and second, the antibody can act as a carrier extending the half-life of the cytokine.

[0081] Indeed, as described below, treatment with the anti-PD-L1/TGFβ Trap elicits a synergistic anti-tumor effect due to the simultaneous blockade of the interaction between PD-L1 on tumor cells and PD-1 on immune cells, and the neutralization of TGFβ in the tumor
25 microenvironment. Without being bound by theory, this presumably is due to a synergistic effect obtained from simultaneous blocking the two major immune escape mechanisms, and in addition, the depletion of the TGFβ in the tumor microenvironment by a single molecular entity. This depletion is achieved by (1) anti-PD-L1 targeting of tumor cells; (2) binding of the TGFβ autocrine/paracrine in the tumor microenvironment by the TGFβ Trap; and (3) destruction of the
30 bound TGFβ through the PD-L1 receptor-mediated endocytosis. Furthermore, the TGFβRII

fused to the C-terminus of Fc (fragment of crystallization of IgG) was several-fold more potent than the TGF β RII-Fc that places the TGF β RII at the N-terminus of Fc.

[0082] TGF β had been a somewhat questionable target in cancer immunotherapy because of its paradoxical roles as the molecular Jekyll and Hyde of cancer (Bierie et al., *Nat. Rev. Cancer*, 2006; 6:506-20). Like some other cytokines, TGF β activity is developmental stage and context dependent. Indeed TGF β can act as either a tumor promoter or a tumor suppressor, affecting tumor initiation, progression and metastasis. The mechanisms underlying this dual role of TGF β remain unclear (Yang et al., *Trends Immunol.* 2010; 31:220–227). Although it has been postulated that Smad-dependent signaling mediates the growth inhibition of TGF β signaling, while the Smad independent pathways contribute to its tumor-promoting effect, there are also data showing that the Smad-dependent pathways are involved in tumor progression (Yang et al., *Cancer Res.* 2008; 68:9107-11).

[0083] Both the TGF β ligand and the receptor have been studied intensively as therapeutic targets. There are three ligand isoforms, TGF β 1, 2 and 3, all of which exist as homodimers. There are also three TGF β receptors (TGF β R), which are called TGF β R type I, II and III (López-Casillas et al., *J. Cell Biol.* 1994; 124:557-68). TGF β RI is the signaling chain and cannot bind ligand. TGF β RII binds the ligand TGF β 1 and 3, but not TGF β 2, with high affinity. The TGF β RII/TGF β complex recruits TGF β RI to form the signaling complex (Won et al., *Cancer Res.* 1999; 59:1273-7). TGF β RIII is a positive regulator of TGF β binding to its signaling receptors and binds all 3 TGF β isoforms with high affinity. On the cell surface, the TGF β /TGF β RIII complex binds TGF β RII and then recruits TGF β RI, which displaces TGF β RIII to form the signaling complex.

[0084] Although the three different TGF β isoforms all signal through the same receptor, they are known to have differential expression patterns and non-overlapping functions *in vivo*. The three different TGF- β isoform knockout mice have distinct phenotypes, indicating numerous non-compensated functions (Bujak et al., *Cardiovasc. Res.* 2007; 74:184-95). While TGF β 1 null mice have hematopoiesis and vasculogenesis defects and TGF β 3 null mice display pulmonary development and defective palatogenesis, TGF β 2 null mice show various developmental abnormalities, the most prominent being multiple cardiac deformities (Bartram et al., *Circulation* 2001; 103:2745-52; Yamagishi et al., *Anat. Rec.* 2012; 295:257-67). Furthermore, TGF β is implicated to play a major role in the repair of myocardial damage after ischemia and

reperfusion injury. In an adult heart, cardiomyocytes secrete TGF β , which acts as an autocrine to maintain the spontaneous beating rate. Importantly, 70-85% of the TGF β secreted by cardiomyocytes is TGF β 2 (Roberts et al., *J. Clin. Invest.* 1992; 90:2056-62). Despite cardiotoxicity concerns raised by treatment with TGF β RI kinase inhibitors, the present applicant
5 has observed a lack of toxicity, including cardiotoxicity, for anti-PD-L1/TGF β Trap in monkeys.

[0085] Therapeutic approaches to neutralize TGF β include using the extracellular domains of TGF β receptors as soluble receptor Traps and neutralizing antibodies. Of the receptor Trap approach, soluble TGF β RIII may seem the obvious choice since it binds all the three TGF β ligands. However, TGF β RIII, which occurs naturally as a 280-330 kD glucosaminoglycan
10 (GAG)-glycoprotein, with extracellular domain of 762 amino acid residues, is a very complex protein for biotherapeutic development. The soluble TGF β RIII devoid of GAG could be produced in insect cells and has been shown to be a potent TGF β neutralizing agent (Vilchis-Landeros et al., *Biochem. J.*, (2001), 355:215). The two separate binding domains (the endoglin-related and the uromodulin-related) of TGF β RIII could be independently expressed, but they
15 were shown to have affinities 20 to 100 times lower than that of the soluble TGF β RIII, and much diminished neutralizing activity (Mendoza et al., *Biochemistry* 2009; 48:11755-65). On the other hand, the extracellular domain of TGF β RII is only 136 amino acid residues in length and can be produced as a glycosylated protein of 25-35 kD. The recombinant soluble TGF β RII was further shown to bind TGF β 1 with a K_D of 200 pM, which is fairly similar to the K_D of 50 pM for the
20 full length TGF β RII on cells (Lin et al., *J. Biol. Chem.* 1995; 270:2747-54). Soluble TGF β RII-Fc was tested as an anti-cancer agent and was shown to inhibit established murine malignant mesothelioma growth in a tumor model (Suzuki et al., *Clin. Cancer Res.*, 2004; 10:5907-18). Because TGF β RII does not bind TGF β 2, and TGF β RIII binds TGF β 1 and 3 with lower affinity than TGF β RII, a fusion protein of the endoglin domain of TGF β RIII and extracellular domain of
25 TGF β RII was produced in bacteria and was shown to inhibit the signaling of TGF β 1 and 2 in cell based assays more effectively than either TGF β RII or RIII (Verona et al., *Protein Eng'g. Des. Sel.* 2008; 21:463-73).

[0086] Still another approach to neutralize all three isoforms of the TGF β ligands is to screen for a pan-neutralizing anti-TGF β antibody, or an anti-receptor antibody that blocks the receptor
30 from binding to TGF β 1, 2 and 3. GC1008, a human antibody specific for all isoforms of TGF β , was in a Phase I/II study in patients with advanced malignant melanoma or renal cell carcinoma

(Morris et al., *J. Clin. Oncol.* 2008; 26:9028 (Meeting abstract)). Although the treatment was found to be safe and well tolerated, only limited clinical efficacy was observed, and hence it was difficult to interpret the importance of anti-TGF β therapy without further characterization of the immunological effects (Flavell et al., *Nat. Rev. Immunol.* 2010; 10:554-67). There were also

5 TGF β -isoform-specific antibodies tested in the clinic. Metelimumab, an antibody specific for TGF β 1, was tested in Phase 2 clinical trial as a treatment to prevent excessive post-operative scarring for glaucoma surgery; and Lerdelimumab, an antibody specific for TGF β 2, was found to be safe but ineffective at improving scarring after eye surgery in a Phase 3 study (Khaw et al., *Ophthalmology* 2007; 114:1822–1830). Anti-TGF β RII antibodies that block the receptor from

10 binding to all the three TGF β isoforms, such as the anti-human TGF β RII antibody TR1 and anti-mouse TGF β RII antibody MT1, have also shown some therapeutic efficacy against primary tumor growth and metastasis in mouse models (Zhong et al., *Clin. Cancer Res.* 2010; 16:1191-205). However, in a recent Phase I study of antibody TR1 (LY3022859), dose escalation beyond 25 mg (flat dose) was considered unsafe due to uncontrolled cytokine release, despite

15 prophylactic treatment (Tolcher et al., *Cancer Chemother. Pharmacol.* 2017; 79:673-680). To date, the vast majority of the studies on TGF β targeted anticancer treatment, including small molecule inhibitors of TGF β signaling that often are quite toxic, are mostly in the preclinical stage and the anti-tumor efficacy obtained has been limited (Calone et al., *Exp Oncol.* 2012; 34:9-16; Connolly et al., *Int. J. Biol. Sci.* 2012; 8:964-78).

20 **[0087]** The antibody-TGF β Trap of the disclosure is a bifunctional protein containing at least portion of a human TGF β Receptor II (TGF β RII) that is capable of binding TGF β . In certain embodiments, the TGF β Trap polypeptide is a soluble portion of the human TGF β Receptor Type 2 Isoform A (SEQ ID NO: 8) that is capable of binding TGF β . In certain embodiments, TGF β Trap polypeptide contains at least amino acids 73-184 of SEQ ID NO: 8. In certain

25 embodiments, the TGF β Trap polypeptide contains amino acids 24-184 of SEQ ID NO: 8. In certain embodiments, the TGF β Trap polypeptide is a soluble portion of the human TGF β Receptor Type 2 Isoform B (SEQ ID NO: 9) that is capable of binding TGF β . In certain embodiments, TGF β Trap polypeptide contains at least amino acids 48-159 of SEQ ID NO: 9. In certain embodiments, the TGF β Trap polypeptide contains amino acids 24-159 of SEQ ID

30 NO: 9. In certain embodiments, the TGF β Trap polypeptide contains amino acids 24-105 of SEQ ID NO: 9. In certain exemplary embodiments, the TGF β Trap polypeptide contains the sequence of SEQ ID NOs: 10, 50, 51, 52, 53, or 54.

[0088] In another embodiment, the antibody-TGF β Trap of the disclosure is one of the fusion proteins disclosed in WO 2018/205985. In some embodiments, the fusion protein is one of the constructs listed in Table 2 of this publication, such as construct 9 or 15 thereof. In other embodiments, the antibody having the heavy chain sequence of SEQ ID NO: 11 and the light chain sequence of SEQ ID NO: 12 of this publication [corresponding to SEQ ID NO: 61 and 62, respectively, of the present disclosure] is fused via a linking sequence (G₄S)_xG, wherein x is 4-5, to the TGF β RII extracellular domain sequence of SEQ ID NO: 14 or SEQ ID NO: 15 of said publication [corresponding to SEQ ID NO: 50 and 51, respectively, of the present disclosure].

Mechanisms of Action

[0089] The approach of targeting T cell inhibition checkpoints for dis-inhibition with therapeutic antibodies is an area of intense investigation (for a review, *see* Pardoll, *Nat. Rev. Cancer* 2012; 12:253-264). In one approach, the antibody moiety or antigen binding fragment thereof targets T cell inhibition checkpoint receptor proteins on the T cell, such as, for example: CTLA-4, PD-1, BTLA, LAG-3, TIM-3, or LAIR1. In another approach, the antibody moiety targets the counter-receptors on antigen presenting cells and tumor cells (which co-opt some of these counter-receptors for their own immune evasion), such as for example: PD-L1 (B7-H1), B7-DC, HVEM, TIM-4, B7-H3, or B7-H4.

[0090] The disclosure contemplates antibody TGF β Traps that target, through their antibody moiety or antigen binding fragment thereof, T cell inhibition checkpoints for dis-inhibition. To that end the applicants have tested the anti-tumor efficacy of combining a TGF β Trap with antibodies targeting various T cell inhibition checkpoint receptor proteins, such as anti-PD-1, anti-PD-L1, anti-TIM-3 and anti-LAG3.

[0091] The programmed death 1 (PD-1)/PD-L1 axis is an important mechanism for tumor immune evasion. Effector T cells chronically sensing antigen take on an exhausted phenotype marked by PD-1 expression, a state under which tumor cells engage by upregulating PD-L1. Additionally, in the tumor microenvironment, myeloid cells, macrophages, parenchymal cells and T cells upregulate PD-L1. Blocking the axis restores the effector function in these T cells. Anti-PD-L1/TGF β Trap also binds TGF β (1, 2, and 3 isoforms), which is an inhibitory cytokine produced in the tumor microenvironment by cells including apoptotic neutrophils, myeloid-derived suppressor cells, T cells and tumor. Inhibition of TGF β by soluble TGF β RII reduced

malignant mesothelioma in a manner that was associated with increases in CD8⁺ T cell anti-tumor effects. The absence of TGF β 1 produced by activated CD4⁺ T cells and Treg cells has been shown to inhibit tumor growth, and protect mice from spontaneous cancer. Thus, TGF β appears to be important for tumor immune evasion.

5 [0092] TGF β has growth inhibitory effects on normal epithelial cells, functioning as a regulator of epithelial cell homeostasis, and it acts as a tumor suppressor during early carcinogenesis. As tumors progress toward malignancy, the growth inhibitory effects of TGF β on the tumor are lost via mutation in one or more TGF β pathway signaling components or through oncogenic reprogramming. Upon loss of sensitivity to TGF β inhibition, the tumor
10 continues to produce high levels of TGF β , which then serve to promote tumor growth. The TGF β cytokine is overexpressed in various cancer types with correlation to tumor stage. Many types of cells in the tumor microenvironment produce TGF β including the tumor cells themselves, immature myeloid cells, regulatory T cells, and stromal fibroblasts; these cells collectively generate a large reservoir of TGF β in the extracellular matrix. TGF β signaling
15 contributes to tumor progression by promoting metastasis, stimulating angiogenesis, and suppressing innate and adaptive anti-tumor immunity. As a broadly immunosuppressive factor, TGF β directly down-regulates the effector function of activated cytotoxic T cells and NK cells and potently induces the differentiation of naïve CD4⁺ T cells to the immunosuppressive regulatory T cells (Treg) phenotype. In addition, TGF β polarizes macrophages and neutrophils to
20 a wound-healing phenotype that is associated with production of immunosuppressive cytokines. As a therapeutic strategy, neutralization of TGF β activity has the potential to control tumor growth by restoring effective anti-tumor immunity, blocking metastasis, and inhibiting angiogenesis.

[0093] The present disclosure provides dosage regimens for targeted TGF- β inhibition with
25 an anti-PD-L1/TGF β Trap molecule in combination with standard chemotherapeutic agents for use in a method of treating a treatment naïve subject diagnosed with advanced NSCLC, or a PDx failure metastatic NSCLC subject. The advanced NSCLC being treated can be squamous or non-squamous NSCLC and is independent of baseline PD-L1 expression levels.

[0094] Concomitant PD-1 and TGF β blockade can restore pro-inflammatory cytokines. Anti-
30 PD-L1/TGF β Trap includes, for example, an extracellular domain of the human TGF β receptor TGF β RII covalently joined via a glycine/serine linker to the C-terminus of each heavy chain of

the fully human IgG1 anti-PD-L1 antibody. Given the emerging picture for the anti-PD-1/PD-L1 class, in which responses are apparent but with room for increase in effect size, it is assumed that co-targeting a complementary immune modulation step will improve tumor response. A similar TGF-targeting agent, fresolimumab, which is a monoclonal antibody targeting TGF β 1, 2 and 3, showed initial evidence of tumor response in a Phase I trial in subjects with melanoma.

[0095] The present disclosure provides experiments that demonstrated that the TGF β RII portion of anti-PD-L1/TGF β Trap (the Trap control “anti-PDL-1(mut)/ TGF β Trap”) elicited antitumor activity. For example, following subcutaneous implantation in a Detroit 562 human pharyngeal carcinoma model, anti-PDL1(mut)/ TGF β Trap elicited a dose-dependent reduction in tumor volume when administered at 25 μ g, 76 μ g, or 228 μ g (**FIG. 5**).

[0096] The present disclosure provides experiments that demonstrated that the protein of the present disclosure simultaneously bound to both PD-L1 and TGF β (**FIG. 2**).

[0097] The present disclosure provides experiments that demonstrated that the protein of the present disclosure (*e.g.* anti-PD-L1/TGF β Trap) inhibited PD-L1 and TGF β dependent signaling *in vitro*. The present disclosure provides experiments that demonstrated that the protein of the present disclosure enhanced T cell effector function *in vitro* via blockade of PD-L1-mediated immune inhibition as measured by an IL-2 induction assay following superantigen stimulation (**FIG. 3**). At approximately 100 ng/ml, the protein of the present disclosure induced a dramatic increase in IL-2 levels *in vitro* (**FIG. 3**).

[0098] The present disclosure provides experiments that demonstrated that the protein of the present disclosure (*e.g.* anti-PD-L1/TGF β Trap) caused depletion of TGF β from blood *in vivo*. Treatment of orthotopically implanted EMT-6 breast cancer cells in JH mice with 55 μ g, or 164 μ g, or 492 μ g of the protein of the present disclosure resulted in efficient and specific depletion of TGF β 1 (**FIG. 4A**), TGF β 2 (**FIG. 4B**), and TGF β 3 (**FIG. 4C**). Furthermore, the present disclosure provides experiments that demonstrated that the protein of the present disclosure occupied the PD-L1 target, supporting the notion that that the protein of the present disclosure fit to a receptor binding model in the EMT-6 tumor system (**FIG. 4D**).

[0099] The present disclosure provides experiments that demonstrated that the protein of the present disclosure efficiently, specifically, and simultaneously bound to PD-L1 and TGF β , possessed potent antitumor activity in a variety of mouse models, suppressed tumor growth and

metastasis, as well as extended survival (e.g., survival of up to and including 6 months, 12 months, 18 months, 22 months, 28 months, 32 months, 38 months, 44 months, 50 months, 56 months, 62 months, 68 months, 74 months, 80 months, 86 months, 92 months, 98 months, 104 months, or 110 months) and conferred long-term protective antitumor immunity. In certain
 5 embodiments, extended survival is at least 108 months.

Anti-PD-L1 Antibodies

[0100] The anti-PD-L1/TGF β Trap molecule of the present disclosure can include any anti-PD-L1 antibody, or antigen-binding fragment thereof, described in the art. Anti-PD-L1 antibodies are commercially available, for example, the 29E2A3 antibody (Biolegend, Cat. No.
 10 329701). Antibodies can be monoclonal, chimeric, humanized, or human. Antibody fragments include Fab, F(ab')₂, scFv and Fv fragments, which are described in further detail below.

[0101] Exemplary antibodies are described in PCT Publication WO 2013/079174. These antibodies can include a heavy chain variable region polypeptide including an HVR-H1, HVR-H2, and HVR-H3 sequence, where:

15 (a) the HVR-H1 sequence is X₁YX₂MX₃ (SEQ ID NO: 21);

(b) the HVR-H2 sequence is SIYPSGGX₄TFYADX₅VKG (SEQ ID NO: 22);

(c) the HVR-H3 sequence is IKLGTVTTVX₆Y (SEQ ID NO: 23);

further where: X₁ is K, R, T, Q, G, A, W, M, I, or S; X₂ is V, R, K, L, M, or I; X₃ is H, T, N, Q, A, V, Y, W, F, or M; X₄ is F or I; X₅ is S or T; X₆ is E or D.

20 [0102] In a one embodiment, X₁ is M, I, or S; X₂ is R, K, L, M, or I; X₃ is F or M; X₄ is F or I; X₅ is S or T; X₆ is E or D.

[0103] In another embodiment X₁ is M, I, or S; X₂ is L, M, or I; X₃ is F or M; X₄ is I; X₅ is S or T; X₆ is D.

[0104] In still another embodiment, X₁ is S; X₂ is I; X₃ is M; X₄ is I; X₅ is T; X₆ is D.

[0105] In another aspect, the polypeptide further includes variable region heavy chain framework sequences juxtaposed between the HVRs according to the formula: (HC-FR1)-(HVR-H1)-(HC-FR2)-(HVR-H2)-(HC-FR3)-(HVR-H3)-(HC-FR4).

[0106] In yet another aspect, the framework sequences are derived from human consensus
5 framework sequences or human germline framework sequences.

[0107] In a still further aspect, at least one of the framework sequences is the following:

HC-FR1 is EVQLLES GGGLVQPGGSLRLSCAASGFTFS (SEQ ID NO: 24);

HC-FR2 is WVRQAPGKGLEWVS (SEQ ID NO: 25);

HC-FR3 is RFTISRDN SKNTLYLQMNSLRAEDTAVYYCAR (SEQ ID NO: 26);

10 HC-FR4 is WGQGTLVTVSS (SEQ ID NO: 27).

[0108] In a still further aspect, the heavy chain polypeptide is further combined with a variable region light chain including an HVR-L1, HVR-L2, and HVR-L3, where:

(a) the HVR-L1 sequence is TGT_{X7}X₈DVGX₉YNYVS (SEQ ID NO: 28);

(b) the HVR-L2 sequence is X₁₀VX₁₁X₁₂RPS (SEQ ID NO: 29);

15 (c) the HVR-L3 sequence is SSX₁₃TX₁₄X₁₅X₁₆X₁₇RV (SEQ ID NO: 30);

further where: X₇ is N or S; X₈ is T, R, or S; X₉ is A or G; X₁₀ is E or D; X₁₁ is I, N or S; X₁₂ is D, H or N; X₁₃ is F or Y; X₁₄ is N or S; X₁₅ is R, T or S; X₁₆ is G or S; X₁₇ is I or T.

[0109] In another embodiment, X₇ is N or S; X₈ is T, R, or S; X₉ is A or G; X₁₀ is E or D; X₁₁ is N or S; X₁₂ is N; X₁₃ is F or Y; X₁₄ is S; X₁₅ is S; X₁₆ is G or S; X₁₇ is T.

20 [0110] In still another embodiment, X₇ is S; X₈ is S; X₉ is G; X₁₀ is D; X₁₁ is S; X₁₂ is N; X₁₃ is Y; X₁₄ is S; X₁₅ is S; X₁₆ is S; X₁₇ is T.

[0111] In a still further aspect, the light chain further includes variable region light chain framework sequences juxtaposed between the HVRs according to the formula: (LC-FR1MHVR-L1)-(LC-FR2)-(HVR-L2)-(LC-FR3)-(HVR-L3)-(LC-FR4).

[0112] In a still further aspect, the light chain framework sequences are derived from human consensus framework sequences or human germline framework sequences.

[0113] In a still further aspect, the light chain framework sequences are lambda light chain sequences.

5 [0114] In a still further aspect, at least one of the framework sequence is the following:

LC-FR1 is QSALTQPASVSGSPGQSITISC (SEQ ID NO: 31);

LC-FR2 is WYQQHPGKAPKLMYI (SEQ ID NO: 32);

LC-FR3 is GVSNRFGSGKSGNTASLTISGLQAEDEADYYC (SEQ ID NO: 33);

LC-FR4 is FGTGTVTL (SEQ ID NO: 34).

10 [0115] In another embodiment, the disclosure provides an anti-PD-L1 antibody or antigen binding fragment including a heavy chain and a light chain variable region sequence, where:

(a) the heavy chain includes an HVR-H1, HVR-H2, and HVR-H3, wherein further: (i) the HVR-H1 sequence is $X_1YX_2MX_3$ (SEQ ID NO: 21); (ii) the HVR-H2 sequence is SIYPSGGX₄TFYADX₅VKG (SEQ ID NO: 22); (iii) the HVR-H3 sequence is

15 IKLGTVTTVX₆Y (SEQ ID NO: 23), and;

(b) the light chain includes an HVR-L1, HVR-L2, and HVR-L3, wherein further: (iv) the HVR-L1 sequence is TGTX₇X₈DVGX₉YNYVS (SEQ ID NO: 28); (v) the HVR-L2 sequence is X₁₀VX₁₁X₁₂RPS (SEQ ID NO: 29); (vi) the HVR-L3 sequence is SSX₁₃TX₁₄X₁₅X₁₆X₁₇RV (SEQ ID NO: 30); wherein: X₁ is K, R, T, Q, G, A, W, M, I, or S; X₂ is V, R, K, L, M, or I; X₃ is H, T, N, Q, A, V, Y, W, F, or M; X₄ is F or I; X₅ is S or T; X₆ is E or D; X₇ is N or S; X₈ is T, R, or S; X₉ is A or G; X₁₀ is E or D; X₁₁ is I, N, or S; X₁₂ is D, H, or N; X₁₃ is F or Y; X₁₄ is N or S; X₁₅ is R, T, or S; X₁₆ is G or S; X₁₇ is I or T.

20 [0116] In one embodiment, X₁ is M, I, or S; X₂ is R, K, L, M, or I; X₃ is F or M; X₄ is F or I; X₅ is S or T; X₆ is E or D; X₇ is N or S; X₈ is T, R, or S; X₉ is A or G; X₁₀ is E or D; X₁₁ is N or S; X₁₂ is N; X₁₃ is F or Y; X₁₄ is S; X₁₅ is S; X₁₆ is G or S; X₁₇ is T.

[0117] In another embodiment, X₁ is M, I, or S; X₂ is L, M, or I; X₃ is F or M; X₄ is I; X₅ is S or T; X₆ is D; X₇ is N or S; X₈ is T, R, or S; X₉ is A or G; X₁₀ is E or D; X₁₁ is N or S; X₁₂ is N; X₁₃ is F or Y; X₁₄ is S; X₁₅ is S; X₁₆ is G or S; X₁₇ is T.

[0118] In still another embodiment, X₁ is S; X₂ is I; X₃ is M; X₄ is I; X₅ is T; X₆ is D; X₇ is S; X₈ is S; X₉ is G; X₁₀ is D; X₁₁ is S; X₁₂ is N; X₁₃ is Y; X₁₄ is S; X₁₅ is S; X₁₆ is S; X₁₇ is T.

[0119] In a further aspect, the heavy chain variable region includes one or more framework sequences juxtaposed between the HVRs as: (HC-FR1)-(HVR-H1)-(HC-FR2)-(HVR-H2)-(HC-FR3)-(HVR-H3)-(HC-FR4), and the light chain variable regions include one or more framework sequences juxtaposed between the HVRs as: (LC-FR1 MHVR-L1)-(LC-FR2)-(HVR-L2)-(LC-FR3)-(HVR-L3)-(LC-FR4).

[0120] In a still further aspect, the framework sequences are derived from human consensus framework sequences or human germline sequences.

[0121] In a still further aspect, one or more of the heavy chain framework sequences is the following:

15 HC-FR1 is EVQLLES GGGLVQPGGSLRLSCAASGFTFS (SEQ ID NO: 24);

HC-FR2 is WVRQAPGKGLEWVS (SEQ ID NO: 25);

HC-FR3 is RFTISRDN SKNTLYLQMNSLRAEDTAVYYCAR (SEQ ID NO: 26);

HC-FR4 is WGQGTLVTVSS (SEQ ID NO: 27).

[0122] In a still further aspect, the light chain framework sequences are lambda light chain sequences.

[0123] In a still further aspect, one or more of the light chain framework sequences is the following:

LC-FR1 is QSALTQPASVSGSPGQSITISC (SEQ ID NO: 31);

LC-FR2 is WYQQHPGKAPKLMIY (SEQ ID NO: 32);

25 LC-FR3 is GVSNRFS GSKSGNTASLTISGLQAEDEADYYC (SEQ ID NO: 33);

LC-FR4 is FGTGTKVTVL (SEQ ID NO: 34).

[0124] In a still further aspect, the heavy chain variable region polypeptide, antibody, or antibody fragment further includes at least a C_H1 domain.

5 [0125] In a more specific aspect, the heavy chain variable region polypeptide, antibody, or antibody fragment further includes a C_H1, a C_H2, and a C_H3 domain.

[0126] In a still further aspect, the variable region light chain, antibody, or antibody fragment further includes a C_L domain.

[0127] In a still further aspect, the antibody further includes a C_H1, a C_H2, a C_H3, and a C_L domain.

10 [0128] In a still further specific aspect, the antibody further includes a human or murine constant region.

[0129] In a still further aspect, the human constant region is selected from the group consisting of IgG1, IgG2, IgG2, IgG3, IgG4.

[0130] In a still further specific aspect, the human or murine constant region is IgG1.

15 [0131] In yet another embodiment, the disclosure features an anti-PD-L1 antibody including a heavy chain and a light chain variable region sequence, where:

(a) the heavy chain includes an HVR-H1, an HVR-H2, and an HVR-H3, having at least 80% overall sequence identity to SYIMM (SEQ ID NO: 35), SIYPSGGITFYADTVKG (SEQ ID NO: 36), and IKLGTVTTVDY (SEQ ID NO: 37), respectively, and

20 (b) the light chain includes an HVR-L1, an HVR-L2, and an HVR-L3, having at least 80% overall sequence identity to TGTSSDVGGYNYVS (SEQ ID NO: 38), DVSNRPS (SEQ ID NO: 39), and SSYTSSSTRV (SEQ ID NO: 40), respectively.

[0132] In a specific aspect, the sequence identity is 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

25 [0133] In yet another embodiment, the disclosure features an anti-PD-L1 antibody including a heavy chain and a light chain variable region sequence, where:

(a) the heavy chain includes an HVR-H1, an HVR-H2, and an HVR-H3, having at least 80% overall sequence identity to MYMMM (SEQ ID NO: 41), SIYPSGGITFYADSVKG (SEQ ID NO: 42), and IKLGTVTTVDY (SEQ ID NO: 37), respectively, and

(b) the light chain includes an HVR-L1, an HVR-L2, and an HVR-L3, having at least 80% overall sequence identity to TGTSSDVGGYNYVS (SEQ ID NO: 43), DVSNRPS (SEQ ID NO: 39), and SSYTSSSTRV (SEQ ID NO: 40), respectively.

[0134] In a specific aspect, the sequence identity is 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

[0135] In a still further aspect, in the antibody or antibody fragment according to the disclosure, as compared to the sequences of HVR-H1, HVR-H2, and HVR-H3, at least those amino acids remain unchanged that are highlighted by underlining as follows:

(a) in HVR-H1 SYIMM (SEQ ID NO: 35),

(b) in HVR-H2 SIYPSGGITFYADTVKG (SEQ ID NO: 36),

(c) in HVR-H3 IKLGTVTTVDY (SEQ ID NO: 37);

and further where, as compared to the sequences of HVR-L1, HVR-L2, and HVR-L3 at least those amino acids remain unchanged that are highlighted by underlining as follows:

(a) HVR-L1 TGTSSDVGGYNYVS (SEQ ID NO: 38)

(b) HVR-L2 DVSNRPS (SEQ ID NO: 39)

(c) HVR-L3 SSYTSSSTRV (SEQ ID NO: 40).

[0136] In another aspect, the heavy chain variable region includes one or more framework sequences juxtaposed between the HVRs as: (HC-FR1)-(HVR-H1)-(HC-FR2)-(HVR-H2)-(HC-FR3)-(HVR-H3)-(HC-FR4), and the light chain variable regions include one or more framework sequences juxtaposed between the HVRs as: (LC-FR1)-(HVR-L1)-(LC-FR2)-(HVR-L2)-(LC-FR3)-(HVR-L3)-(LC-FR4).

[0137] In yet another aspect, the framework sequences are derived from human germline sequences.

[0138] In a still further aspect, one or more of the heavy chain framework sequences is the following:

HC-FR1 is EVQLLESGGGLVQPGGSLRLSCAASGFTFS (SEQ ID NO: 24);

HC-FR2 is WVRQAPGKGGLEWVS (SEQ ID NO: 25);

5 HC-FR3 is RFTISRDN SKNTLYLQMNSLRAEDTAVYYCAR (SEQ ID NO: 26);

HC-FR4 is WGQGTLVTVSS (SEQ ID NO: 27).

[0139] In a still further aspect, the light chain framework sequences are derived from a lambda light chain sequence.

10 [0140] In a still further aspect, one or more of the light chain framework sequences is the following:

LC-FR1 is QSALTQPASVSGSPGQSITISC (SEQ ID NO: 31);

LC-FR2 is WYQQHPGKAPKLMYI (SEQ ID NO: 32);

LC-FR3 is GVSNRFGSGSKSGNTASLTISGLQAEDEADYYC (SEQ ID NO: 33);

LC-FR4 is FGTGTKVTVL (SEQ ID NO: 34).

15 [0141] In a still further specific aspect, the antibody further includes a human or murine constant region.

[0142] In a still further aspect, the human constant region is selected from the group consisting of IgG1, IgG2, IgG2, IgG3, IgG4.

20 [0143] In certain embodiments, the disclosure features an anti-PD-L1 antibody including a heavy chain and a light chain variable region sequence, where:

(a) the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence:

EVQLLES GGGLVQPGGSLRLSCAASGFTFSSYIMMVWRQAPGKGLEWVSSIYPSGGITF
YADWKGRFTISRDN SKNTLYLQMNSLRAEDTAVYYCARIKLGTVTTVDYWGQGT LVT
VSS (SEQ ID NO: 44), and

(b) the light chain sequence has at least 85% sequence identity to the light chain
5 sequence:

QSALTQPASVSGSPGQSITISCTGTSSDVGGYNYVSWYQQHPGKAPKLMYDVSN
RPSGVSNRFGSGKSGNTASLTISGLQAEDEADYYCSSYTSSSTRVFGTGT KVTVL (SEQ
ID NO: 45).

[0144] In various embodiments, the heavy chain sequence has at least 86% sequence identity
10 to SEQ ID NO: 44 and the light chain sequence has at least 86% sequence identity to SEQ ID
NO: 45; the heavy chain sequence has at least 87% sequence identity to SEQ ID NO: 44 and the
light chain sequence has at least 87% sequence identity to SEQ ID NO: 45; the heavy chain
sequence has at least 88% sequence identity to SEQ ID NO: 44 and the light chain sequence has
at least 88% sequence identity to SEQ ID NO: 45; the heavy chain sequence has at least 89%
15 sequence identity to SEQ ID NO: 44 and the light chain sequence has at least 89% sequence
identity to SEQ ID NO: 45; the heavy chain sequence has at least, 90% sequence identity to SEQ
ID NO: 44 and the light chain sequence has at least 90% sequence identity to SEQ ID NO: 45;
the heavy chain sequence has at least 91% sequence identity to SEQ ID NO: 44 and the light
chain sequence has at least 91% sequence identity to SEQ ID NO: 45; the heavy chain sequence
20 has at least 92% sequence identity to SEQ ID NO: 44 and the light chain sequence has at least
92% sequence identity to SEQ ID NO: 45; the heavy chain sequence has at least 93% sequence
identity to SEQ ID NO: 44 and the light chain sequence has at least 93% sequence identity to
SEQ ID NO: 45; the heavy chain sequence has at least 94% sequence identity to SEQ ID NO: 44
and the light chain sequence has at least 94% sequence identity to SEQ ID NO: 45; the heavy
25 chain sequence has at least 95% sequence identity to SEQ ID NO: 44 and the light chain
sequence has at least 95% sequence identity to SEQ ID NO: 45; the heavy chain sequence has at
least 96% sequence identity to SEQ ID NO: 44 and the light chain sequence has at least 96%
sequence identity to SEQ ID NO: 45; the heavy chain sequence has at least 97% sequence
identity to SEQ ID NO: 44 and the light chain sequence has at least 97% sequence identity to
30 SEQ ID NO: 45; the heavy chain sequence has at least 98% sequence identity to SEQ ID NO: 44
and the light chain sequence has at least 98% sequence identity to SEQ ID NO: 45; the heavy

chain sequence has at least 99% sequence identity to SEQ ID NO: 44 and the light chain sequence has at least 99% sequence identity to SEQ ID NO: 45; or the heavy chain sequence comprises SEQ ID NO: 44 and the light chain sequence comprises SEQ ID NO: 45.

[0145] In certain embodiments, the disclosure provides for an anti-PD-L1 antibody including a heavy chain and a light chain variable region sequence, where:

(a) the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence:

EVQLLES GGGLVQPGGSLRLSCAASGFTFSMYMMMWVRQAPGKGLEVWSSIYPSGGIT
FYADSVKGRFTISRDN SKNTLYLQMNSLRAEDTAIYYCARIKLGT VTTVDYWG

10 QGTLVTVSS (SEQ ID NO: 46), and

(b) the light chain sequence has at least 85% sequence identity to the light chain sequence:

QSALTQPASVSGSPGQSITISCTGTSSDVGAYNYVSWYQQHPGKAPKLMYDVSNR
PSGVSNRFGSKSGNTASLTISGLQAEDEADYYCSSYTSSSTRVFGTGT KVTVL (SEQ ID

15 NO: 47).

[0146] In various embodiments, the heavy chain sequence has at least 86% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 86% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 87% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 87% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 88% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 88% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 89% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 89% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least, 90% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 90% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 91% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 91% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 92% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 92% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 93% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 93% sequence identity to

SEQ ID NO: 47; the heavy chain sequence has at least 94% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 94% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 95% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 95% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 96% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 96% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 97% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 97% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 98% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 98% sequence identity to SEQ ID NO: 47; the heavy chain sequence has at least 99% sequence identity to SEQ ID NO: 46 and the light chain sequence has at least 99% sequence identity to SEQ ID NO: 47; or the heavy chain sequence comprises SEQ ID NO: 46 and the light chain sequence comprises SEQ ID NO: 47.

[0147] In another embodiment the antibody binds to human, mouse, or cynomolgus monkey PD-L1. In a specific aspect the antibody is capable of blocking the interaction between human, mice, or cynomolgus monkey PD-L1 and the respective human, mouse, or cynomolgus monkey PD-1 receptors.

[0148] In another embodiment, the antibody binds to human PD-L1 with a KD of 5×10^{-9} M or less, preferably with a KD of 2×10^{-9} M or less, and even more preferred with a KD of 1×10^{-9} M or less.

[0149] In yet another embodiment, the disclosure relates to an anti-PD-L1 antibody or antigen binding fragment thereof which binds to a functional epitope including residues Y56 and D61 of human PD-L1.

[0150] In a specific aspect, the functional epitope further includes E58, E60, Q66, R113, and M115 of human PD-L1.

[0151] In a more specific aspect, the antibody binds to a conformational epitope, including residues 54-66 and 112-122 of human PD-L1.

[0152] In certain embodiments, the disclosure is related to an anti-PD-L1 antibody, or antigen binding fragment thereof, which cross-competes for binding to PD-L1 with an antibody according to the disclosure as described herein.

[0153] In certain embodiments, the disclosure features proteins and polypeptides including any of the above described anti-PD-L1 antibodies in combination with at least one pharmaceutically acceptable carrier.

5 [0154] In certain embodiments, the disclosure features an isolated nucleic acid encoding a polypeptide, or light chain or a heavy chain variable region sequence of an anti-PD-L1 antibody, or antigen binding fragment thereof, as described herein. In certain embodiments, the disclosure provides for an isolated nucleic acid encoding a light chain or a heavy chain variable region sequence of an anti-PD-L1 antibody, wherein:

10 (a) the heavy chain includes an HVR-H1, an HVR-H2, and an HVR-H3 sequence having at least 80% sequence identity to SYIMM (SEQ ID NO: 35), SIYPSGGITFYADTVKG (SEQ ID NO: 36), and IKLGTVTTVDY (SEQ ID NO: 37), respectively, or

(b) the light chain includes an HVR-L1, an HVR-L2, and an HVR-L3 sequence having at least 80% sequence identity to TGTSSDVGGYNYVS (SEQ ID NO: 38), DVSNRPS (SEQ ID NO: 39), and SSYTSSSTRV (SEQ ID NO: 40), respectively.

15 [0155] In a specific aspect, the sequence identity is 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

[0156] In a further aspect, the nucleic acid sequence for the heavy chain is:

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atggagttgc ctgttaggct gttggtgctg atgttctgga ttcttgctag ctccagcgag      60
gtgcagctgc tggaaatccg cggaggactg gtgcagcctg gcggctccct gagactgtct      120
tgcgccgcct ccggtttcac cttctccagc tacatcatga tgtgggtgcg acaggccctt      180
ggcaagggcc tggaatgggt gtcctccatc taccctcccg gcggcatcac cttctacgcc      240
gacaccgtga agggccgggt caccatctcc cgggacaact ccaagaacac cctgtacctg      300
cagatgaact ccctgcgggc cgaggacacc gccgtgtact actgcgcccc gatcaagctg      360
ggcaccgtga ccaccgtgga ctactggggc cagggcaccc tggtgacagt gtcctccgcc      420
tccaccaagg gcccatcggt cttccccctg gcaccctcct ccaagagcac ctctgggggc      480
acagcggccc tgggctgcct ggtcaaggac tacttccccg aaccgggtgac ggtgtcgtgg      540
aactcaggcg ccctgaccag cggcgtgcac accttccccg ctgtcctaca gtcctcagga      600
ctctactccc tcagcagcgt ggtgaccgtg cctccagca gcttgggcac ccagacctac      660
atctgcaacg tgaatcaca gcccagcaac accaagggtg acaagaaagt tgagcccaaa      720
tcttgtgaca aaactcacac atgcccaccg tgcccagcac ctgaactcct ggggggaccg      780
tcagtcttcc tcttcccccc aaaacccaag gacacctca tgatctcccg gacctctgag      840
gtcacatgcy tgggtggtgga cgtgagccac gaagaccctg aggtcaagtt caactggtag      900
gtggacggcg tggaggtgca taatgccaa gcaaagccgc gggaggagca gtacaacagc      960
acgtaccgtg tggtcagcgt cctcaccgtc ctgcaccagg actggctgaa tggcaaggag     1020
tacaagtgca aggtctccaa caaagccctc ccagcccca tcgagaaaac catctccaaa     1080
gccaaagggc agccccgaga accacagggtg tacaccctgc ccccatcacg ggatgagctg     1140
accaagaacc aggtcagcct gacctgcctg gtcaaaggct tctatcccag cgacatcgcc     1200
gtggagtggg agagcaatgg gcagccggag aacaactaca agaccacgcc tcccgtgctg     1260
gactccgacg gctccttctt cctctatagc aagctcaccg tggacaagag cagggtggcag     1320
caggggaacg tcttctcatg ctccgtgatg catgaggctc tgcacaacca ctacacgcag     1380
aagagcctct ccctgtcccc gggtaaa                                     1407

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(SEQ ID NO: 48)

and the nucleic acid sequence for the light chain is:

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atggagttgc ctgttaggct gttggtgctg atgttctgga ttcttgcctc ctttaagccag      60
tccgccctga cccagcctgc ctccgtgtct ggctcccctg gccagtcctc caccatcagc      120
tgcaccggca cctccagcga cgtgggcggc tacaactacg tgtcctggta tcagcagcac      180
cccggcaagg cccccaagct gatgatctac gacgtgtcca accggccctc cggcgtgtcc      240
aacagattct cgggtccaa gtccggcaac accgcctccc tgaccatcag cggactgcag      300
gcagaggacg aggccgacta ctactgctcc tcctacacct cctccagcac cagagtgttc      360
ggcaccggca caaaagtgc cgtgctgggc cagcccaagg ccaacccaac cgtgacactg      420
ttcccccat cctccgagga actgcaggcc aacaaggcca ccctggctctg cctgatctca      480
gatttctatc caggcgccgt gaccgtggcc tgggaaggctg atggctcccc agtgaaggcc      540
ggcgtggaaa ccaccaagcc ctccaagcag tccaacaaca aatacgccgc ctctctctac      600
ctgtccctga ccccgagca gtggaagtcc caccggctct acagctgccca ggtcacacac      660
gagggtcca cgtggaaaa gaccgtcgcc cccaccgagt gctca                          705

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(SEQ ID NO: 49).

- 5 **[0157]** Further exemplary anti-PD-L1 antibodies that can be used in an anti-PD-L1/TGFβ Trap are described in US patent application publication US 2010/0203056. In one embodiment of the disclosure, the antibody moiety is YW243.55S70. In another embodiment of the disclosure, the antibody moiety is MPDL3289A.

- [0158]** In certain embodiments, the disclosure features an anti-PD-L1 antibody moiety
 10 including a heavy chain and a light chain variable region sequence, where:

(a) the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence:

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EVQLVESGGGLVQPGGSLRLSCAASGFTFSDSWIHWVRQAPGKGLEWVAWISPYGGST
YYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCARRHWPGGFDYWGQGTLVT

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- 15 VSS (SEQ ID NO: 12), and

(b) the light chain sequence has at least 85% sequence identity to the light chain sequence:

DIQMTQSPSSLSASVGDRVTITCRASQDVSTAVAWYQQKPGKAPKLLIYSASFLYSGVPS
RFSGSGSGTDFTLTISSLQPEDFATYYCQQYLYHPATFGGQGTKVEIKR (SEQ ID NO: 13).

[0159] In various embodiments, the heavy chain sequence has at least 86% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 86% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 87% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 87% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 88% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 88% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 89% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 89% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least, 90% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 90% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 91% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 91% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 92% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 92% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 93% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 93% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 94% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 94% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 95% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 95% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 96% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 96% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 97% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 97% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 98% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 98% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 99% sequence identity to SEQ ID NO: 12 and the light chain sequence has at least 99% sequence identity to SEQ ID NO: 13; or the heavy chain sequence comprises SEQ ID NO: 12 and the light chain sequence comprises SEQ ID NO: 13.

[0160] In certain embodiments, the disclosure features an anti-PD-L1 antibody moiety including a heavy chain and a light chain variable region sequence, where:

(a) the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence:

EVQLVESGGGLVQPGGSLRLSCAASGFTFSDSWIHWVRQAPGKGLEWVAVWISPYGGST
YYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCARRHWPGGFDYWGQGTTLVT
5 VSA (SEQ ID NO: 14), and

(b) the light chain sequence has at least 85% sequence identity to the light chain sequence:

DIQMTQSPSSLSASVGDRVTITCRASQDVSTAVAWYQQKPGKAPKLLIYSASFLYSGVPS
RFGSGSGTDFTLTISSLQPEDFATYYCQQYLYHPATFGQGTKVEIKR (SEQ ID NO: 13).

- 10 **[0161]** In various embodiments, the heavy chain sequence has at least 86% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 86% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 87% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 87% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 88% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 88% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 89% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 89% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 90% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 90% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 91% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 91% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 92% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 92% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 93% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 93% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 94% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 94% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 95% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 95% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 96% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 96% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 97% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 97% sequence identity to

SEQ ID NO: 13; the heavy chain sequence has at least 98% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 98% sequence identity to SEQ ID NO: 13; the heavy chain sequence has at least 99% sequence identity to SEQ ID NO: 14 and the light chain sequence has at least 99% sequence identity to SEQ ID NO: 13; or the heavy chain sequence
 5 comprises SEQ ID NO: 14 and the light chain sequence comprises SEQ ID NO: 13.

[0162] Further exemplary anti-PD-L1 antibodies that can be used in an anti-PD-L1/TGFβ Trap are described in US patent application publication US 2018/0334504.

[0163] In certain embodiments, the disclosure features an anti-PD-L1 antibody moiety including a heavy chain and a light chain variable region sequence, where

- 10 (a) the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence:

QVQLQESGPGLVKPSQTLSTCTVSGGSISNDYWTWIRQHPGKGGLEYIGYISYTGSTYYN
 PSLKSRVTISRDTSKNQFSLKLSSVTAADTAVYYCARSGGWLAPFDYWGRGTLVTVSS
 (SEQ ID NO: 55), and

- 15 (b) the light chain sequence has at least 85% sequence identity to the light chain sequence:

DIVMTQSPDSLAVSLGERATINCKSSQSLFYHSNQKHSLAWYQQKPGQPPLLIYGAST
 RESGVPDRFSGSGSGTDFTLTISSLQAEDVAVYYCQQYYGYPYTFGGGGTKVEIK (SEQ ID
 NO: 56).

- 20 **[0164]** In various embodiments, the heavy chain sequence has at least 86% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 86% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 87% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 87% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 88% sequence identity to SEQ ID NO: 55 and the light chain sequence has
 25 at least 88% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 89% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 89% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least, 90% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 90% sequence identity to SEQ ID NO: 56;

the heavy chain sequence has at least 91% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 91% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 92% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 92% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 93% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 93% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 94% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 94% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 95% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 95% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 96% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 96% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 97% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 97% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 98% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 98% sequence identity to SEQ ID NO: 56; the heavy chain sequence has at least 99% sequence identity to SEQ ID NO: 55 and the light chain sequence has at least 99% sequence identity to SEQ ID NO: 56; or the heavy chain sequence comprises SEQ ID NO: 55 and the light chain sequence comprises SEQ ID NO: 56.

[0165] In certain embodiments, the disclosure features an anti-PD-L1 antibody moiety including a heavy chain and a light chain variable region sequence, where

(a) the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence:

QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYWMHWVRQAPGQGLEWMGRIGPNSG
FTSYNEKFKNRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARGGSSYDYFDYWGGQTT
VTVSS (SEQ ID NO: 57), and

(b) the light chain sequence has at least 85% sequence identity to the light chain sequence:

DIVLTQSPASLAVSPGQRATITCRASESVSIHGTHLMHWYQQKPGQPPKLLIYAASNLES
GVPARFSGSGSGTDFTLTINPVEAEDTANYCQQSFEDPLTFGQGTKLEIK(SEQ ID NO:
58).

[0166] In various embodiments, the heavy chain sequence has at least 86% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 86% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 87% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 87% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 88% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 88% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 89% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 89% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 90% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 90% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 91% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 91% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 92% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 92% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 93% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 93% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 94% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 94% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 95% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 95% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 96% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 96% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 97% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 97% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 98% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 98% sequence identity to SEQ ID NO: 58; the heavy chain sequence has at least 99% sequence identity to SEQ ID NO: 57 and the light chain sequence has at least 99% sequence identity to SEQ ID NO: 58; or the heavy chain sequence comprises SEQ ID NO: 57 and the light chain sequence comprises SEQ ID NO: 58.

[0167] In certain embodiments, the disclosure features an anti-PD-L1 antibody moiety including a heavy chain and a light chain sequence, where

(a) the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence:

QVQLQESGPGLVKPSQTLSTCTVSGGSISNDYWTWIRQHPGKGGLEYIGYISYTGSTYYN
 PSLKSRVTISRDTSKNQFSLKLSSVTAADTAVYYCARSGGWLAPFDYWGRGTLVTVSSA
 STKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSG
 LYSLSVVTVPSSSLGTKTYTCNVDPHKPSNTKVDKRVESKYGPPCPPCPAPEAAGGPSVF
 5 LFPPKPKDTLMISRTPEVTCVVDVDSQEDPEVQFNWYVDGVEVHNAKTKPREEQFNST
 YRVVSVLTVLHQDWLNGKEYKCKVSNKGLPSSIEKTISKAKGQPREPQVYTLPPSQEEM
 TKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSRLTVDKSRW
 QEGNVFSCSVMHEALHNHYTQKSLSLGLK (SEQ ID NO: 59), and

(b) the light chain sequence has at least 85% sequence identity to the light chain
 10 sequence:

DIVMTQSPDSLAVSLGERATINCKSSQSLFYHSNQKHS LAWYQQKPGQPPKLLIYGAST
 RESGVDPDRFSGSGSGTDFTLTISSLQAEDVAVYYCQQYYGYPTFGGGTKVEIKRTVAA
 PSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDS
 TYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC (SEQ ID NO: 60).

15 **[0168]** In various embodiments, the heavy chain sequence has at least 86% sequence identity
 to SEQ ID NO: 59 and the light chain sequence has at least 86% sequence identity to SEQ ID
 NO: 60; the heavy chain sequence has at least 87% sequence identity to SEQ ID NO: 59 and the
 light chain sequence has at least 87% sequence identity to SEQ ID NO: 60; the heavy chain
 sequence has at least 88% sequence identity to SEQ ID NO: 59 and the light chain sequence has
 20 at least 88% sequence identity to SEQ ID NO: 60; the heavy chain sequence has at least 89%
 sequence identity to SEQ ID NO: 59 and the light chain sequence has at least 89% sequence
 identity to SEQ ID NO: 60; the heavy chain sequence has at least, 90% sequence identity to SEQ
 ID NO: 59 and the light chain sequence has at least 90% sequence identity to SEQ ID NO: 60;
 the heavy chain sequence has at least 91% sequence identity to SEQ ID NO: 59 and the light
 25 chain sequence has at least 91% sequence identity to SEQ ID NO: 60; the heavy chain sequence
 has at least 92% sequence identity to SEQ ID NO: 59 and the light chain sequence has at least
 92% sequence identity to SEQ ID NO: 60; the heavy chain sequence has at least 93% sequence
 identity to SEQ ID NO: 59 and the light chain sequence has at least 93% sequence identity to
 SEQ ID NO: 60; the heavy chain sequence has at least 94% sequence identity to SEQ ID NO: 59
 30 and the light chain sequence has at least 94% sequence identity to SEQ ID NO: 60; the heavy
 chain sequence has at least 95% sequence identity to SEQ ID NO: 59 and the light chain

sequence has at least 95% sequence identity to SEQ ID NO: 60; the heavy chain sequence has at least 96% sequence identity to SEQ ID NO: 59 and the light chain sequence has at least 96% sequence identity to SEQ ID NO: 60; the heavy chain sequence has at least 97% sequence identity to SEQ ID NO: 59 and the light chain sequence has at least 97% sequence identity to SEQ ID NO: 60; the heavy chain sequence has at least 98% sequence identity to SEQ ID NO: 59 and the light chain sequence has at least 98% sequence identity to SEQ ID NO: 60; the heavy chain sequence has at least 99% sequence identity to SEQ ID NO: 59 and the light chain sequence has at least 99% sequence identity to SEQ ID NO: 60; or the heavy chain sequence comprises SEQ ID NO: 59 and the light chain sequence comprises SEQ ID NO: 60.

10 **[0169]** In certain embodiments, the disclosure features an anti-PD-L1 antibody moiety including a heavy chain and a light chain sequence, where

(a) the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence:

QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYWMHWVRQAPGQGLEWMGRIGPNSG
 15 FTSYNEKFKNRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARGGSSYDYFDYWGGQTT
 VTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPA
 VLQSSGLYSLSSVVTVPSSSLGKTYTCNVDHKPSNTKVDKRVESKYGPPCPPCPAPEAA
 GGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSQEDPEVQFNWYVDGVEVHNAKTKPRE
 EQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKGLPSSIEKTISKAKGQPREPQVYTLT
 20 PSQEEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSRLT
 VDKSRWQEGNVFSCSVMHEALHNHYTQKSLSLGA (SEQ ID NO: 61), and

(b) the light chain sequence has at least 85% sequence identity to the light chain sequence:

DIVLTQSPASLAVSPGQRATITCRASESVSIHGTHLMHWYQQKPGQPPKLLIYAASNLES
 25 GVPARFSGSGSGTDFTLTINPVEAEDTANYYCQQSFEDPLTFGQGTKLEIKRTVAAPSVFI
 FPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSSTYSLS
 STLTLKADYEEKHKVYACEVTHQGLSSPVTKSFNRGEC (SEQ ID NO: 62).

[0170] In various embodiments, the heavy chain sequence has at least 86% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 86% sequence identity to SEQ ID

NO: 62; the heavy chain sequence has at least 87% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 87% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 88% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 88% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 89% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 89% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least, 90% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 90% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 91% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 91% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 92% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 92% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 93% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 93% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 94% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 94% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 95% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 95% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 96% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 96% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 97% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 97% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 98% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 98% sequence identity to SEQ ID NO: 62; the heavy chain sequence has at least 99% sequence identity to SEQ ID NO: 61 and the light chain sequence has at least 99% sequence identity to SEQ ID NO: 62; or the heavy chain sequence comprises SEQ ID NO: 61 and the light chain sequence comprises SEQ ID NO: 62.

[0171] Yet further exemplary anti-PD-L1 antibodies that can be used in an anti-PD-L1/TGF β Trap are described in US patent publication US 7,943,743.

[0172] In one embodiment of the disclosure, the anti-PD-L1 antibody is MDX-1105.

[0173] In certain embodiments, the anti-PD-L1 antibody is MEDI-4736.

Constant Region

[0174] The proteins and peptides of the disclosure can include a constant region of an immunoglobulin or a fragment, analog, variant, mutant, or derivative of the constant region. In certain embodiments, the constant region is derived from a human immunoglobulin heavy chain, for example, IgG1, IgG2, IgG3, IgG4, or other classes. In certain embodiments, the constant region includes a CH2 domain. In certain embodiments, the constant region includes CH2 and CH3 domains or includes hinge-CH2-CH3. Alternatively, the constant region can include all or a portion of the hinge region, the CH2 domain and/or the CH3 domain.

[0175] In one embodiment, the constant region contains a mutation that reduces affinity for an Fc receptor or reduces Fc effector function. For example, the constant region can contain a mutation that eliminates the glycosylation site within the constant region of an IgG heavy chain. In some embodiments, the constant region contains mutations, deletions, or insertions at an amino acid position corresponding to Leu234, Leu235, Gly236, Gly237, Asn297, or Pro331 of IgG1 (amino acids are numbered according to EU nomenclature). In a particular embodiment, the constant region contains a mutation at an amino acid position corresponding to Asn297 of IgG1. In alternative embodiments, the constant region contains mutations, deletions, or insertions at an amino acid position corresponding to Leu281, Leu282, Gly283, Gly284, Asn344, or Pro378 of IgG1.

[0176] In some embodiments, the constant region contains a CH2 domain derived from a human IgG2 or IgG4 heavy chain. Preferably, the CH2 domain contains a mutation that eliminates the glycosylation site within the CH2 domain. In one embodiment, the mutation alters the asparagine within the Gln-Phe-Asn-Ser (SEQ ID NO: 15) amino acid sequence within the CH2 domain of the IgG2 or IgG4 heavy chain. Preferably, the mutation changes the asparagine to a glutamine. Alternatively, the mutation alters both the phenylalanine and the asparagine within the Gln-Phe-Asn-Ser (SEQ ID NO: 15) amino acid sequence. In one embodiment, the Gln-Phe-Asn-Ser (SEQ ID NO: 15) amino acid sequence is replaced with a Gln-Ala-Gln-Ser (SEQ ID NO: 16) amino acid sequence. The asparagine within the Gln-Phe-Asn-Ser (SEQ ID NO: 15) amino acid sequence corresponds to Asn297 of IgG1.

[0177] In another embodiment, the constant region includes a CH2 domain and at least a portion of a hinge region. The hinge region can be derived from an immunoglobulin heavy

chain, *e.g.*, IgG1, IgG2, IgG3, IgG4, or other classes. Preferably, the hinge region is derived from human IgG1, IgG2, IgG3, IgG4, or other suitable classes. More preferably the hinge region is derived from a human IgG1 heavy chain. In one embodiment the cysteine in the Pro-Lys-Ser-Cys-Asp-Lys (SEQ ID NO: 17) amino acid sequence of the IgG1 hinge region is altered. In
5 certain embodiments, the Pro-Lys-Ser-Cys-Asp-Lys (SEQ ID NO: 17) amino acid sequence is replaced with a Pro-Lys-Ser-Ser-Asp-Lys (SEQ ID NO: 18) amino acid sequence. In certain embodiments, the constant region includes a CH2 domain derived from a first antibody isotype and a hinge region derived from a second antibody isotype. In certain embodiments, the CH2 domain is derived from a human IgG2 or IgG4 heavy chain, while the hinge region is derived
10 from an altered human IgG1 heavy chain.

[0178] The alteration of amino acids near the junction of the Fc portion and the non-Fc portion can dramatically increase the serum half-life of the Fc fusion protein (PCT publication WO 0158957, the disclosure of which is hereby incorporated by reference). Accordingly, the junction region of a protein or polypeptide of the present disclosure can contain alterations that,
15 relative to the naturally-occurring sequences of an immunoglobulin heavy chain and erythropoietin, preferably lie within about 10 amino acids of the junction point. These amino acid changes can cause an increase in hydrophobicity. In one embodiment, the constant region is derived from an IgG sequence in which the C-terminal lysine residue is replaced. Preferably, the C-terminal lysine of an IgG sequence is replaced with a non-lysine amino acid, such as alanine
20 or leucine, to further increase serum half-life. In another embodiment, the constant region is derived from an IgG sequence in which the Leu-Ser-Leu-Ser (SEQ ID NO: 19) amino acid sequence near the C-terminus of the constant region is altered to eliminate potential junctional T-cell epitopes. For example, in one embodiment, the Leu-Ser-Leu-Ser (SEQ ID NO: 19) amino acid sequence is replaced with an Ala-Thr-Ala-Thr (SEQ ID NO: 20) amino acid sequence. In
25 other embodiments, the amino acids within the Leu-Ser-Leu-Ser (SEQ ID NO: 19) segment are replaced with other amino acids such as glycine or proline. Detailed methods of generating amino acid substitutions of the Leu-Ser-Leu-Ser (SEQ ID NO: 19) segment near the C-terminus of an IgG1, IgG2, IgG3, IgG4, or other immunoglobulin class molecule have been described in U.S. Patent Publication No. 20030166877, the disclosure of which is hereby incorporated by
30 reference.

[0179] Suitable hinge regions for the present disclosure can be derived from IgG1, IgG2, IgG3, IgG4, and other immunoglobulin classes. The IgG1 hinge region has three cysteines, two of which are involved in disulfide bonds between the two heavy chains of the immunoglobulin. These same cysteines permit efficient and consistent disulfide bonding formation between Fc portions. Therefore, a hinge region of the present disclosure is derived from IgG1, *e.g.*, human IgG1. In some embodiments, the first cysteine within the human IgG1 hinge region is mutated to another amino acid, preferably serine. The IgG2 isotype hinge region has four disulfide bonds that tend to promote oligomerization and possibly incorrect disulfide bonding during secretion in recombinant systems. A suitable hinge region can be derived from an IgG2 hinge; the first two cysteines are each preferably mutated to another amino acid. The hinge region of IgG4 is known to form interchain disulfide bonds inefficiently. However, a suitable hinge region for the present disclosure can be derived from the IgG4 hinge region, preferably containing a mutation that enhances correct formation of disulfide bonds between heavy chain-derived moieties (Angal S., et al. (1993) *Mol. Immunol.*, 30:105-8).

[0180] In accordance with the present disclosure, the constant region can contain CH2 and/or CH3 domains and a hinge region that are derived from different antibody isotypes, *e.g.*, a hybrid constant region. For example, in one embodiment, the constant region contains CH2 and/or CH3 domains derived from IgG2 or IgG4 and a mutant hinge region derived from IgG1. Alternatively, a mutant hinge region from another IgG subclass is used in a hybrid constant region. For example, a mutant form of the IgG4 hinge that allows efficient disulfide bonding between the two heavy chains can be used. A mutant hinge can also be derived from an IgG2 hinge in which the first two cysteines are each mutated to another amino acid. Assembly of such hybrid constant regions has been described in U.S. Patent Publication No. 20030044423, the disclosure of which is hereby incorporated by reference.

[0181] In accordance with the present disclosure, the constant region can contain one or more mutations described herein. The combinations of mutations in the Fc portion can have additive or synergistic effects on the prolonged serum half-life and increased *in vivo* potency of the bifunctional molecule. Thus, in one exemplary embodiment, the constant region can contain (i) a region derived from an IgG sequence in which the Leu-Ser-Leu-Ser (SEQ ID NO: 19) amino acid sequence is replaced with an Ala-Thr-Ala-Thr (SEQ ID NO: 20) amino acid sequence; (ii) a C-terminal alanine residue instead of lysine; (iii) a CH2 domain and a hinge region that are

derived from different antibody isotypes, for example, an IgG2 CH2 domain and an altered IgG1 hinge region; and (iv) a mutation that eliminates the glycosylation site within the IgG2-derived CH2 domain, for example, a Gln-Ala-Gln-Ser (SEQ ID NO: 16) amino acid sequence instead of the Gln-Phe-Asn-Ser (SEQ ID NO: 15) amino acid sequence within the IgG2-derived CH2 domain.

Antibody fragments

[0182] The proteins and polypeptides of the disclosure can also include antigen-binding fragments of antibodies. Exemplary antibody fragments include scFv, Fv, Fab, F(ab')₂, and single domain VHH fragments such as those of camelid origin.

[0183] Single-chain antibody fragments, also known as single-chain antibodies (scFvs), are recombinant polypeptides which typically bind antigens or receptors; these fragments contain at least one fragment of an antibody variable heavy-chain amino acid sequence (V_H) tethered to at least one fragment of an antibody variable light-chain sequence (V_L) with or without one or more interconnecting linkers. Such a linker may be a short, flexible peptide selected to assure that the proper three-dimensional folding of the V_L and V_H domains occurs once they are linked so as to maintain the target molecule binding-specificity of the whole antibody from which the single-chain antibody fragment is derived. Generally, the carboxyl terminus of the V_L or V_H sequence is covalently linked by such a peptide linker to the amino acid terminus of a complementary V_L and V_H sequence. Single-chain antibody fragments can be generated by molecular cloning, antibody phage display library or similar techniques. These proteins can be produced either in eukaryotic cells or prokaryotic cells, including bacteria.

[0184] Single-chain antibody fragments contain amino acid sequences having at least one of the variable regions or CDRs of the whole antibodies described in this specification, but are lacking some or all of the constant domains of those antibodies. These constant domains are not necessary for antigen binding, but constitute a major portion of the structure of whole antibodies. Single-chain antibody fragments may therefore overcome some of the problems associated with the use of antibodies containing part or all of a constant domain. For example, single-chain antibody fragments tend to be free of undesired interactions between biological molecules and the heavy-chain constant region, or other unwanted biological activity. Additionally, single-chain antibody fragments are considerably smaller than whole antibodies and may therefore have

greater capillary permeability than whole antibodies, allowing single-chain antibody fragments to localize and bind to target antigen-binding sites more efficiently. Also, antibody fragments can be produced on a relatively large scale in prokaryotic cells, thus facilitating their production. Furthermore, the relatively small size of single-chain antibody fragments makes them less likely than whole antibodies to provoke an immune response in a recipient.

[0185] Fragments of antibodies that have the same or comparable binding characteristics to those of the whole antibody may also be present. Such fragments may contain one or both Fab fragments or the F(ab')₂ fragment. The antibody fragments may contain all six CDRs of the whole antibody, although fragments containing fewer than all of such regions, such as three, four or five CDRs, are also functional.

Pharmaceutical Compositions

[0186] The present disclosure also features pharmaceutical compositions that contain a therapeutically effective amount of a protein described herein. The composition can be formulated for use in a variety of drug delivery systems. One or more physiologically acceptable excipients or carriers can also be included in the composition for proper formulation. Suitable formulations for use in the present disclosure are found in Remington's Pharmaceutical Sciences, Mack Publishing Company, Philadelphia, Pa., 17th ed., 1985. For a brief review of methods for drug delivery, *see, e.g.*, Langer (Science 249:1527-1533, 1990).

[0187] In one aspect, the present disclosure provides an intravenous drug delivery formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject that includes 500 mg – 2400 mg of a protein including a first polypeptide and a second polypeptide, the first polypeptide includes: (a) at least a variable region of a heavy chain of an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGF β RII), or a fragment thereof, capable of binding Transforming Growth Factor β (TGF β), the second polypeptide includes at least a variable region of a light chain of an antibody that binds PD-L1, and the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1.

[0188] In certain embodiments, a protein product of the present disclosure includes a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide

that includes the amino acid sequence of SEQ ID NO: 1. In certain embodiments, a protein product of the present disclosure includes a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40.

5 [0189] In certain embodiments of the present disclosure, the intravenous drug delivery formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject may include an about 1200 mg to about 2400 mg dose (*e.g.*, about 1200 mg to about 2300 mg, about 1200 mg to about 2200 mg, about 1200 mg to about 2100 mg, about 1200 mg to about 2000 mg, about 1200 mg to about 1900 mg, about 1200 mg to about 1800 mg, about 1200 mg to about 1700 mg, about 1200 mg to about 1600 mg, about 1200 mg to about 1500 mg, about 1200 mg to about 1400 mg, about 1200 mg to about 1300 mg, about 1300 mg to 2400 mg, about 1400 mg to 2400 mg, about 1500 mg to 2400 mg, about 1600 mg to 2400 mg, about 1700 mg to 2400 mg, about 1800 mg to 2400 mg, about 1900 mg to 2400 mg, about 2000 mg to 2400 mg, about 2100 mg to 2400 mg, about 2200 mg to 2400 mg, or about 2300 mg to 2400 mg) of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1)). In certain embodiments, the intravenous drug delivery formulation may include an about 2100 to about 2000 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1)). In certain embodiments, the intravenous drug delivery formulation may include an about 2100 mg dose of a protein product of the present disclosure with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1. In certain embodiments, the intravenous drug delivery formulation may include a 2100 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1)). In certain embodiments, the intravenous drug delivery formulation may include an about 1200 mg dose of a protein product of the present disclosure with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1. In certain embodiments, the intravenous drug delivery formulation may include

a 1200 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1)). In certain embodiments, the intravenous drug delivery formulation may include an about 1800 mg dose of a protein product of the present disclosure with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1. In certain embodiments, the intravenous drug delivery formulation may include a 1800 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1)).

[0190] In certain embodiments, the intravenous drug delivery formulation may include an about 2400 mg dose of a protein product of the present disclosure with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1. In certain embodiments, the intravenous drug delivery formulation may include a 2400 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1)).

[0191] In certain embodiments, the intravenous drug delivery formulation may include a 1800 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide comprising the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide comprising the amino acid sequences of SEQ ID NOs: 38, 39, and 40)). In certain embodiments, the intravenous drug delivery formulation may include a 2100 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide comprising the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide comprising the amino acid sequences of SEQ ID NOs: 38, 39, and 40)). In certain embodiments, the intravenous drug delivery formulation may include a 2400 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide comprising the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide comprising the amino acid sequences of SEQ ID NOs: 38, 39, and 40)).

[0192] In certain embodiments, the intravenous drug delivery formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a

PDx failure metastatic NSCLC subject may include an about 1200 mg to about 3000 mg (*e.g.*, about 1200 mg to about 3000 mg, about 1200 mg to about 2900 mg, about 1200 mg to about 2800 mg, about 1200 mg to about 2700 mg, about 1200 mg to about 2600 mg, about 1200 mg to about 2500 mg, about 1200 mg to about 2400 mg, about 1200 mg to about 2300 mg, about 1200 mg to about 2200 mg, about 1200 mg to about 2100 mg, about 1200 mg to about 2000 mg, about 1200 mg to about 1900 mg, about 1200 mg to about 1800 mg, about 1200 mg to about 1700 mg, about 1200 mg to about 1600 mg, about 1200 mg to about 1500 mg, about 1200 mg to about 1400 mg, about 1200 mg to about 1300 mg, about 1300 mg to about 3000 mg, about 1400 mg to about 3000 mg, about 1500 mg to about 3000 mg, about 1600 mg to about 3000 mg, about 1700 mg to about 3000 mg, about 1800 mg to about 3000 mg, about 1900 mg to about 3000 mg, about 2000 mg to about 3000 mg, about 2100 mg to about 3000 mg, about 2200 mg to about 3000 mg, about 2300 mg to about 3000 mg, about 2400 mg to about 3000 mg, about 2500 mg to about 3000 mg, about 2600 mg to about 3000 mg, about 2700 mg to about 3000 mg, about 2800 mg to about 3000 mg, about 2900 mg to about 3000 mg, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, about 2500 mg, about 2600 mg, about 2700 mg, about 2800 mg, about 2900 mg, or about 3000 mg) of a protein product of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap). In certain embodiments, the intravenous drug delivery formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject may include an about 1200 mg to about 3000 mg (*e.g.*, about 1200 mg to about 3000 mg, about 1200 mg to about 2900 mg, about 1200 mg to about 2800 mg, about 1200 mg to about 2700 mg, about 1200 mg to about 2600 mg, about 1200 mg to about 2500 mg, about 1200 mg to about 2400 mg, about 1200 mg to about 2300 mg, about 1200 mg to about 2200 mg, about 1200 mg to about 2100 mg, about 1200 mg to about 2000 mg, about 1200 mg to about 1900 mg, about 1200 mg to about 1800 mg, about 1200 mg to about 1700 mg, about 1200 mg to about 1600 mg, about 1200 mg to about 1500 mg, about 1200 mg to about 1400 mg, about 1200 mg to about 1300 mg, about 1300 mg to about 3000 mg, about 1400 mg to about 3000 mg, about 1500 mg to about 3000 mg, about 1600 mg to about 3000 mg, about 1700 mg to about 3000 mg, about 1800 mg to about 3000 mg, about 1900 mg to about 3000 mg, about 2000 mg to about 3000 mg, about 2100 mg to about 3000 mg, about 2200 mg to about 3000 mg, about 2300 mg to about 3000 mg, about 2400 mg to about 3000 mg, about 2500 mg to about 3000 mg, about 2600 mg to about 3000 mg, about

2700 mg to about 3000 mg, about 2800 mg to about 3000 mg, about 2900 mg to about 3000 mg, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, about 2500 mg, about 2600 mg, about 2700 mg, about 2800 mg, about 2900 mg, or about 3000 mg) of a protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40.

10 [0193] In certain embodiments, the intravenous drug delivery formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject may include about 1200 mg, about 1225 mg, about 1250 mg, about 1275 mg, about 1300 mg, about 1325 mg, about 1350 mg, about 1375 mg, about 1400 mg, about 1425 mg, about 1450 mg, about 1475 mg, about 1500 mg, about 1525 mg, about 1550 mg, about 1575 mg, about 1600 mg, about 1625 mg, about 1650 mg, about 1675 mg, about 1700 mg, about 1725 mg, about 1750 mg, about 1775 mg, about 1800 mg, about 1825 mg, about 1850 mg, about 1875 mg, about 1900 mg, about 1925 mg, about 1950 mg, about 1975 mg, about 2000 mg, about 2025 mg, about 2050 mg, about 2075 mg, about 2100 mg, about 2125 mg, about 2150 mg, about 2175 mg, about 2200 mg, about 2225 mg, about 2250 mg, about 2275 mg, about 2300 mg, about 2325 mg, about 2350 mg, about 2375 mg, or about 2400 mg of the protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap comprising a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40).

25 [0194] The intravenous drug delivery formulation of the present disclosure for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject may be contained in a bag, a pen, or a syringe. In certain embodiments, the bag may be connected to a channel comprising a tube and/or a needle. In certain embodiments, the formulation may be a lyophilized formulation or a liquid formulation. In certain embodiments, the formulation may freeze-dried (lyophilized) and 30 contained in about 12-60 vials. In certain embodiments, the formulation may be freeze-dried and about 45 mg of the freeze-dried formulation may be contained in one vial. In certain

embodiments, the about 40 mg – about 100 mg of freeze-dried formulation may be contained in one vial. In certain embodiments, freeze dried formulation from 12, 27, or 45 vials are combined to obtain a therapeutic dose of the protein in the intravenous drug formulation. In certain embodiments, the formulation may be a liquid formulation of a protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40, and stored as about 250 mg/vial to about 2000 mg/vial (*e.g.*, about 250 mg/vial to about 2000 mg/vial, about 250 mg/vial to about 1900 mg/vial, about 250 mg/vial to about 1800 mg/vial, about 250 mg/vial to about 1700 mg/vial, about 250 mg/vial to about 1600 mg/vial, about 250 mg/vial to about 1500 mg/vial, about 250 mg/vial to about 1400 mg/vial, about 250 mg/vial to about 1300 mg/vial, about 250 mg/vial to about 1200 mg/vial, about 250 mg/vial to about 1100 mg/vial, about 250 mg/vial to about 1000 mg/vial, about 250 mg/vial to about 900 mg/vial, about 250 mg/vial to about 800 mg/vial, about 250 mg/vial to about 700 mg/vial, about 250 mg/vial to about 600 mg/vial, about 250 mg/vial to about 500 mg/vial, about 250 mg/vial to about 400 mg/vial, about 250 mg/vial to about 300 mg/vial, about 300 mg/vial to about 2000 mg/vial, about 400 mg/vial to about 2000 mg/vial, about 500 mg/vial to about 2000 mg/vial, about 600 mg/vial to about 2000 mg/vial, about 700 mg/vial to about 2000 mg/vial, about 800 mg/vial to about 2000 mg/vial, about 900 mg/vial to about 2000 mg/vial, about 1000 mg/vial to about 2000 mg/vial, about 1100 mg/vial to about 2000 mg/vial, about 1200 mg/vial to about 2000 mg/vial, about 1300 mg/vial to about 2000 mg/vial, about 1400 mg/vial to about 2000 mg/vial, about 1500 mg/vial to about 2000 mg/vial, about 1600 mg/vial to about 2000 mg/vial, about 1700 mg/vial to about 2000 mg/vial, about 1800 mg/vial to about 2000 mg/vial, or about 1900 mg/vial to about 2000 mg/vial). In certain embodiments, the formulation may be a liquid formulation and stored as about 600 mg/vial. In certain embodiments, the formulation may be a liquid formulation and stored as about 1200 mg/vial. In certain embodiments, the formulation may be a liquid formulation and stored as about 1800 mg/vial. In certain embodiments, the formulation may be a liquid formulation and stored as about 2400 mg/vial. In certain embodiments, the formulation may be a liquid formulation and stored as about 250 mg/vial.

[0195] This disclosure provides a liquid aqueous pharmaceutical formulation including a therapeutically effective amount of the protein of the present disclosure (*e.g.*, anti-PD-L1/TGFβ

Trap) in a buffered solution forming a formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject.

[0196] These compositions for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject may be sterilized by conventional sterilization techniques, or may be sterile filtered. The resulting aqueous solutions may be packaged for use as-is, or lyophilized, the lyophilized preparation being combined with a sterile aqueous carrier prior to administration. The pH of the preparations typically will be between 3 and 11, more preferably between 5 and 9 or between 6 and 8, and most preferably between 7 and 8, such as 7 to 7.5. The resulting compositions in solid form may be packaged in multiple single dose units, each containing a fixed amount of the above-mentioned agent or agents. The composition in solid form can also be packaged in a container for a flexible quantity.

[0197] In certain embodiments, the present disclosure provides for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject, a formulation with an extended shelf life including a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1)), in combination with mannitol, citric acid monohydrate, sodium citrate, disodium phosphate dihydrate, sodium dihydrogen phosphate dihydrate, sodium chloride, polysorbate 80, water, and sodium hydroxide.

[0198] In certain embodiments, an aqueous formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject is prepared including a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40) in a pH-buffered solution. The buffer of this invention may have a pH ranging from about 4 to about 8, *e.g.*, from about 4 to about 8, from about 4.5 to about 8, from about 5 to about 8, from about 5.5 to about 8, from about 6 to about 8, from about 6.5 to

about 8, from about 7 to about 8, from about 7.5 to about 8, from about 4 to about 7.5, from about 4.5 to about 7.5, from about 5 to about 7.5, from about 5.5 to about 7.5, from about 6 to about 7.5, from about 6.5 to about 7.5, from about 4 to about 7, from about 4.5 to about 7, from about 5 to about 7, from about 5.5 to about 7, from about 6 to about 7, from about 4 to about 6.5, from about 4.5 to about 6.5, from about 5 to about 6.5, from about 5.5 to about 6.5, from about 4 to about 6.0, from about 4.5 to about 6.0, from about 5 to about 6, or from about 4.8 to about 5.5, or may have a pH of about 5.0 to about 5.2. Ranges intermediate to the above recited pH's are also intended to be part of this disclosure. For example, ranges of values using a combination of any of the above recited values as upper and/or lower limits are intended to be included.

10 Examples of buffers that will control the pH within this range include acetate (*e.g.* sodium acetate), succinate (such as sodium succinate), gluconate, histidine, citrate and other organic acid buffers.

[0199] In certain embodiments, the formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject includes a buffer system which contains citrate and phosphate to maintain the pH in a range of about 4 to about 8. In certain embodiments the pH range may be from about 4.5 to about 6.0, or from about pH 4.8 to about 5.5, or in a pH range of about 5.0 to about 5.2. In certain embodiments, the buffer system includes citric acid monohydrate, sodium citrate, disodium phosphate dihydrate, and/or sodium dihydrogen phosphate dihydrate. In certain embodiments, the buffer system includes about 1.3 mg/ml of citric acid (*e.g.*, 1.305 mg/ml), about 0.3 mg/ml of sodium citrate (*e.g.*, 0.305 mg/ml), about 1.5 mg/ml of disodium phosphate dihydrate (*e.g.*, 1.53 mg/ml), about 0.9 mg/ml of sodium dihydrogen phosphate dihydrate (*e.g.*, 0.86), and about 6.2 mg/ml of sodium chloride (*e.g.*, 6.165 mg/ml). In certain embodiments, the buffer system includes about 1-1.5 mg/ml of citric acid, about 0.25 to about 0.5 mg/ml of sodium citrate, about 1.25 to about 1.75 mg/ml of disodium phosphate dihydrate, about 0.7 to about 1.1 mg/ml of sodium dihydrogen phosphate dihydrate, and 6.0 to 6.4 mg/ml of sodium chloride. In certain embodiments, the pH of the formulation is adjusted with sodium hydroxide.

[0200] A polyol, which acts as a tonicifier and may stabilize the antibody, may also be included in the formulation. The polyol is added to the formulation in an amount which may vary with respect to the desired isotonicity of the formulation. In certain embodiments, the aqueous formulation may be isotonic. The amount of polyol added may also alter with respect to

the molecular weight of the polyol. For example, a lower amount of a monosaccharide (*e.g.* mannitol) may be added, compared to a disaccharide (such as trehalose). In certain embodiments, the polyol which may be used in the formulation as a tonicity agent is mannitol. In certain embodiments, the mannitol concentration may be about 5 to about 20 mg/ml. In certain
5 embodiments, the concentration of mannitol may be about 7.5 to about 15 mg/ml. In certain
embodiments, the concentration of mannitol may be about 10 – about 14 mg/ml. In certain
embodiments, the concentration of mannitol may be about 12 mg/ml. In certain embodiments,
the polyol sorbitol may be included in the formulation.

[0201] A detergent or surfactant may also be added to the formulation. Exemplary detergents
10 include nonionic detergents such as polysorbates (*e.g.* polysorbates 20, 80 etc.) or poloxamers
(*e.g.*, poloxamer 188). The amount of detergent added is such that it reduces aggregation of the
formulated antibody and/or minimizes the formation of particulates in the formulation and/or
reduces adsorption. In certain embodiments, the formulation may include a surfactant which is a
polysorbate. In certain embodiments, the formulation may contain the detergent polysorbate 80
15 or Tween 80. Tween 80 is a term used to describe polyoxyethylene (20) sorbitanmonooleate (*see*
Fiedler, Lexikon der Hilfsstoffe, Editio Cantor Verlag Aulendorf, 4th edi., 1996). In certain
embodiments, the formulation may contain between about 0.1 mg/mL and about 10 mg/mL of
polysorbate 80, or between about 0.5 mg/mL and about 5 mg/mL. In certain embodiments, about
0.1% polysorbate 80 may be added in the formulation.

20 ***Lyophilized Formulation***

[0202] The lyophilized formulation for use in a method of treating advanced NSCLC or
inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC
subject includes the anti-PD-L1/TGFβ Trap molecule and a lyoprotectant. The lyoprotectant may
be sugar, *e.g.*, disaccharides. In certain embodiments, the lyoprotectant may be sucrose or
25 maltose. The lyophilized formulation may also include one or more of a buffering agent, a
surfactant, a bulking agent, and/or a preservative.

[0203] The amount of sucrose or maltose useful for stabilization of the lyophilized drug
product may be in a weight ratio of at least 1:2 protein to sucrose or maltose. In certain
embodiments, the protein to sucrose or maltose weight ratio may be of from 1:2 to 1:5.

[0204] In certain embodiments, the pH of the formulation, prior to lyophilization, may be set by addition of a pharmaceutically acceptable acid and/or base. In certain embodiments the pharmaceutically acceptable acid may be hydrochloric acid. In certain embodiments, the pharmaceutically acceptable base may be sodium hydroxide.

5 [0205] Before lyophilization, the pH of the solution containing the protein of the present disclosure may be adjusted between about 6 to about 8. In certain embodiments, the pH range for the lyophilized drug product may be from about 7 to about 8.

[0206] In certain embodiments, a salt or buffer components may be added in an amount of about 10 mM – about 200 mM. The salts and/or buffers are pharmaceutically acceptable and are
10 derived from various known acids (inorganic and organic) with “base forming” metals or amines. In certain embodiments, the buffer may be phosphate buffer. In certain embodiments, the buffer may be glycinate, carbonate, citrate buffers, in which case, sodium, potassium or ammonium ions can serve as counterion.

[0207] In certain embodiments, a “bulking agent” may be added. A “bulking agent” is a
15 compound which adds mass to a lyophilized mixture and contributes to the physical structure of the lyophilized cake (*e.g.*, facilitates the production of an essentially uniform lyophilized cake which maintains an open pore structure). Illustrative bulking agents include mannitol, glycine, polyethylene glycol and sorbitol. The lyophilized formulations of the present invention may contain such bulking agents.

20 [0208] A preservative may be optionally added to the formulations herein to reduce bacterial action. The addition of a preservative may, for example, facilitate the production of a multi-use (multiple-dose) formulation.

[0209] In certain embodiments, the lyophilized drug product for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure
25 metastatic NSCLC subject may be constituted with an aqueous carrier. The aqueous carrier of interest herein is one which is pharmaceutically acceptable (*e.g.*, safe and non-toxic for administration to a human) and is useful for the preparation of a liquid formulation, after lyophilization. Illustrative diluents include sterile water for injection (SWFI), bacteriostatic water for injection (BWFI), a pH buffered solution (*e.g.* phosphate-buffered saline), sterile saline
30 solution, Ringer's solution or dextrose solution.

[0210] In certain embodiments, the lyophilized drug product of the current disclosure is reconstituted with either Sterile Water for Injection, USP (SWFI) or 0.9% Sodium Chloride Injection, USP. During reconstitution, the lyophilized powder dissolves into a solution.

5 [0211] In certain embodiments, the lyophilized protein product of the instant disclosure is constituted to about 4.5 mL water for injection and diluted with 0.9% saline solution (sodium chloride solution).

Liquid Formulation

[0212] In embodiments, the protein product of the present disclosure is formulated as a liquid formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject. The liquid
10 formulation may be presented at a 10 mg/mL concentration in either a USP / Ph Eur type I 50R vial closed with a rubber stopper and sealed with an aluminum crimp seal closure. The stopper may be made of elastomer complying with USP and Ph Eur. In certain embodiments vials may be filled with about 61.2 mL of the protein product solution in order to allow an extractable
15 volume of 60 mL. In certain embodiments, the liquid formulation may be diluted with 0.9% saline solution. In certain embodiments vials may contain about 61.2 mL of the protein product (*e.g.*, anti-PD-L1/TGFβ Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of
20 SEQ ID NO: 1)) solution of about 20 mg/mL to about 50 mg/mL (*e.g.*, about 20 mg/mL, about 25 mg/mL, about 30 mg/mL, about 35 mg/mL, about 40 mg/mL, about 45 mg/mL or about 50 mg/mL) in order to allow an extractable volume of 60 mL for delivering about 1200 mg to about 3000 mg (*e.g.*, about 1200 mg to about 3000 mg, about 1200 mg to about 2900 mg, about 1200 mg to about 2800 mg, about 1200 mg to about 2700 mg, about 1200 mg to about 2600 mg, about 1200 mg to about 2500 mg, about 1200 mg to about 2400 mg, about 1200 mg to about 2300 mg,
25 about 1200 mg to about 2200 mg, about 1200 mg to about 2100 mg, about 1200 mg to about 2000 mg, about 1200 mg to about 1900 mg, about 1200 mg to about 1800 mg, about 1200 mg to about 1700 mg, about 1200 mg to about 1600 mg, about 1200 mg to about 1500 mg, about 1200 mg to about 1400 mg, about 1200 mg to about 1300 mg, about 1300 mg to about 3000 mg, about 1400 mg to about 3000 mg, about 1500 mg to about 3000 mg, about 1600 mg to about 3000 mg,
30 about 1700 mg to about 3000 mg, about 1800 mg to about 3000 mg, about 1900 mg to about 3000 mg, about 2000 mg to about 3000 mg, about 2100 mg to about 3000 mg, about 2200 mg to

about 3000 mg, about 2300 mg to about 3000 mg, about 2400 mg to about 3000 mg, about 2500 mg to about 3000 mg, about 2600 mg to about 3000 mg, about 2700 mg to about 3000 mg, about 2800 mg to about 3000 mg, about 2900 mg to about 3000 mg, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, about 2500 mg, about 2600 mg, about 2700 mg, about 2800 mg, about 2900 mg, or about 3000 mg) of the protein product (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40)) to a subject.

[0213] In certain embodiments, vials may contain about 61.2 mL of the protein product solution (protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40) of about 20 mg/mL to about 50 mg/mL (*e.g.*, about 20 mg/mL, about 25 mg/mL, about 30 mg/mL, about 35 mg/mL, about 40 mg/mL, about 45 mg/mL or about 50 mg/mL) in order to allow an extractable volume of 60 mL for delivering about 1200 mg to about 3000 mg (*e.g.*, about 1200 mg to about 3000 mg, about 1200 mg to about 2900 mg, about 1200 mg to about 2800 mg, about 1200 mg to about 2700 mg, about 1200 mg to about 2600 mg, about 1200 mg to about 2500 mg, about 1200 mg to about 2400 mg, about 1200 mg to about 2300 mg, about 1200 mg to about 2200 mg, about 1200 mg to about 2100 mg, about 1200 mg to about 2000 mg, about 1200 mg to about 1900 mg, about 1200 mg to about 1800 mg, about 1200 mg to about 1700 mg, about 1200 mg to about 1600 mg, about 1200 mg to about 1500 mg, about 1200 mg to about 1400 mg, about 1200 mg to about 1300 mg, about 1300 mg to about 3000 mg, about 1400 mg to about 3000 mg, about 1500 mg to about 3000 mg, about 1600 mg to about 3000 mg, about 1700 mg to about 3000 mg, about 1800 mg to about 3000 mg, about 1900 mg to about 3000 mg, about 2000 mg to about 3000 mg, about 2100 mg to about 3000 mg, about 2200 mg to about 3000 mg, about 2300 mg to about 3000 mg, about 2400 mg to about 3000 mg, about 2500 mg to about 3000 mg, about 2600 mg to about 3000 mg, about 2700 mg to about 3000 mg, about 2800 mg to about 3000 mg, about 2900 mg to about 3000 mg, about 1200 mg, about 1300 mg,

about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, about 2500 mg, about 2600 mg, about 2700 mg, about 2800 mg, about 2900 mg, or about 3000 mg) of the protein product to a treatment naïve subject or a PDx failure metastatic NSCLC subject.

5 [0214] In certain embodiments, the liquid formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject may be prepared as a 10 mg/mL concentration solution in combination with a sugar at stabilizing levels. In certain embodiments the liquid formulation may be prepared in an aqueous carrier. In certain embodiments, a stabilizer may be added in an
10 amount no greater than that which may result in a viscosity undesirable or unsuitable for intravenous administration. In certain embodiments, the sugar may be disaccharides, *e.g.*, sucrose. In certain embodiments, the liquid formulation may also include one or more of a buffering agent, a surfactant, and a preservative.

[0215] In certain embodiments, the pH of the liquid formulation may be set by addition of a
15 pharmaceutically acceptable acid and/or base. In certain embodiments, the pharmaceutically acceptable acid may be hydrochloric acid. In certain embodiments, the base may be sodium hydroxide.

[0216] In addition to aggregation, deamidation is a common product variant of peptides and proteins that may occur during fermentation, harvest/cell clarification, purification, drug
20 substance/drug product storage and during sample analysis. Deamidation is the loss of NH₃ from a protein forming a succinimide intermediate that can undergo hydrolysis. The succinimide intermediate results in a 17 u mass decrease of the parent peptide. The subsequent hydrolysis results in an 18 u mass increase. Isolation of the succinimide intermediate is difficult due to instability under aqueous conditions. As such, deamidation is typically detectable as 1 u mass
25 increase. Deamidation of an asparagine results in either aspartic or isoaspartic acid. The parameters affecting the rate of deamidation include pH, temperature, solvent dielectric constant, ionic strength, primary sequence, local polypeptide conformation and tertiary structure. The amino acid residues adjacent to Asn in the peptide chain affect deamidation rates. Gly and Ser following an Asn in protein sequences results in a higher susceptibility to deamidation.

[0217] In certain embodiments, the liquid formulation for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject may be preserved under conditions of pH and humidity to prevent deamidation of the protein product.

5 [0218] The aqueous carrier of interest herein is one which is pharmaceutically acceptable (safe and non-toxic for administration to a human) and is useful for the preparation of a liquid formulation. Illustrative carriers include sterile water for injection (SWFI), bacteriostatic water for injection (BWFI), a pH buffered solution (*e.g.* phosphate-buffered saline), sterile saline solution, Ringer's solution or dextrose solution.

10 [0219] A preservative may be optionally added to the formulations herein to reduce bacterial action. The addition of a preservative may, for example, facilitate the production of a multi-use (multiple-dose) formulation.

[0220] Intravenous (IV) formulations may be the preferred administration route in particular instances, such as when a patient is in the hospital after transplantation receiving all drugs via the
15 IV route. In certain embodiments, the liquid formulation is diluted with 0.9% Sodium Chloride solution before administration. In certain embodiments, the diluted drug product for injection is isotonic and suitable for administration by intravenous infusion.

[0221] In certain embodiments, a salt or buffer components may be added in an amount of 10 mM - 200 mM. The salts and/or buffers are pharmaceutically acceptable and are derived from
20 various known acids (inorganic and organic) with "base forming" metals or amines. In certain embodiments, the buffer may be phosphate buffer. In certain embodiments, the buffer may be glycinate, carbonate, citrate buffers, in which case, sodium, potassium or ammonium ions can serve as counterion.

[0222] A preservative may be optionally added to the formulations herein to reduce bacterial
25 action. The addition of a preservative may, for example, facilitate the production of a multi-use (multiple-dose) formulation.

[0223] The aqueous carrier of interest herein is one which is pharmaceutically acceptable (safe and non-toxic for administration to a human) and is useful for the preparation of a liquid formulation. Illustrative carriers include sterile water for injection (SWFI), bacteriostatic water

for injection (BWFI), a pH buffered solution (*e.g.* phosphate-buffered saline), sterile saline solution, Ringer's solution or dextrose solution.

[0224] A preservative may be optionally added to the formulations herein to reduce bacterial action. The addition of a preservative may, for example, facilitate the production of a multi-use (multiple-dose) formulation.

Method of Treating Cancer or Inhibiting Tumor Growth

[0225] In one aspect the present disclosure provides a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve subject or a PDx failure metastatic NSCLC subject in need thereof, the method including administering to the subject a dose of at least 1200 mg of a protein including a first polypeptide and a second polypeptide in combination with systemic chemotherapeutic agents. The first polypeptide includes: (a) at least a variable region of a heavy chain of an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGF β RII), or a fragment thereof, capable of binding Transforming Growth Factor β (TGF β). The second polypeptide includes at least a variable region of a light chain of an antibody that binds PD-L1, and the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1.

[0226] In certain embodiments, the method of treating advanced NSCLC or inhibiting tumor growth of the present disclosure involves administering to a treatment naïve subject a protein including two peptides in which the first polypeptide includes the amino acid sequence of SEQ ID NO: 3, and the second polypeptide includes the amino acid sequence of SEQ ID NO: 1. In certain embodiments, the method of treating advanced NSCLC or inhibiting tumor growth of the present disclosure involves administering to a PDx failure metastatic NSCLC subject a protein including two peptides in which the first polypeptide includes the amino acid sequence of SEQ ID NO: 3, and the second polypeptide includes the amino acid sequence of SEQ ID NO: 1. In certain embodiments, the protein is an anti-PD-L1/TGF β Trap molecule.

[0227] In certain embodiments, systemic chemotherapeutic agents administered in combination with the bifunctional protein of the present disclosure are platinum-based agents, such as cisplatin or carboplatin. In certain embodiments, platinum-based agents are administered with other chemotherapeutic agents, such as paclitaxel (or nab-paclitaxel), gemcitabine, or

pemetrexed. In certain embodiments, systemic chemotherapeutic agents administered in combination with the bifunctional protein of the present disclosure are non-platinum-based drugs such as docetaxel. As will be readily appreciated by those skilled in the art, dosages for these chemotherapeutic agents can be administered according to FDA-approved regimens and adjusted according to the judgment of the practitioner.

[0228] Contemplated herein are methods of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve subject by administering systemic chemotherapeutic agents in combination with the bifunctional protein of the present disclosure. For example, in some embodiments, the present disclosure provides the method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve subject by administering to a treatment naïve subject systemic chemotherapeutic agents such as cisplatin or carboplatin in combination with the bifunctional protein of the present disclosure. In certain embodiments, cisplatin or carboplatin are administered with other chemotherapeutic agents, such as paclitaxel (or nab-paclitaxel), gemcitabine, or pemetrexed.

[0229] Contemplated herein are methods of treating advanced NSCLC or inhibiting tumor growth in a PDx failure metastatic NSCLC subject by administering systemic chemotherapeutic agent in combination with the bifunctional protein of the present disclosure. For example, in some embodiments, the present disclosure provides the method of treating advanced NSCLC or inhibiting tumor growth in a PDx failure metastatic NSCLC subject by administering to a PDx failure metastatic NSCLC subject docetaxel in combination with the bifunctional protein of the present disclosure.

[0230] In an embodiment, the treatment naïve subject treated in accordance with the methods disclosed herein has not received prior therapy with the bifunctional protein of the present disclosure (anti-PD-L1/TGF β Trap molecule). In some embodiments, the treatment naïve cancer patient to be treated in accordance with the methods of the present disclosure does not have a mutation selected from an epidermal growth factor receptor (EGFR) sensitizing (activating) mutation, an anaplastic lymphoma kinase (ALK) translocation, a ROS1 mutation, and a BRAF V600E mutation. In some embodiments, the treatment naïve cancer (*e.g.*, advanced NSCLC (*i.e.*, metastatic NSCLC) of non-squamous histology) patient to be treated in accordance with the methods of the present disclosure does not have an epidermal growth factor receptor (EGFR) sensitizing (activating) mutation. In some embodiments, the treatment naïve cancer (*e.g.*,

advanced NSCLC (*i.e.*, metastatic NSCLC) of non-squamous histology) patient to be treated in accordance with the methods of the present disclosure does not have an anaplastic lymphoma kinase (ALK) translocation (*i.e.*, is not ALK positive). In some embodiments, the treatment naïve cancer (*e.g.*, advanced NSCLC (*i.e.*, metastatic NSCLC) of non-squamous histology) patient to be treated in accordance with the methods of the present disclosure does not have a ROS1 mutation. In some embodiments, the treatment naïve cancer (*e.g.*, advanced NSCLC (*i.e.*, metastatic NSCLC) of non-squamous histology) patient to be treated in accordance with the methods of the present disclosure does not have a BRAF V600E mutation.

[0231] In certain embodiments, the present disclosure provides a method of treating advanced non-small cell lung cancer (NSCLC) or inhibiting NSCLC tumor growth in subject indicated as having metastatic NSCLC disease progression on previous treatment with an immunotherapy in combination with chemotherapy, or on previous treatment with chemotherapy followed by treatment with an immunotherapy, or on previous treatment with an immunotherapy followed by platinum-based chemotherapy, the method comprising a first step of administering to the subject a dose of at least 1800 mg of an anti-PD-L1/ TGF β Trap protein as provided in the present disclosure, with concurrent systemic chemotherapy comprising docetaxel, and a second step of administering at least 1800 mg of anti-PD-L1/ TGF β Trap protein.

[0232] In certain embodiments, the present disclosure provides a method of treating advanced non-small cell lung cancer (NSCLC) or inhibiting NSCLC tumor growth in subject indicated as having metastatic NSCLC disease progression on previous treatment with an immunotherapy in combination with chemotherapy, or on previous treatment with chemotherapy followed by treatment with an immunotherapy, or on previous treatment with an immunotherapy followed by platinum-based chemotherapy, the method comprising a first step of administering to the subject a dose of about 2400 mg of an anti-PD-L1/ TGF β Trap protein as provided in the present disclosure, with concurrent systemic chemotherapy comprising docetaxel, and a second step of administering about 2400 mg of anti-PD-L1/ TGF β Trap protein.

[0233] In certain embodiments, the method of treating advanced NSCLC or inhibiting tumor growth of the present disclosure involves administering to a treatment naïve subject or a PDx failure metastatic NSCLC subject a protein (*e.g.*, an anti-PD-L1/TGF β Trap molecule (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product

with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40)) at a dose of about 1200 mg to about 3000 mg (*e.g.*, about 1200 mg to about 3000 mg, about 1200 mg to about 2900 mg, about 1200 mg to about 2800 mg, about 1200 mg to about 2700 mg, about 1200 mg to about 2600 mg, about 1200 mg to about 2500 mg, about 1200 mg to about 2400 mg, about 1200 mg to about 2300 mg, about 1200 mg to about 2200 mg, about 1200 mg to about 2100 mg, about 1200 mg to about 2000 mg, about 1200 mg to about 1900 mg, about 1200 mg to about 1800 mg, about 1200 mg to about 1700 mg, about 1200 mg to about 1600 mg, about 1200 mg to about 1500 mg, about 1200 mg to about 1400 mg, about 1200 mg to about 1300 mg, about 1300 mg to about 3000 mg, about 1400 mg to about 3000 mg, about 1500 mg to about 3000 mg, about 1600 mg to about 3000 mg, about 1700 mg to about 3000 mg, about 1800 mg to about 3000 mg, about 1900 mg to about 3000 mg, about 2000 mg to about 3000 mg, about 2100 mg to about 3000 mg, about 2200 mg to about 3000 mg, about 2300 mg to about 3000 mg, about 2400 mg to about 3000 mg, about 2500 mg to about 3000 mg, about 2600 mg to about 3000 mg, about 2700 mg to about 3000 mg, about 2800 mg to about 3000 mg, about 2900 mg to about 3000 mg, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, about 2500 mg, about 2600 mg, about 2700 mg, about 2800 mg, about 2900 mg, or about 3000 mg). In certain embodiments, about 1200 mg of anti-PD-L1/TGF β Trap molecule is administered to a treatment naïve advanced NSCLC subject or a PDx failure metastatic NSCLC subject once every two weeks. In certain embodiments, about 1800 mg of anti-PD-L1/TGF β Trap molecule is administered to a treatment naïve advanced NSCLC subject or a PDx failure metastatic NSCLC subject once every three weeks. In certain embodiments, about 1200 mg of a protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3 and the second polypeptide that includes the amino acid sequence of SEQ ID NO: 1 is administered to a treatment naïve subject or a PDx failure metastatic NSCLC subject once every two weeks. In certain embodiments, about 1800 mg of a protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3 and the second polypeptide that includes the amino acid sequence of SEQ ID NO: 1 is administered to a treatment naïve advanced NSCLC subject or a PDx failure metastatic NSCLC subject once every three weeks. In certain embodiments, about 1800 mg of a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second

polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40 is administered to a treatment naïve advanced NSCLC subject or a PDx failure metastatic NSCLC subject once every three weeks.

[0234] In certain embodiments, the dose administered to a treatment naïve advanced NSCLC subject or a PDx failure metastatic NSCLC subject may be about 1200 mg, about 1225 mg, about 1250 mg, about 1275 mg, about 1300 mg, about 1325 mg, about 1350 mg, about 1375 mg, about 1400 mg, about 1425 mg, about 1450 mg, about 1475 mg, about 1500 mg, about 1525 mg, about 1550 mg, about 1575 mg, about 1600 mg, about 1625 mg, about 1650 mg, about 1675 mg, about 1700 mg, about 1725 mg, about 1750 mg, about 1775 mg, about 1800 mg, about 1825 mg, about 1850 mg, about 1875 mg, about 1900 mg, about 1925 mg, about 1950 mg, about 1975 mg, about 2000 mg, about 2025 mg, about 2050 mg, about 2075 mg, 2100 mg, about 2125 mg, about 2150 mg, about 2175 mg, about 2200 mg, about 2225 mg, about 2250 mg, about 2275 mg, about 2300 mg, about 2325 mg, about 2350 mg, about 2375 mg, or about 2400 mg.

[0235] In certain embodiments, the dose administered to a treatment naïve advanced NSCLC subject or a PDx failure metastatic NSCLC subject may be administered once every two weeks. In certain embodiments, the dose administered to a treatment naïve advanced NSCLC subject or a PDx failure metastatic NSCLC subject may be administered once every three weeks. In certain embodiments, the protein may be administered by intravenous administration, *e.g.*, with a prefilled bag, a prefilled pen, or a prefilled syringes. In certain embodiments, the protein is administered intravenously from a 250 ml saline bag, and the intravenous infusion may be for about one hour (*e.g.*, 50 to 80 minutes). In certain embodiments, the bag is connected to a channel comprising a tube and/or a needle.

[0236] In some embodiments, the advanced NSCLC exhibits squamous or non-squamous histology. For example, in an embodiment, the method treats squamous advanced NSCLC. In some embodiments, the method treats non-squamous advanced NSCLC.

[0237] In certain embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering at least 1200 mg (*e.g.*, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, or more) of

anti-PD-L1/TGF β Trap, which includes a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1. In certain embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are

5 treated by intravenously administering at least 1200 mg (*e.g.*, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, or more) of anti-PD-L1/TGF β Trap, which includes a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino

10 acid sequences of SEQ ID NOs: 38, 39, and 40.

[0238] In certain embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering about 1200 mg – about 2400 mg (*e.g.*, about 1200 mg to about 2400 mg, about 1200 mg to about 2300 mg, about 1200 mg to about 2200 mg, about

15 1200 mg to about 2100 mg, about 1200 mg to about 2000 mg, about 1200 mg to about 1900 mg, about 1200 mg to about 1800 mg, about 1200 mg to about 1700 mg, about 1200 mg to about 1600 mg, about 1200 mg to about 1500 mg, about 1200 mg to about 1400 mg, about 1200 mg to about 1300 mg, about 1300 mg to about 2400 mg, about 1400 mg to about 2400 mg, about 1500 mg to about 2400 mg, about 1600 mg to about 2400 mg, about 1700 mg to about 2400 mg, about

20 1800 mg to about 2400 mg, about 1900 mg to about 2400 mg, about 2000 mg to about 2400 mg, about 2100 mg to about 2400 mg, about 2200 mg to about 2400 mg, or about 2300 mg to about 2400 mg) of anti-PD-L1/TGF β Trap, which includes a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1.

25 **[0239]** In certain embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering about 1200 mg – about 2400 mg (*e.g.*, about 1200 mg to about 2400 mg, about 1200 mg to about 2300 mg, about 1200 mg to about 2200 mg, about 1200 mg to about 2100 mg, about 1200 mg to about 2000 mg, about 1200 mg to about 1900 mg,

30 about 1200 mg to about 1800 mg, about 1200 mg to about 1700 mg, about 1200 mg to about 1600 mg, about 1200 mg to about 1500 mg, about 1200 mg to about 1400 mg, about 1200 mg to

about 1300 mg, about 1300 mg to about 2400 mg, about 1400 mg to about 2400 mg, about 1500 mg to about 2400 mg, about 1600 mg to about 2400 mg, about 1700 mg to about 2400 mg, about 1800 mg to about 2400 mg, about 1900 mg to about 2400 mg, about 2000 mg to about 2400 mg, about 2100 mg to about 2400 mg, about 2200 mg to about 2400 mg, or about 2300 mg to about 2400 mg) of anti-PD-L1/TGF β Trap, which includes a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40.

[0240] In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGF β Trap at a dose of about 1200 mg once every 2 weeks. In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGF β Trap at a dose of 1200 mg once every 2 weeks. In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGF β Trap at a dose of about 1800 mg once every 3 weeks. In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGF β Trap at a dose of 1800 mg once every 3 weeks. In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGF β Trap at a dose of about 2400 mg once every 3 weeks. In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGF β Trap at a dose of 2400 mg once every 3 weeks.

[0241] In certain embodiments, the advanced NSCLC to be treated is PD-L1 positive. For example, in certain embodiments, the advanced NSCLC to be treated exhibits PD-L1 expression (*e.g.*, PD-L1 positive or high PD-L1 expression). In certain embodiments, the advanced NSCLC to be treated does not exhibit PD-L1 expression. In exemplary embodiments, the advanced NSCLC are treated irrespective of PD-L1 expression.

[0242] In certain embodiments, the PD-L1 expression levels in the advanced NSCLC is detected using an anti-PD-L1 antibody. The tissue sample may be a formalin-fixed, paraffin-embedded advanced stage IV NSCLC tissue.

[0243] In some embodiments for example, PD-L1 high may be defined as $\geq 80\%$ PD-L1 positive tumor cells (tumor proportion score [TPS]) as determined by the 73-10 assay. In some
5 embodiments, PD-L1 high may be defined as Tumor Proportion Score (TPS) $\geq 50\%$ as determined by the PD-L1 IHC 22C3 pharmDx assay. Methods of detecting a biomarker, such as PD-L1 for example, on a cancer or tumor, are routine in the art and are contemplated herein. Non-limiting examples include immunohistochemistry, immunofluorescence and fluorescence
10 activated cell sorting (FACS). In some embodiments for example, PD-L1 expression level is determined by VENTANA PD-L1 (SP263) assay, which is a quantitative immunochemical assay using rabbit monoclonal anti-PD-L1 clone SP263 intended for use in the assessment of the PD-L1 protein in formalin-fixed, paraffin-embedded (FFPE) cancer tissue. PD-L1 status is determined by the percentage of tumor cells with any membrane staining above background or
15 by the percentage of tumor-associated immune cells with staining (IC+) at any intensity above background. For example, the US FDA approved SP263 test for the identification of patients with locally advanced or metastatic urothelial carcinoma most likely to benefit from durvalumab, the percent of tumor area occupied by any tumor-associated immune cells (Immune Cells Present, ICP) is used to determine IC+, which is the percent area of ICP exhibiting PD-L1
20 positive immune cell staining. PD-L1 status is considered high if any of the following are met: $\geq 25\%$ of tumor cells exhibit membrane staining; or, ICP $> 1\%$ and IC+ $\geq 25\%$; or, ICP = 1% and IC+ = 100% . Methods of detecting PD-L1 on NSCLC are described in “PD-L1 expression testing in non-small cell lung cancer,” Teixidó *et al.*, *Ther. Adv. Med. Oncol.* (2018), 10. In certain embodiments, patients are enrolled irrespective of PD-L1 expression.

[0244] In some embodiments, treatment naïve subjects or patients with PD-L1 high, advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGF β Trap at a dose of about 1200 mg once every 2 weeks. In some embodiments, treatment naïve subjects or patients with PD-L1 high, advanced NSCLC (*e.g.*, squamous or non-squamous advanced
30 NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGF β Trap at a dose of about 1800 mg once every 3 weeks. In some embodiments,

treatment naïve subjects or patients with PD-L1 high, advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGFβ Trap at a dose of about 2100 mg once every 3 weeks. In some embodiments, treatment naïve subjects or patients with PD-L1 high, advanced NSCLC (*e.g.*, squamous or non-squamous advanced NSCLC) or PDx failure metastatic NSCLC subjects are treated by intravenously administering anti-PD-L1/TGFβ Trap at a dose of about 2400 mg once every 3 weeks.

[0245] In some embodiments, the treatment naïve subject or patient to be treated does not have a mutation selected from an EGFR sensitizing mutation, an ALK translocation, a ROS1 mutation, and a BRAF V600E mutation. For example, in some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, metastatic NSCLC of non-squamous histology), but who do not have a mutation selected from an EGFR sensitizing mutation, an ALK translocation, a ROS1 mutation, and a BRAF V600E mutation, are treated by intravenously administering anti-PD-L1/TGFβ Trap at a dose of at least 500 mg (*e.g.*, about 500 mg, about 600 mg, about 700 mg, about 800 mg, about 900 mg, about 1000 mg, about 1100 mg, about 1200 mg, about 1300 mg, about 1400 mg, about 1500 mg, about 1600 mg, about 1700 mg, about 1800 mg, about 1900 mg, about 2000 mg, about 2100 mg, about 2200 mg, about 2300 mg, about 2400 mg, or more). In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, metastatic NSCLC of non-squamous histology) who do not have a mutation selected from an EGFR sensitizing mutation, an ALK translocation, a ROS1 mutation, and a BRAF V600E mutation are treated by intravenously administering anti-PD-L1/TGFβ Trap at a dose of about 1200 mg once every 2 weeks. In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, metastatic NSCLC of non-squamous histology) who do not have a mutation selected from an EGFR sensitizing mutation, an ALK translocation, a ROS1 mutation, and a BRAF V600E mutation are treated by intravenously administering anti-PD-L1/TGFβ Trap at a dose of about 1800 mg once every 3 weeks. In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, metastatic NSCLC of non-squamous histology) who do not have a mutation selected from an EGFR sensitizing mutation, an ALK translocation, a ROS1 mutation, and a BRAF V600E mutation are treated by intravenously administering anti-PD-L1/TGFβ Trap at a dose of about 2100 mg once every 3 weeks. In some embodiments, treatment naïve subjects or patients with advanced NSCLC (*e.g.*, metastatic NSCLC of non-squamous histology) who do not have a mutation selected from an EGFR

sensitizing mutation, an ALK translocation, a ROS1 mutation, and a BRAF V600E mutation are treated by intravenously administering anti-PD-L1/TGF β Trap at a dose of about 2400 mg once every 3 weeks.

[0246] In some embodiments, the methods of treatment disclosed herein result in a disease response or improved survival (*e.g.*, survival of up to and including 6 months, 12 months, 18 months, 22 months, 28 months, 32 months, 38 months, 44 months, 50 months, 56 months, 62 months, 68 months, 74 months, 80 months, 86 months, 92 months, 98 months, 104 months, or 110 months) of the subject or patient. In certain embodiments, improved survival is at least 108 months. In some embodiments for example, the disease response may be a complete response, a partial response, or a stable disease. In some embodiments for example, the improved survival (*e.g.*, survival of up to and including 6 months, 12 months, 18 months, 22 months, 28 months, 32 months, 38 months, 44 months, 50 months, 56 months, 62 months, 68 months, 74 months, 80 months, 86 months, 92 months, 98 months, 104 months, or 110 months) could be progression-free survival (PFS) or overall survival (OS). In certain embodiments, improved survival of PFS and/or OS is at least 108 months. In some embodiments, improvement (*e.g.*, in PFS) is determined relative to a period prior to initiation of treatment with an anti-PD-L1/TGF β Trap of the present disclosure. Methods of determining disease response (*e.g.*, complete response, partial response, or stable disease) and patient survival (*e.g.*, PFS, overall survival (*e.g.*, survival of up to and including 6 months, 12 months, 18 months, 22 months, 28 months, 32 months, 38 months, 44 months, 50 months, 56 months, 62 months, 68 months, 74 months, 80 months, 86 months, 92 months, 98 months, 104 months, or 110 months)) for cancer or tumor therapy are routine in the art and are contemplated herein. In certain embodiments, patient survival is at least 108 months. In some embodiments, disease response is evaluated according to RECIST 1.1 after subjecting the treated patient to contrast-enhanced computed tomography (CT) or magnetic resonance imaging (MRI) of the affected area (*e.g.*, chest/abdomen and pelvis covering the area from the superior extent of the thoracic inlet to the symphysis pubis).

Delivery Device

[0247] In one aspect, the present disclosure provides a drug delivery device for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject, wherein the device includes a formulation comprising about 500 mg – about 3000 mg of a protein including a first polypeptide and a

second polypeptide, the first polypeptide includes: (a) at least a variable region of a heavy chain of an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGF β RII), or a fragment thereof, capable of binding Transforming Growth Factor β (TGF β), the second polypeptide includes at least a variable
 5 region of a light chain of an antibody that binds PD-L1, and the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1.

[0248] In certain embodiments, the device may be a bag, a pen, or a syringe. In certain embodiments, the bag may be connected to a channel comprising a tube and/or a needle.

10 **[0249]** In certain embodiments of the present disclosure, the drug delivery device for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject may include an about 1200 mg to about 3000 mg (*e.g.*, about 1200 mg to about 3000 mg, about 1200 mg to about 2900 mg, about 1200 mg to about 2800 mg, about 1200 mg to about 2700 mg, about 1200 mg to about 2600 mg, about 1200
 15 mg to about 2500 mg, about 1200 mg to about 2400 mg, about 1200 mg to about 2300 mg, about 1200 mg to about 2200 mg, about 1200 mg to about 2100 mg, about 1200 mg to about 2000 mg, about 1200 mg to about 1900 mg, about 1200 mg to about 1800 mg, about 1200 mg to about 1700 mg, about 1200 mg to about 1600 mg, about 1200 mg to about 1500 mg, about 1200 mg to about 1400 mg, about 1200 mg to about 1300 mg, about 1300 mg to about 3000 mg, about 1400
 20 mg to about 3000 mg, about 1500 mg to about 3000 mg, about 1600 mg to about 3000 mg, about 1700 mg to about 3000 mg, about 1800 mg to about 3000 mg, about 1900 mg to about 3000 mg, about 2000 mg to about 3000 mg, about 2100 mg to about 3000 mg, about 2200 mg to about 3000 mg, about 2300 mg to about 3000 mg, about 2400 mg to about 3000 mg, about 2500 mg to about 3000 mg, about 2600 mg to about 3000 mg, about 2700 mg to about 3000 mg, about 2800
 25 mg to about 3000 mg, or about 2900 mg to about 3000 mg) of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap, which includes a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino
 30 acid sequences of SEQ ID NOs: 38, 39, and 40). In certain embodiments, the drug delivery device may include about 1200 to about 2400 mg dose of a protein of the present disclosure

(*e.g.*, anti-PD-L1/TGF β Trap, which includes a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1). In certain embodiments, the drug delivery device may include about 1800 mg, about 2100 mg, or about 2400 mg dose of the protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap, which includes a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40).

10 [0250] In certain embodiments, the drug delivery device includes an about 1200 mg, about 1800 mg, about 2100 mg, or about 2400 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap, which includes a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40). In certain embodiments, the drug delivery device for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject, includes an about 1800 mg dose of a protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap, which includes a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40). In certain embodiments, the drug delivery device for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject, includes an about 1200 mg, about 1800 mg, about 2100 mg, or about 2400 mg dose of the protein product with a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40).

[0251] In certain embodiments, the drug delivery device for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject, includes an about 1200 mg dose of the protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40)). In certain embodiments, the drug delivery device for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject, includes an about 1800 mg dose of the protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1; or a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40)).

[0252] In certain embodiments, the drug delivery device for use in a method of treating advanced NSCLC or inhibiting tumor growth in a treatment naïve cancer patient or a PDx failure metastatic NSCLC subject may include about 1200 mg, about 1225 mg, about 1250 mg, about 1275 mg, about 1300 mg, about 1325 mg, about 1350 mg, about 1375 mg, about 1400 mg, about 1425 mg, about 1450 mg, about 1475 mg, about 1500 mg, about 1525 mg, about 1550 mg, about 1575 mg, about 1600 mg, about 1625 mg, about 1650 mg, about 1675 mg, about 1700 mg, about 1725 mg, about 1750 mg, about 1775 mg, about 1800 mg, about 1825 mg, about 1850 mg, about 1875 mg, about 1900 mg, about 1925 mg, about 1950 mg, about 1975 mg, about 2000 mg, about 2025 mg, about 2050 mg, about 2075 mg, about 2100 mg, about 2125 mg, about 2150 mg, about 2175 mg, about 2200 mg, about 2225 mg, about 2250 mg, about 2275 mg, about 2300 mg, about 2325 mg, about 2350 mg, about 2375 mg, or about 2400 mg of the protein of the present disclosure (*e.g.*, anti-PD-L1/TGF β Trap, *e.g.*, a protein product with a first polypeptide that comprises the amino acid sequences of SEQ ID NOs: 35, 36, and 37, and a second polypeptide that comprises the amino acid sequences of SEQ ID NOs: 38, 39, and 40).

Protein Production

[0253] The antibody-cytokine Trap proteins are generally produced recombinantly, using mammalian cells containing a nucleic acid engineered to express the protein. Although one example of a suitable cell line and protein production method is described in Examples 1 and 2 of US 20150225483 A1, a wide variety of suitable vectors, cell lines and protein production methods have been used to produce antibody-based biopharmaceuticals and could be used in the synthesis of these antibody-cytokine Trap proteins.

Therapeutic Indications

[0254] The anti-PD-L1/TGF β Trap proteins described in the application (*e.g.*, including a first polypeptide that includes the amino acid sequence of SEQ ID NO: 3, and a second polypeptide that includes the amino acid sequence of SEQ ID NO: 1), as well as the disclosed intravenous drug delivery formulations and delivery devices comprising said anti-PD-L1/TGF β Trap proteins, can be used to treat advanced NSCLC or reduce tumor growth in a treatment naïve patient or a PDx failure metastatic NSCLC subject.

[0255] The advanced NSCLC or tumor to be treated with an anti-PD-L1/ TGF β Trap may be squamous or non-squamous NSCLC. The advanced NSCLC or tumor to be treated with an anti-PD-L1/ TGF β Trap may have expression of PD-L1 and/or TGF β in the tumor, the correlation of their expression levels with prognosis or disease progression, and preclinical and clinical experience on the sensitivity of the tumor to treatments targeting PD-L1 and TGF β . In certain embodiments, the advanced NSCLC or tumor to be treated with an anti-PD-L1/ TGF β Trap does not have expression of PD-L1 in the tumor.

[0256] In some embodiments, the treatment naïve cancer (*e.g.*, advanced NSCLC (*i.e.*, metastatic NSCLC) of non-squamous histology) patient to be treated in accordance with the methods of the present disclosure does not have a mutation selected from an epidermal growth factor receptor (EGFR) sensitizing (activating) mutation, an anaplastic lymphoma kinase (ALK) translocation, a ROS1 mutation, and a BRAF V600E mutation. In some embodiments, the treatment naïve cancer (*e.g.*, advanced NSCLC (*i.e.*, metastatic NSCLC) of non-squamous histology) patient to be treated in accordance with the methods of the present disclosure does not have an epidermal growth factor receptor (EGFR) sensitizing (activating) mutation. In some embodiments, the treatment naïve cancer (*e.g.*, advanced NSCLC (*i.e.*, metastatic NSCLC) of

non-squamous histology) patient to be treated in accordance with the methods of the present disclosure does not have an anaplastic lymphoma kinase (ALK) translocation (i.e., is not ALK positive). In some embodiments, the treatment naïve cancer (e.g., advanced NSCLC (i.e., metastatic NSCLC) of non-squamous histology)) patient to be treated in accordance with the methods of the present disclosure does not have a ROS1 mutation. In some embodiments, the treatment naïve cancer (e.g., advanced NSCLC (i.e., metastatic NSCLC) of non-squamous histology) patient to be treated in accordance with the methods of the present disclosure does not have a BRAF V600E mutation.

EXAMPLES

10 [0257] The disclosure now being generally described, will be more readily understood by reference to the following examples, which are included merely for purposes of illustration of certain aspects and embodiments of the present disclosure, and are not intended to limit the scope of the disclosure in any way.

EXAMPLE 1: Packaging of Intravenous Drug Formulation

15 [0258] The formulation of anti-PD-L1/TGF β Trap is prepared as a lyophilized formulation or a liquid formulation. For preparing the lyophilized formulation, freeze-dried anti-PD-L1/TGF β Trap is sterilized and stored in single-use glass vials. Several such glass vials are then packaged in a kit for delivering a specific body weight independent dose to a subject diagnosed with a cancer or a tumor. Depending on the dose requirement, the kit contains 12-60 vials.

20 Alternatively, the formulation is prepared and packaged as a liquid formulation and stored as 250 mg/vial to 1000 mg/vial. For example, the formulation is a liquid formulation and stored as 600 mg/vial, or stored as 250 mg/vial. In another example, the anti-PD-L1/TGF β Trap is formulated as a 10 mg/mL solution and is supplied in USP/Ph Eur type I vials filled to allow an extractable volume of 60 mL (600 mg/60 mL) and closed with rubber stoppers in serum format complying

25 with USP and Ph Eur with an aluminum crimpseal closure.

[0259] A subject diagnosed with advanced NSCLC is intravenously administered a formulation containing 1800 mg to 2400 mg of anti-PD-L1/TGF β Trap. For example, the subject is intravenously administered 1800 mg of anti-PD-L1/TGF β Trap once every three weeks or 2100 mg or 2400 mg of anti-PD-L1/TGF β Trap once every three weeks. The intravenous

administration is from a saline bag. The amount of the anti-PD-L1/TGF β Trap administered to a subject is independent of the subject's body weight.

EXAMPLE 2: Anti-PD-L1/TGF β Trap Administration with Concomitant Chemotherapy of an Advanced NSCLC Patient Cohort

5 [0260] In an exemplary embodiment, treatment-naïve patients with non-squamous advanced stage IV NSCLC, irrespective of PD-L1 expression, are treated with a first step of administering anti-PD-L1/TGF β Trap in combination with systemic chemotherapy (cisplatin or carboplatin in combination with pemetrexed) followed by a second step of administering anti-PD-L1/TGF β Trap in combination with pemetrexed (Cohort A) (**FIG. 8**). In an exemplary embodiment,

10 treatment-naïve patients with either squamous or non-squamous advanced stage IV non-small cell lung cancer (NSCLC), irrespective of PD-L1 expression, are treated with a first step of administering anti-PD-L1/TGF β Trap in combination with systemic chemotherapy (cisplatin or carboplatin in combination with gemcitabine (Cohort C), or carboplatin in combination with paclitaxel (or nab-paclitaxel (Cohort B)) followed by a second step of administering anti-PD-

15 L1/TGF β Trap alone (**FIG. 8**). In an exemplary embodiment, patients with either squamous or non-squamous advanced stage IV NSCLC who had PDx failure metastatic NSCLC, irrespective of PD-L1 expression, are treated with a first step of administering anti-PD-L1/TGF β Trap in combination with docetaxel followed by a second step of administering anti-PD-L1/TGF β Trap alone (Cohort D) (**FIG. 8**).

20 [0261] The patients in Cohorts A-C must not have an epidermal growth factor receptor (EGFR) sensitizing mutation or an anaplastic lymphoma kinase (ALK) translocation, and if tested, a ROS1 mutation, or a BRAF V600E mutation if targeted therapy is approved.

[0262] In one exemplary embodiment, systemic chemotherapy is administered as either cisplatin/carboplatin concurrently with gemcitabine. In one exemplary embodiment, cisplatin is

25 administered in the first step at a dose of 100 mg/m² intravenously on Day 1 for every 21 days, concurrently with gemcitabine administration at a dose of 1000 mg/m² intravenously, on Days 1, 8, and 15 for every 21 days, for at least four cycles, and systemic chemotherapy is discontinued in the second step. In one exemplary embodiment, carboplatin is administered in the first step at a dose of AUC 4-6 intravenously on Day 1 for every 21 days, concurrently with gemcitabine

30 administration at a dose of 1000 mg/m² intravenously, on Days 1, 8, and 15 for every 21 days, for at least four cycles, and systemic chemotherapy is discontinued in the second step.

[0263] In one exemplary embodiment, systemic chemotherapy is administered as carboplatin concurrently with paclitaxel (bound or unbound to albumin). In one exemplary embodiment, carboplatin is administered in the first step at a dose of AUC 4-6 intravenously, concurrently with paclitaxel (bound or unbound to albumin) administration at a dose of 225 mg/m² intravenously over 3 hours, on Day 1 for every 21 days for at least four cycles, and systemic chemotherapy is discontinued for the second step. In one exemplary embodiment, carboplatin is administered in the first step at a dose of AUC 4-6 intravenously on Day 1 for every 21 days, concurrently with albumin-bound paclitaxel administration at a dose of 100 mg/m² intravenously, on Days 1, 8, and 15 for every 21 days, for at least four cycles, and systemic chemotherapy is discontinued for the second step.

[0264] In one exemplary embodiment, systemic chemotherapy is administered to non-squamous NSCLC patients as cisplatin/carboplatin concurrently with pemetrexed. In one exemplary embodiment, cisplatin is administered in the first step at a dose of 75 mg/m² intravenously, concurrently with pemetrexed administration at a dose of 500mg/m² intravenously, on Day 1 for every 21 days for at least four cycles, and pemetrexed is administered alone with anti-PD-L1/TGFβ Trap in the second step at a dose of 500mg/m² intravenously, on Day 1 for every 21 days. In one exemplary embodiment, carboplatin is administered in the first step at a dose of AUC 4-6 intravenously, concurrently with pemetrexed administration at a dose of 500 mg/m² intravenously, on Day 1 for every 21 days for at least four cycles, and pemetrexed is administered alone with anti-PD-L1/TGFβ Trap in the second step at a dose of 500 mg/m² intravenously, on Day 1 for every 21 days.

[0265] In one exemplary embodiment, PDx failure metastatic NSCLC subjects are administered docetaxel in combination with anti-PD-L1/TGFβ Trap. Docetaxel is administered in the first step at a dose of 75 mg/m² intravenously over 60 minutes every 3 weeks. For example, docetaxel is administered on Day 1 of every 21 days, for at least four cycles. In one exemplary embodiment, docetaxel is administered on Day 1 of every 21 days for four cycles.

[0266] Standard premedication consisting of an H2-blocker, antiemetics, dexamethasone (oral or intravenous) are administered according to local guidelines. For patients receiving paclitaxel (or nab-paclitaxel), standard premedication consisting of diphenhydramine 25-50 mg, an H2-blocker, and dexamethasone (oral or IV is acceptable) according to local standards should be given at least 30 minutes prior to paclitaxel (or nab-paclitaxel). For patients receiving

carboplatin and paclitaxel (or nab-paclitaxel), carboplatin will be given with standard antiemetics after the paclitaxel (or nab-paclitaxel) is administered.

[0267] In one exemplary embodiment, anti-PD-L1/TGF β Trap is administered as a BW-independent dose of 1800 mg to cancer patients with advanced non-small cell lung cancer (NSCLC) once every three weeks. The administration is performed intravenously for about an hour (-10 minutes / +20 minutes, *e.g.*, 50 minutes to 80 minutes). In one exemplary embodiment, anti-PD-L1/TGF β Trap is administered as a BW-independent dose of 2100 mg to cancer patients with advanced non-small cell lung cancer (NSCLC) once every three weeks. The administration is performed intravenously for about an hour (-10 minutes / +20 minutes, *e.g.*, 50 minutes to 80 minutes).

[0268] For phase Ib/2, phase 2 and phase 3 studies in which anti-PD-L1/TGF β Trap is administered in combination with systemic chemotherapies, a modeling approach is used to select the once every three weeks dose of anti-PD-L1/TGF β Trap. Because most chemotherapies are administered once every three weeks, the same dosing interval for anti-PD-L1/TGF β Trap can be employed for convenience and compliance. For the selection of the once every three weeks dose, an efficacy profile comparable to that for 1200 mg once every two weeks dose can be achieved. $C_{\text{trough,ss}}$ and average concentration over the dosing interval at steady-state are similar to or higher than that achieved with 1200 mg once every two weeks dosing and most patients can have $C_{\text{trough,ss}}$ above the target concentration of 50 $\mu\text{g/mL}$. Based on the Pharmacokinetics-Pharmacodynamics (PK-PD) profile characterized during dosing for dose-escalation and population PK-based simulations, 2400 mg once every three weeks is expected to achieve median $C_{\text{trough,ss}}$ similar to 1200 mg once every two weeks dosing. If the elimination half-life of anti-PD-L1/TGF β Trap is about 7 days, an approximate doubling of dose will maintain the same C_{trough} with once every three weeks dosing as with once every two weeks dosing.

[0269] In one exemplary embodiment, anti-PD-L1/TGF β Trap is administered as BW-independent dose of 2400 mg to cancer patients with advanced non-small cell lung cancer (NSCLC) once every three weeks. The administration is performed intravenously for about an hour (-10 minutes / +20 minutes, *e.g.*, 50 minutes to 80 minutes). In one exemplary embodiment, anti-PD-L1/TGF β Trap is administered as BW-independent dose of 2600 mg, 2800 mg, or 3000 mg to cancer patients with advanced non-small cell lung cancer (NSCLC) once every three

weeks. The administration is performed intravenously for about an hour (-10 minutes / +20 minutes, *e.g.*, 50 minutes to 80 minutes). In one or more exemplary embodiments, in order to mitigate potential infusion-related reactions, premedication with an antihistamine and with paracetamol (acetaminophen) (for example, 25-50 mg diphenhydramine and 500-650 mg paracetamol [acetaminophen] IV or oral equivalent) approximately 30 to 60 minutes prior to each anti-PD-L1/TGF β Trap dose is administered for the first 2 infusions. If Grade $\geq$ 2 infusion reactions are observed during the first two infusions, premedication is not stopped. Steroids as premedication are not permitted.

[0270] The following describes the inclusion criteria for patients used in this example.

10 Patients:

- are $\geq$ 18 years, inclusive at the time of informed consent
- have histologically confirmed diagnosis of advanced NSCLC
- have measurable disease based on RECIST 1.1 (*see* Eisenhauer et al., EJC. 2009; 45:228-247)
- 15 – have not received prior systemic therapy treatment, or any antibody or drug targeting T-cell coregulatory proteins (immune checkpoints) such as anti-PDL1, or anti-CTLA-4 antibody, since diagnosis of advanced NSCLC (completion of treatment with cytotoxic chemotherapy, biological therapy, and/or radiation as part of neoadjuvant/adjuvant therapy is allowed as long as therapy was completed
- 20 at least 6 months prior to the diagnosis of metastatic disease) (enrolled in Cohorts A, B, and C)
- had disease progression on previous treatment with PD-(L)1 inhibitors in combination with chemotherapy, or treatment with chemotherapy followed by treatment with PD-(L)1 inhibitors, or PD-(L)1 inhibitor followed by a platinum-based chemotherapy (enrolled in Cohort D)
- 25 – have a life expectancy of at least 12 weeks (based on physician's assessment of the prognosis of the patient after diagnosis)
- have available tumor material (< 6 months old) adequate for biomarker analysis
- have Eastern Cooperative Oncology Group Performance Status (ECOG PS) of 0
- 30 to 1

- have adequate pulmonary function defined as a forced expiratory volume in 1 second (FEV_1) ≥ 1.2 liters or $\geq 50\%$ of predicted normal volume measured within 3 weeks prior to randomization
- have adequate hematological function defined by absolute neutrophil count (ANC) $\geq 1.5 \times 10^9/L$, platelet count $\geq 100 \times 10^9/L$, and Hgb ≥ 9 g/ dL
- have adequate hepatic function defined by a total bilirubin level $\leq 1.5 \times$ upper limit of normal (ULN), an aspartate aminotransferase (AST) level $\leq 3.0 \times$ ULN, an alanine aminotransferase (ALT) level $\leq 3.0 \times$ ULN and alkaline phosphatase ≤ 2.5 ULN. For participants with liver involvement in their tumor, aspartate aminotransferase (AST) $\leq 5.0 \times$ ULN, alanine aminotransferase (ALT) $\leq 5.0 \times$ ULN, and bilirubin $\leq 3.0 \times$ ULN is acceptable
- have adequate renal function defined by creatinine $\leq 1.5 \times$ ULN or a calculated creatinine clearance > 30 mL/min; and
- have adequate coagulation function defined as international normalized ratio (INR) or prothrombin time (PT) $\leq 1.5 \times$ ULN unless the participant is receiving anticoagulant therapy, and activated partial thromboplastin time (aPTT) $\leq 1.5 \times$ ULN unless the participant is receiving anticoagulant therapy.

EXAMPLE 3: Therapeutic Efficacy in Treatment of Advanced NSCLC Patients with Anti-PD-L1/TGF β Trap with Concomitant Chemotherapy

[0271] *Objective:* The purpose of this study is to evaluate the safety, tolerability, and efficacy of anti-PD-L1/TGF β Trap in combination with systemic chemotherapeutic agents as a treatment for patients with advanced squamous or non-squamous non-small cell lung cancer (NSCLC), irrespective of PD-L1 tumor expression. Additional purpose of the study is to evaluate the safety, tolerability, and efficacy of anti-PD-L1/TGF β Trap in combination with systemic chemotherapeutic agents as a treatment for PDx failure metastatic NSCLC patients. The rationale for using anti-PD-L1/TGF β Trap in these NSCLC patient cohorts is that anti-PD-L1/TGF β Trap targets PD-L1 and TGF β , two major mechanisms of immunosuppression in the tumor microenvironment. Preclinical data suggest that anti-PD-L1/TGF β Trap strongly enhances antitumor activity and prolongs survival in mouse tumor models above the effect of either the anti PD-L1 antibody avelumab or the TGF β Trap control alone. Thus, simultaneous

neutralization of TGF- β , a molecule known to inhibit tumor immune activation, might stimulate clinical response in patients in combination with systemic chemotherapeutic agents.

[0272] *Study Design:* This study evaluates disease response and survival primary endpoints to assess clinical benefit of an anti-PD-L1/TGF β Trap in combination with systemic

5 chemotherapeutic agents as a treatment for patients with advanced NSCLC or PDx failure metastatic NSCLC patients. The patients in this study meet the inclusion criteria of patients described in Example 2, and either have not received prior systemic therapy treatment for their advanced NSCLC (patients are treatment naïve in Cohorts A, B, or C) or PDx failure metastatic NSCLC patients (patients in Cohort D).

10 **[0273]** The patients are administered intravenous dosages of anti-PD-L1/TGF β Trap and systemic chemotherapeutic agents according to a two-step process as described in Example 2. In one exemplary embodiment, as described in Example 2, participants with either squamous or non-squamous NSCLC are administered anti-PD-L1/TGF β Trap in combination with cisplatin/carboplatin and gemcitabine in the first step, and then administered anti-PD-L1/TGF β
15 Trap alone in the second step. In one exemplary embodiment, as described in Example 2, participants with either squamous or non-squamous NSCLC are administered anti-PD-L1/TGF β Trap in combination with carboplatin and paclitaxel (or nab-paclitaxel) in the first step, and then administered anti-PD-L1/TGF β Trap alone in the second step.

[0274] In one exemplary embodiment, as described in Example 2, participants with non-squamous NSCLC are administered anti-PD-L1/TGF β Trap in combination with
20 cisplatin/carboplatin and pemetrexed in the first step, and then administered anti-PD-L1/TGF β Trap with pemetrexed only in the second step.

[0275] In one exemplary embodiment, as described in Example 2, PDx failure metastatic NSCLC patients are administered anti-PD-L1/TGF β Trap in combination with docetaxel in the
25 first step, and then administered anti-PD-L1/TGF β Trap alone in the second step.

[0276] Treatment is continued until confirmed progressive disease (PD) per Response Evaluation Criteria in Solid Tumors version 1.1 (RECIST 1.1), unacceptable toxicity, or for up to 24 months. In the case of PD, treatment may continue past the initial determination of PD or confirmed PD if the patient's Eastern Cooperative Oncology Group Performance Status (ECOG
30 PS) remains stable, and if the participant will benefit from continued treatment. Patients who

experience stable disease (SD), partial response (PR), or complete response (CR) will continue treatment until the end of 24 months, although additional treatment is be possible.

[0277] Throughout treatment, safety is assessed through the recording, reporting and analysis of baseline medical conditions, adverse events (AEs), physical examination findings, including vital signs, ECOG performance status, and laboratory tests. For each cohort of 8 participants, the combination is considered safe when DLTs (Dose-Limiting Toxicity) is observed in ≤ 2 of the 8 evaluable participants.

[0278] *Pharmacokinetic profile:* Pharmacokinetic (PK) profile of anti-PD-L1/TGF β Trap is generated in terms of concentration immediately at the end of infusion (C_{eoi}) and concentration immediately before next dosing (C_{trough}). In one exemplary embodiment, pharmacokinetic (PK) profile of anti-PD-L1/TGF β Trap in participants (*e.g.*, participants in safety part of the study) is evaluated in terms of AUC_{0-t} , $AUC_{0-\infty}$, C_{max} , t_{max} , and $t_{1/2}$. [AUC_{0-t} = The area under the concentration-time curve (AUC) from time zero (= dosing time) to the last sampling time (t_{last}) at which the concentration is at or above the lower limit of quantification. Calculated using the mixed log-linear trapezoidal rule (linear up, log down); $AUC_{0-\infty}$ = The AUC from time zero (dosing time) extrapolated to infinity, based on the predicted value for the concentration at t_{last} , as estimated using the linear regression from λz determination. $AUC_{0-\infty} = AUC_{0-t} + C_{\text{last}} \text{ pred} / \lambda z$; C_{max} = Maximum serum concentration observed post-dose].

[0279] *Immunogenicity:* Immunogenicity of PD-L1/TGF β Trap is measured by antidrug antibody (ADA) assay, from screening through the last safety follow-up visit.

[0280] *Efficacy Assessments:* Tumor response to anti-PD-L1/TGF β Trap is assessed by CT scan or MRI. Scans performed at baseline are repeated at subsequent visits. In general, lesions detected at baseline are followed using the same imaging methodology and preferably the same imaging equipment at subsequent tumor evaluation visits. Skin metastasis can be used as target lesions according to RECIST 1.1 using measurements by caliper, if they fulfill RECIST 1.1 for target lesions.

[0281] *Potential markers of clinical response in blood and tumor:* Several potential markers of clinical response from blood and tumor can be associated with clinical outcome. For example: tumor mutational burden (TMB) in plasma and in tumor tissue, level of circulating tumor DNA (CtDNA) in plasma, genes or gene signatures assessed in tumors, and/or level of PD-L1

expression accessed with Immunohistochemistry (IHC) in tumors are associated with clinical outcome.

[0282] *Results:* Objective tumor response is evaluated by the overall response rate (ORR), defined as the number of participants having reached a best overall response (BOR) of complete response (CR) or partial response (PR) divided by the number of participants in the analysis population. Progression-free survival is defined as the time from randomization to the date of the first documentation of objective progression of disease (PD) as assessed according to RECIST 1.1 or death due to any cause, whichever occurs first. It is contemplated that treatment with anti-PD-L1/TGF β Trap in combination with systemic chemotherapeutic agents results in initial clinical activity in treatment naïve, advanced NSCLC patients. Treated patients exhibit disease response (*e.g.*, partial response, complete response, stable disease) and/or improved survival (*e.g.*, progression-free survival and/or overall survival). It is contemplated that treatment with anti-PD-L1/TGF β Trap in combination with systemic chemotherapeutic agents followed by anti-PD-L1/TGF β Trap consolidation treatment results in superior survival of treatment naïve, advanced NSCLC patients or patients who have received and failed platinum-based chemotherapy and anti-PD-1 or anti-PD-L1 as monotherapy compared to systemic chemotherapy alone.

[0283] In summary, anti-PD-L1/TGF β Trap is found to be an innovative first-in-class bifunctional fusion protein designed to simultaneously target 2 immune suppressive pathways: PD-L1 and TGF- β , which when administrated in combination with systemic chemotherapeutic agents improves the treatment of advanced NSCLC.

SEQUENCES

SEQ ID NO: 1

Peptide sequence of the secreted anti-PD-L1 lambda light chain

25 QSALTQPASVSGSPGQSITISCTGTSSDVGGYNYVSWYQQHPGKAPKLMIYDVSNRPSG
VSNRFSGSKSGNTASLTISGLQAEDEADYYCSSYTSSSTRVFGTGTKVTVLGQPKANPTV
TLFPPSSEELQANKATLVCLISDFYPGAVTVAWKADGSPVKAGVETTKPSKQSNKYAA
SSYLSLTPEQWKSHRSYSCQVTHEGSTVEKTVAPTECS

SEQ ID NO: 2

Peptide sequence of the secreted H chain of anti-PDL1

EVQLLES GGGLVQPGGSLRLSCAASGFTFSSYIMMWVRQAPGKGLEWVSSIYPSGGITF
 YADTVKGRFTISRDN SKNTLYLQMNSLRAEDTAVYYCARIKLGTVTTVDYWGQGT LVT
 5 VSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVL
 QSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRV EPKSCDKTHTCPPCPAPEL
 LGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPR
 EEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYT
 LPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKL
 10 TVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

SEQ ID NO: 3

Peptide sequence of the secreted H chain of anti-PDL1/TGFβ Trap

EVQLLES GGGLVQPGGSLRLSCAASGFTFSSYIMMWVRQAPGKGLEWVSSIYPSGGITF
 15 YADTVKGRFTISRDN SKNTLYLQMNSLRAEDTAVYYCARIKLGTVTTVDYWGQGT LVT
 VSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVL
 QSSGLYSLSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRV EPKSCDKTHTCPPCPAPEL
 LGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPR
 EEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYT
 20 LPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKL
 TVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGAGGGGSGGGGSGGGGSGGGGS
 GIPPHVQKSVNNDMIVTDNNGAVKFPQLCKFCDFRSTCDNQKSCMSNCSITSICEKPQE
 VCVAVWRKNDENITLETVCHDPKLPYHDFILED AASPKCIMKEKKKPGETFFMCSCSSD
 ECNDNIIFSEEYNTSNPD

25

SEQ ID NO: 4

DNA sequence from the translation initiation codon to the translation stop codon of the anti-PD-L1 lambda light chain (the leader sequence preceding the VL is the signal peptide from urokinase plasminogen activator)

30 atgagggccctgctggctagactgctgctgtgcgtgctgtccgacagcaagggcCAGTCCGCCCTGACCCAGC
 CTGCCTCCGTGTCTGGCTCCCCTGGCCAGTCCATCACCATCAGCTGCACCGGCACCT
 CCAGCGACGTGGGCGGCTACAAC TACGTGTCCTGGTATCAGCAGCACCCCGGCAAG

GCCCCAAGCTGATGATCTACGACGTGTCCAACCGGCCCTCCGGCGTGTCCAACAG
 ATTCTCCGGCTCCAAGTCCGGCAACACCGCCTCCCTGACCATCAGCGGACTGCAGGC
 AGAGGACGAGGCCGACTACTACTGCTCCTCTACACCTCCTCCAGCACCAGAGTGTT
 CGGCACCGGCACAAAAGTGACCGTGCTGggccagcccaaggccaaccaaccgtgacactgttcccccatc
 5 ctccgaggaactgcagccaacaaggccaccctggtctgctgatctcagatttctatccaggcgcgtgaccgtggcctggaaggctgat
 gggtccccagtgaaggccggcgtggaaccaccaagccctccaagcagtcacaacaatacgccgcctcctctacgtgcctgac
 ccccgagcagtggaagtcaccggctctacagctgccaggtcacacagagggtccaccgtggaagaccgtgccccaccgag
 tgctcaTGA

10 SEQ ID NO: 5

DNA sequence from the translation initiation codon to the translation stop codon (mVK SP
 leader: small underlined; VH: capitals; IgG1m3 with K to A mutation: small letters; (G4S)x4-G
 (SEQ ID NO: 11) linker: bold capital letters; TGFβRII: bold underlined small letters; two stop
 codons: bold underlined capital letters)

15 atggaacagacacacctgctgctgtgggtgctgctgctgtgggtgccccggctccacaggcGAGGTGCAGCTGCTGGAAT
 CCGGCGGAGGACTGGTGCAGCCTGGCGGCTCCCTGAGACTGTCTTGCGCCGCTCCG
 GCTTCACCTTCTCCAGCTACATCATGATGTGGGTGCGACAGGCCCTGGCAAGGGCC
 TGAATGGGTGTCCTCCATCTACCCCTCCGGCGGCATCACCTTCTACGCCGACACCG
 TGAAGGGCCGGTTCACCATCTCCCGGGACAACCTCCAAGAACACCCTGTACCTGCAG
 20 ATGAACTCCCTGCGGGCCGAGGACACCGCCGTGTACTACTGCGCCCGGATCAAGCT
 GGGCACCGTGACCACCGTGGACTACTGGGGCCAGGGCACCTGGTGACAGTGTCTCT
 CCgtagcaccaaggccccatcggtcttccccctggcacctctccaagagcacctctgggggcacagcggccctgggtgctgctgt
 caaggactacttccccgaaccggtgacgggtgctggaactcaggcgccctgaccagcggcgtgcacaccttcccggtgtcctacagtc
 ctcaggacttactccctcagcagcgtggtgacctgcccctcagcagcttgggcacccagacctacatctgcaactgaatcacaagccc
 25 agcaacaccaaggtggacaagagagttgagcccaaatctgtgacaaaactcacacatgccaccgtgccagcacctgaactctggg
 gggaccgtcagtcttcttccccccaaaaccaaggacacctcatgatctccggaccttgaggtcacatgcgtggtggtggacgtg
 agccacgaagacctgaggtcaagttcaactggtacgtggacggcgtggaggtgcataatgccaagacaagccgcgggaggagcagt
 acaacagcagctaccgtgtggtcagcgtcctcaccgtcctgcaccaggactggctgaatggcaaggagtacaagtgaaggtctccaaca
 aagccctcccagcccccatcgagaaaaccatctcaaaagccaaaggcagccccgagaaccacaggtgtacacctgccccatcccc
 30 ggaggagatgaccaagaaccaggtcagcctgacctgctggtcaaaagcttctatccagcgacatcgccgtggagtgaggagagcaatg
 ggcagccgggagaacaactacaagaccacgcctcccgctggtgactccgacggctccttctctctatagcaagctcaccgtggacaaga
 gcaggtggcagcaggggaacgtcttctcatgctccgtgatgcatgaggctctgcacaaccactacacgcagaagagcctctccctgtcccc

gggtgctGGCGGCGGAGGAAGCGGAGGAGGTGGCAGCGGTGGCGGTGGCTCCGG
 CGGAGGTGGCTCCGGAAatccctccccacgtgcagaagtcctgaacaacgacatgacgtgaccgacaacaacg
gcgccgtgaagtccctcagctgtgcaagttctgcgacgtgaggttcagcacctgcgacaaccagaagtctgcatgagcaactgc
agcatcacaagcatctgcgagaagccccaggaggtgtgtgtggccgtgtggaggaagaacgacgaaaacatcacctcgagacc
 5 gtgtgcatgaccccaagctgccctaccacgacttcactcctggaagacgccgcctcccccaagtgcacatgaaggagaagaaga
agccccggcgagaccttctcatgtgcagctgcagcagcgacgagtgcaatgacaacatcatcttagcgaggagtacaacaccag
caaccccgacTGATAA

SEQ ID NO: 6

- 10 Polypeptide sequence of the secreted lambda light chain of anti-PD-L1(mut)/ TGFβ Trap, with
 mutations A31G,D52E,R99Y
 QSALTQPASVSGSPGQSITISCTGTSSDVGGYNYVSWYQQHPGKAPKLMIYEVSNRPSG
 VSNRFSGSKSGNTASLTISGLQAEDEADYYCSSYTSSSTYVFGTGTKVTVLGQPKANPTV
 TLFPPSSEELQANKATLVCLISDFYPGAVTVAWKADGSPVKAGVETTKPSKQSNNKYAA
 15 SSYLSLTPEQWKSHRSYSCQVTHEGSTVEKTVAPTECS

SEQ ID NO: 7

- Polypeptide sequence of the secreted heavy chain of anti-PD-L1(mut)/ TGFβ Trap
 EVQLLESGLLVQPGSLRLSCAASGFTFSMYMMMWVRQAPGKGLEWVSSIYPSGGIT
 20 FYADSVKGRFTISRDN SKNTLYLQMNSLRAEDTAIYYCARIKLGTVTTVDYWGQGTLLV
 TVSSASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAV
 LQSSGLYSLSSVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPE
 LLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKP
 REEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVY
 25 TLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSK
 LTVDKSRWQQGNVFCFSVMHEALHNHYTQKSLSLSPGAGGGGSGGGGSGGGGSGGGG
 SGIPPHVQKSVNNDMIVTDNNGAVKFPQLCKFCDVRFSTCDNQKSCMSNCSITSICEKPQ
 EVCVAVWRKNDENITLETVCHDPKLPYHDFILEDAAAPKCMKEKKKPGETFFMCSCSS
 DECNDNIIFSEYNTSNPD

SEQ ID NO: 8

Human TGF β RII Isoform A Precursor Polypeptide (NCBI RefSeq Accession No: NP_001020018)

5 MGRGLLRGLWPLHIVLWTRIASTIPPHVQKSDVEMEAQKDEIICPSCNRTAHPLRHINND
 MIVTDNNGAVKFPQLCKFCDFRSTCDNQKSCMSNCSITSICEKPQEVCAVWRKNDE
 NITLETVCHDPKLPYHDFILEDAAAPKCMKEKKKKPGETFFMCSCSSDECNDNIIFSEEYN
 TSNPDLLLVIQVGTGISLLPPLGVAISVIIIIFYCYRVNRQQKLSSTWETGKTRKLMEFSEHC
 AIIEDDRSDISSTCANNINHNTPELLDLVGKGRFAEVYKAKLKQNTSEQFETVAVK
 10 IFPYEEYASWKTEKDIFSDINLKHENILQFLTAERKTELGKQYWLITAFHAKGNLQEYL
 TRHVISWEDLRKLGSSSLARGIAHLSDHTPCGRPKMPIVHRDLKSSNILVKNDLTCCLCD
 FGLSLRLDPTLSVDDLANSQGVTARYMAPEVLESRMNLENVESFKQTDVYSMALVL
 WEMTSRCNAVGEVKDYEPFSGSKVREHPCVESMKDNVLRDRGRPEIPSWLNHQGIQM
 VCETLTECWDHDPPEARLTAQCVAERFSELEHLDRLSGRSCSEEKIPEDGSLNTTK

15

SEQ ID NO: 9

Human TGF β RII Isoform B Precursor Polypeptide (NCBI RefSeq Accession No: NP_003233)

MGRGLLRGLWPLHIVLWTRIASTIPPHVQKSVNNDMIVTDNNGAVKFPQLCKFCDFRFS
 TCDNQKSCMSNCSITSICEKPQEVCAVWRKNDENITLETVCHDPKLPYHDFILEDAAAP
 20 KCIMKEKKKKPGETFFMCSCSSDECNDNIIFSEEYNTSNPDLLLVIQVGTGISLLPPLGVAIS
 VIIIIFYCYRVNRQQKLSSTWETGKTRKLMEFSEHCAIIEDDRSDISSTCANNINHNTPELL
 IELDLVGKGRFAEVYKAKLKQNTSEQFETVAVKIFPYEEYASWKTEKDIFSDINLKHEN
 ILQFLTAERKTELGKQYWLITAFHAKGNLQEYLTRHVISWEDLRKLGSSSLARGIAHLSD
 DHTPCGRPKMPIVHRDLKSSNILVKNDLTCCLCDFGLSLRLDPTLSVDDLANSQGVTAR
 25 RYMAPEVLESRMNLENVESFKQTDVYSMALVLWEMTSRCNAVGEVKDYEPFSGSKVR
 EHPCVESMKDNVLRDRGRPEIPSWLNHQGIQMVCELTETECWDHDPPEARLTAQCVAER
 FSELEHLDRLSGRSCSEEKIPEDGSLNTTK

SEQ ID NO: 10

A Human TGF β RII Isoform B Extracellular Domain Polypeptide

IPPHVQKSVNNDMIVTDNNGAVKFPQLCKFCDVRFSTCDNQKSCMSNCSITSICEKPQEV
CVAVWRKNDENITLETVCHDPKLPYHDFILEDAAAPKCIMKEKKKKPGETFFMCSCSSDE
5 CNDNIIFSEEYNTSNPD

SEQ ID NO: 11

(Gly₄Ser)₄Gly linker

GGGGSGGGGSGGGGSGGGGSG

10

SEQ ID NO: 12

Polypeptide sequence of the secreted heavy chain variable region of anti-PD-L1 antibody

MPDL3289A

EVQLVESGGGLVQPGGSLRLSCAASGFTFSDSWIHWVRQAPGKGLEWVAWISPYGGST
15 YYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCARRHWPGGFDYWGQGTSLV
VSS

SEQ ID NO: 13

Polypeptide sequence of the secreted light chain variable region of anti-PD-L1 antibody

20 MPDL3289A

DIQMTQSPSSLSASVGDRVTITCRASQDVSTAVAWYQQKPGKAPKLLIYSASFLYSGVPS
RFGSGSGTDFTLTISSLQPEDFATYYCQQYLYHPATFGQGTKVEIKR

SEQ ID NO: 14

Polypeptide sequence of the secreted heavy chain variable region of anti-PD-L1 antibody

25 YW243.55S70

EVQLVESGGGLVQPGGSLRLSCAASGFTFSDSWIHWVRQAPGKGLEWVAWISPYGGST
YYADSVKGRFTISADTSKNTAYLQMNSLRAEDTAVYYCARRHWPGGFDYWGQGTSLV
VSA

SEQ ID NO: 50A Truncated Human TGF β RII Isoform B Extracellular Domain Polypeptide

GAVKFPQLCKFCDVRFSTCDNQKSCMSNCSITSICEKPQEVCAVWRKNDENITLETVC
HDPKLPYHDFILEDAAAPK CIMKEKKKPGETFFMCSCSSDECNDNIIFSEEYNTSNPD

5

SEQ ID NO: 51A Truncated Human TGF β RII Isoform B Extracellular Domain Polypeptide

VKFPQLCKFCDVRFSTCDNQKSCMSNCSITSICEKPQEVCAVWRKNDENITLETVCHD
PKLPYHDFILEDAAAPK CIMKEKKKPGETFFMCSCSSDECNDNIIFSEEYNTSNPD

10

SEQ ID NO: 52A Truncated Human TGF β RII Isoform B Extracellular Domain Polypeptide

VTDNNGAVKFPQLCKFCDVRFSTCDNQKSCMSNCSITSICEKPQEVCAVWRKNDENIT
LETVCHDPKLPYHDFILEDAAAPK CIMKEKKKPGETFFMCSCSSDECNDNIIFSEEYNTSN

15 PD

SEQ ID NO: 53A Truncated Human TGF β RII Isoform B Extracellular Domain Polypeptide

LCKFCDVRFSTCDNQKSCMSNCSITSICEKPQEVCAVWRKNDENITLETVCHDPKLPY
HDFILEDAAAPK CIMKEKKKPGETFFMCSCSSDECNDNIIFSEEYNTSNPD

20

SEQ ID NO: 54A Mutated Human TGF β RII Isoform B Extracellular Domain Polypeptide

VTDNAGAVKFPQLCKFCDVRFSTCDNQKSCMSNCSITSICEKPQEVCAVWRKNDENIT
LETVCHDPKLPYHDFILEDAAAPK CIMKEKKKPGETFFMCSCSSDECNDNIIFSEEYNTSN

25 PD

SEQ ID NO: 55Polypeptide sequence of the heavy chain variable region of anti-PD-L1 antibody

QVQLQESGPGLVKPSQTLSTCTVSGGISNDYWTWIRQHPGKGLYIGYISYTGSTYYN
PSLKSRVTISRDTSKNQFSLKLSSVTAADTAVYYCARSGGWLAPFDYWGRGTLTVSS

30

SEQ ID NO: 56

Polypeptide sequence of the light chain variable region of anti-PD-L1 antibody

DIVMTQSPDSLAVSLGERATINCKSSQSLFYHSNQKHS LAWYQQKPGQPPKLLIYGAST
RESGVPDRFSGSGSGTDFTLTIS SLQAEDVAVYYCQYYGYPTYTFGGGTKVEIK

5

SEQ ID NO: 57

Polypeptide sequence of the heavy chain variable region of anti-PD-L1 antibody

QVQLVQSGAEVKKPGASVKV SCKASGYTFTSYWMHWVRQAPGQGLEWMGRIGPNSG
FTSYNEKFKNRVTMTRDTSTSTVYME LSSLRSEDTAVYYCARGGSSYDYFDYWGQGT

10 VTVSS

SEQ ID NO: 58

Polypeptide sequence of the light chain variable region of anti-PD-L1 antibody

DIVLTQSPASLAVSPGQRATITCRASESVSIHGTHLMHWYQQKPGQPPKLLIYAASNLES
GVPARFSGSGSGTDFTLTINPVEAEDTANYYCQQSFEDPLTFGQGTKLEIK

15

SEQ ID NO: 59

Polypeptide sequence of the heavy chain of anti-PD-L1 antibody

QVQLQESGPGLVKPSQTL SLTCTVSGGSISNDYWTWIRQHPGKGLE YIGYISYTGSTYYN
PSLKS RVTISRDTSKNQFSLKLSSVTAADTAVYYCARSGGWLAPFDYWG RGT LVTVSSA

20 STKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSG

LYSLSSVVTVPSSSLG TKTYTCNV DHKPSNTKVDKRVESKYGP PCPPCAPEAAGGPSVF
LFPPKPKD TLMISRTPEVTCVVVDVSQEDPEVQFNWYVDGVEVHNAKTKPREEQFNST

YRVVSVLTVLHQDWLNGKEYKCKVSNKGLPSSIEKTISKAKGQPREPQVYTLPPSQEEM

TKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSRLTVDKSRW

25 QEGNVFSCSVMHEALHNHYTQKSLSLGLK

SEQ ID NO: 60

Polypeptide sequence of the light chain of anti-PD-L1 antibody

DIVMTQSPDSLAVSLGERATINCKSSQSLFYHSNQKHS LAWYQQKPGQPPKLLIYGAST
RESGVPDRFSGSGSGTDFTLTIS SLQAEDVAVYYCQYYGYPTYTFGGGTKVEIKRTVAA

30 PSVFIFPPSDEQLKSGTASVVC LLNFPYPREAKVQWKVDNALQSGNSQESVTEQDSKDS

TYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFNRGEC

SEQ ID NO: 61Polypeptide sequence of the heavy chain of anti-PD-L1 antibody

QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYWMHWVRQAPGQGLEWMGRIGPNSG
 FTSYNEKFKNRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARGGSSYDYFDYWGQGT
 5 VTVSSASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPA
 VLQSSGLYSLSSVVTVPSSSLGTCTYTCNVDHKPSNTKVDKRVESKYGPPCPPCPAPEAA
 GGPSVFLFPPKPKDTLMISRTPEVTCVVDVDSQEDPEVQFNWYVDGVEVHNAKTKPRE
 EQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKGLPSSIEKTISKAKGQPREPQVYTL
 PSQEEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSRLT
 10 VDKSRWQEGNVFSCSVMHEALHNHYTQKSLSLGLA

SEQ ID NO: 62Polypeptide sequence of the light chain of anti-PD-L1 antibody

DIVLTQSPASLAVSPGQRATITCRASESVSIHGTHLMHWYQQKPGQPPKLLIYAASNLES
 15 GVPARFSGSGSGTDFTLTINPVEAEDTANYCYQQSFEDPLTFGQGTKLEIKRTVAAPSVFI
 FPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYSLS
 STLTLTKADYEEKHKVYACEVTHQGLSSPVTKSFNRGEC

INCORPORATION BY REFERENCE

20 **[0284]** The entire disclosure of each of the patent documents and scientific articles referred to
 herein is incorporated by reference for all purposes.

EQUIVALENTS

[0285] The disclosure may be embodied in other specific forms without departing from the
 spirit or essential characteristics thereof. The foregoing embodiments are therefore to be
 considered in all respects illustrative rather than limiting the disclosure described herein. Various
 25 structural elements of the different embodiments and various disclosed method steps may be
 utilized in various combinations and permutations, and all such variants are to be considered
 forms of the disclosure. Scope of the disclosure is thus indicated by the appended claims rather
 than by the foregoing description, and all changes that come within the meaning and range of
 equivalency of the claims are intended to be embraced therein.

CLAIMS

WHAT IS CLAIMED IS:

1. A method of treating advanced non-small cell lung cancer (NSCLC) or inhibiting
5 NSCLC tumor growth in a treatment naïve patient in need thereof, the method comprising a first step of administering to the patient a dose of at least 1800 mg of a protein comprising a first polypeptide and a second polypeptide, with concurrent systemic chemotherapy, and a second step comprising administering at least 1800 mg of the protein,

wherein the first polypeptide comprises: (a) at least a variable region of a heavy chain of
10 an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGF β RII), or a fragment thereof, capable of binding Transforming Growth Factor β (TGF β),

wherein the second polypeptide comprises at least a variable region of a light chain of an antibody that binds PD-L1, and

15 wherein the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1.
2. The method of claim 1, wherein the first polypeptide comprises the amino acid sequence of SEQ ID NO: 3, and the second polypeptide comprises the amino acid sequence of SEQ ID NO: 1.
- 20 3. The method of claim 1 or 2, wherein the dose is 1800 mg to 3000 mg.
4. The method of any one of claims 1-3, wherein the dose is 2100 mg to 2400 mg.
5. The method of any one of claims 1-4, wherein the dose is 2100 mg.
6. The method of any one of claims 1-4, wherein the dose is 2400 mg.
7. The method of any one of claims 1-4, wherein the protein is administered once every
25 three weeks.
8. The method of claim 7, wherein the dose is 2100 mg, administered once every three weeks.

9. The method of claim 7, wherein the dose is 2400 mg, administered once every three weeks.
10. The method of any one of claims 1-3, wherein the dose is 3000 mg, administered once every three weeks.
- 5 11. The method of any one of claims 1-10, wherein the systemic chemotherapy comprises carboplatin or cisplatin.
12. The method of claim 11, wherein the systemic chemotherapy comprises carboplatin administered once every three weeks.
13. The method of claim 12, wherein the carboplatin is administered AUC 4 to AUC 6 IV on
10 day 1 every 21 days for at least four cycles.
14. The method of claim 11, wherein the systemic chemotherapy comprises cisplatin.
15. The method of claim 14, wherein the cisplatin is administered 75-100 mg/m² IV on day 1 every 21 days for at least four cycles.
16. The method of any one of claims 11-15, wherein the systemic chemotherapy comprises
15 concurrently administering gemcitabine or paclitaxel to the patient.
17. The method of claim 16, wherein the systemic chemotherapy comprises concurrently administering gemcitabine to the patient.
18. The method of claim 17, wherein the gemcitabine is administered 1000-1250 mg/m² IV on days 1 and 8 every 21 days for at least four cycles.
- 20 19. The method of claim 16, wherein the systemic chemotherapy comprises concurrently administering paclitaxel to the patient.
20. The method of claim 19, wherein the paclitaxel is administered 175-225 mg/m² IV on day 1 every 21 days for at least four cycles.
21. The method of claim 19, wherein the paclitaxel is administered albumin-bound 100
25 mg/m² on days 1, 8, and 15 every 21 days for at least four cycles.

22. The method of any one of claims 1-21, wherein the second step further comprises administering a systemic chemotherapeutic agent.
23. The method of any one of claims 1-22, wherein the NSCLC is squamous NSCLC.
24. The method of any one of claims 1-22, wherein the NSCLC is non-squamous NSCLC.
- 5 25. A method of treating advanced non-squamous non-small cell lung cancer (NSCLC) or inhibiting non-squamous NSCLC tumor growth in a treatment naïve patient in need thereof, the method comprising a first step of administering to the patient a dose of at least 1800 mg of a protein comprising a first polypeptide and a second polypeptide, with concurrent systemic chemotherapy comprising pemetrexed, and a second step of administering at least 1800 mg of
10 the protein in combination with systemic chemotherapy consisting of pemetrexed to the patient,
- wherein the first polypeptide comprises: (a) at least a variable region of a heavy chain of an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGF β RII), or a fragment thereof, capable of binding Transforming Growth Factor β (TGF β),
- 15 wherein the second polypeptide comprises at least a variable region of a light chain of an antibody that binds PD-L1, and
- wherein the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1.
26. The method of claim 25, wherein the first polypeptide comprises the amino acid
20 sequence of SEQ ID NO: 3, and the second polypeptide comprises the amino acid sequence of SEQ ID NO: 1.
27. The method of claim 25 or 26, wherein the dose is 1800 mg to 3000 mg.
28. The method of any one of claims 25-27, wherein the dose is 2100 mg to 2400 mg.
29. The method of any one of claims 25-28, wherein the dose is 2100 mg.
- 25 30. The method of any one of claims 25-28, wherein the dose is 2400 mg.

31. The method of any one of claims 25-28, wherein the protein is administered once every three weeks.
32. The method of claim 31, wherein the dose is 2100 mg, administered once every three weeks.
- 5 33. The method of claim 31, wherein the dose is 2400 mg, administered once every three weeks.
34. The method of any one of claims 25-27, wherein the dose is 3000 mg, administered once every three weeks.
35. The method of any one of claims 25-34, wherein the systemic chemotherapy comprises
10 carboplatin or cisplatin administered once every three weeks.
36. The method of claim 35, wherein the systemic chemotherapy comprises carboplatin.
37. The method of claim 36, wherein the carboplatin is administered AUC 4 to AUC 6 IV on day 1 every 21 days for at least four cycles.
38. The method of claim 35, wherein the systemic chemotherapy comprises cisplatin.
- 15 39. The method of claim 38, wherein the cisplatin is administered 75 mg/m² IV on day 1 every 21 days for at least four cycles.
40. The method of any one of claims 25-39, wherein the pemetrexed is administered once every three weeks.
41. The method of any one of claims 25-40, wherein the pemetrexed is administered 500
20 mg/m² IV on day 1 every 21 days for at least four cycles.
42. The method of any one of claims 1-41, wherein the NSCLC is PD-L1 positive, or the NSCLC is not PD-L1 positive.
43. A method of treating advanced non-small cell lung cancer (NSCLC) or inhibiting
NSCLC tumor growth in patient indicated as having metastatic NSCLC disease progression on
25 previous treatment with an immunotherapy in combination with chemotherapy, or on previous
treatment with chemotherapy followed by treatment with an immunotherapy, or on previous

treatment with an immunotherapy followed by platinum-based chemotherapy, the method comprising a first step of administering to the patient a dose of at least 1800 mg of a protein comprising a first polypeptide and a second polypeptide, with concurrent systemic chemotherapy comprising docetaxel, and a second step of administering at least 1800 mg of the protein,

5 wherein the first polypeptide comprises: (a) at least a variable region of a heavy chain of an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGF β RII), or a fragment thereof, capable of binding Transforming Growth Factor β (TGF β),

 wherein the second polypeptide comprises at least a variable region of a light chain of an
10 antibody that binds PD-L1, and wherein the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1.

44. The method of claim 43, wherein the first polypeptide comprises the amino acid sequence of SEQ ID NO: 3, and the second polypeptide comprises the amino acid sequence of SEQ ID NO: 1.

15 45. The method of claim 43 or 44, wherein the dose is 1800 mg to 3000 mg.

46. The method of any one of claims 43-45, wherein the dose is 2100 mg to 2400 mg.

47. The method of any one of claims 43-46, wherein the dose is 2100 mg.

48. The method of any one of claims 43-46, wherein the dose is 2400 mg.

49. The method of any one of claims 43-46, wherein the protein is administered once every
20 three weeks.

50. The method of claim 49, wherein the dose is 2100 mg, administered once every three weeks.

51. The method of claim 49, wherein the dose is 2400 mg, administered once every three weeks.

25 52. The method of claim 45, wherein the dose is 3000 mg, administered once every three weeks.

53. The method of any one of claims 43-52, wherein the immunotherapy comprises an anti-PD-L1 or an anti-PD-1 antibody.
54. The method of any one of claims 43-53, wherein the docetaxel is administered 75-100 mg/m² IV on day 1 every 21 days for four cycles.
- 5 55. The method of any one of claims 43-54, wherein the NSCLC is squamous NSCLC.
56. The method of any one of claims 43-54, wherein the NSCLC is non-squamous NSCLC.
57. The method of any one of claims 1-56, wherein the treatment results in a disease response or improved survival of the patient.
58. The method of claim 57, wherein the disease response is a complete response, a partial
10 response, or a stable disease.
59. The method of claim 57, wherein the survival is progression-free survival (PFS).
60. The method of any one of claims 1-59, wherein the protein is administered by intravenous administration.
61. The method of claim 60, wherein the intravenous administration is performed with a
15 prefilled bag, a prefilled pen, or a prefilled syringe comprising a formulation comprising the protein.
62. The method of claim 61, wherein the bag is connected to a channel comprising a tube and/or a needle.
63. The method of any one of claims 1-62, wherein the second step is continued for 31 or
20 more cycles.
64. An anti-PD-L1/TGF β Trap protein comprising a first polypeptide and a second polypeptide for use in a method of treating advanced non-small cell lung cancer (NSCLC) or inhibiting NSCLC tumor growth in a treatment naïve patient in need thereof, the method comprising a first step of administering to the patient a dose of at least 1800 mg of the protein
25 with concurrent systemic chemotherapy, and a second step comprising administering at least 1800 mg of the protein,

wherein the first polypeptide comprises: (a) at least a variable region of a heavy chain of an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGF β RII), or a fragment thereof, capable of binding Transforming Growth Factor β (TGF β),

5 wherein the second polypeptide comprises at least a variable region of a light chain of an antibody that binds PD-L1, and

wherein the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1.

65. An anti-PD-L1/TGF β Trap protein comprising a first polypeptide and a second
10 polypeptide for use in a method of treating advanced non-squamous non-small cell lung cancer (NSCLC) or inhibiting non-squamous NSCLC tumor growth in a treatment naïve patient in need thereof, the method comprising a first step of administering to the patient a dose of at least 1800 mg of the protein with concurrent systemic chemotherapy comprising pemetrexed, and a second step of administering at least 1800 mg of the protein in combination with systemic chemotherapy
15 consisting of pemetrexed to the patient,

wherein the first polypeptide comprises: (a) at least a variable region of a heavy chain of an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGF β RII), or a fragment thereof, capable of binding Transforming Growth Factor β (TGF β),

20 wherein the second polypeptide comprises at least a variable region of a light chain of an antibody that binds PD-L1, and

wherein the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1.

66. An anti-PD-L1/TGF β Trap protein comprising a first polypeptide and a second
25 polypeptide for use in a method of treating advanced non-small cell lung cancer (NSCLC) or inhibiting NSCLC tumor growth in patient indicated as having metastatic NSCLC disease progression on previous treatment with an immunotherapy in combination with chemotherapy, or on previous treatment with chemotherapy followed by treatment with an immunotherapy, or

on previous treatment with an immunotherapy followed by platinum-based chemotherapy, the method comprising a first step of administering to the patient a dose of at least 1800 mg of the protein with concurrent systemic chemotherapy comprising docetaxel, and a second step of administering at least 1800 mg of the protein,

5 wherein the first polypeptide comprises: (a) at least a variable region of a heavy chain of an antibody that binds to human protein Programmed Death Ligand 1 (PD-L1); and (b) human Transforming Growth Factor β Receptor II (TGF β RII), or a fragment thereof, capable of binding Transforming Growth Factor β (TGF β),

 wherein the second polypeptide comprises at least a variable region of a light chain of an
10 antibody that binds PD-L1, and wherein the heavy chain of the first polypeptide and the light chain of the second polypeptide, when combined, form an antigen binding site that binds PD-L1.

67. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-66, wherein the first polypeptide comprises the amino acid sequence of SEQ ID NO: 3, and the second polypeptide comprises the amino acid sequence of SEQ ID NO: 1.

15 68. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-67, wherein the dose is 1800 mg to 3000 mg.

69. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-68, wherein the dose is 2100 mg to 2400 mg.

20 70. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-69, wherein the dose is 2100 mg.

71. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-69, wherein the dose is 2400 mg.

72. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-69, wherein the protein is administered once every three weeks.

25 73. The anti-PD-L1/TGF β Trap protein for use of claim 72, wherein the dose is 2100 mg, administered once every three weeks.

74. The anti-PD-L1/TGF β Trap protein for use of claim 72, wherein the dose is 2400 mg, administered once every three weeks.
75. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-68, wherein the dose is 3000 mg, administered once every three weeks.
- 5 76. The anti-PD-L1/TGF β Trap protein for use of claim 64 or 65, or any one of claims 67-75, which depend from claim 64 or 65, wherein the systemic chemotherapy comprises carboplatin or cisplatin.
77. The anti-PD-L1/TGF β Trap protein for use of claim 76, wherein the systemic chemotherapy comprises carboplatin administered once every three weeks.
- 10 78. The anti-PD-L1/TGF β Trap protein for use of claim 77, wherein the carboplatin is administered AUC 4 to AUC 6 IV on day 1 every 21 days for at least four cycles.
79. The anti-PD-L1/TGF β Trap protein for use of claim 76, wherein the systemic chemotherapy comprises cisplatin.
80. The anti-PD-L1/TGF β Trap protein for use of claim 79, wherein the cisplatin is
15 administered 75-100 mg/m² IV on day 1 every 21 days for at least four cycles.
81. The anti-PD-L1/TGF β Trap protein for use of any one of claims 76-80, which depends from claim 64, wherein the systemic chemotherapy comprises concurrently administering gemcitabine or paclitaxel to the patient.
82. The anti-PD-L1/TGF β Trap protein for use of claim 81, wherein the systemic
20 chemotherapy comprises concurrently administering gemcitabine to the patient.
83. The anti-PD-L1/TGF β Trap protein for use of claim 82, wherein the gemcitabine is administered 1000-1250 mg/m² IV on days 1 and 8 every 21 days for at least four cycles.
84. The anti-PD-L1/TGF β Trap protein for use of claim 81, wherein the systemic chemotherapy comprises concurrently administering paclitaxel to the patient.
- 25 85. The anti-PD-L1/TGF β Trap protein for use of claim 84, wherein the paclitaxel is administered 175-225 mg/m² IV on day 1 every 21 days for at least four cycles.

86. The anti-PD-L1/TGF β Trap protein for use of claim 84, wherein the paclitaxel is administered albumin-bound 100 mg/m² on days 1, 8, and 15 every 21 days for at least four cycles.
87. The anti-PD-L1/TGF β Trap protein for use of claim 64 or any one of claims 67-86,
5 which depends from claim 64, wherein the second step further comprises administering a systemic chemotherapeutic agent.
88. The anti-PD-L1/TGF β Trap protein for use of claim 64 or any one of claims 67-87, which depends from claim 64, wherein the NSCLC is squamous NSCLC.
89. The anti-PD-L1/TGF β Trap protein for use of claim 64 or any one of claims 67-87,
10 which depends from claim 64, wherein the NSCLC is non-squamous NSCLC.
90. The anti-PD-L1/TGF β Trap protein for use of claim 65 or any one of claims 67-80, which depends from claim 65, wherein the pemetrexed is administered once every three weeks.
91. The anti-PD-L1/TGF β Trap protein for use of claim 65, or any one of claims 67-80 or 90, which depends from claim 65, wherein the pemetrexed is administered 500 mg/m² IV on day 1
15 every 21 days for at least four cycles.
92. The anti-PD-L1/TGF β Trap protein for use of claim 64 or 65, or any one of claims 67-91, which depends from claim 64 or 65, wherein the NSCLC is PD-L1 positive, or the NSCLC is not PD-L1 positive.
93. The anti-PD-L1/TGF β Trap protein for use of claim 66 or any one of claims 67-75,
20 which depends from claim 66, wherein the immunotherapy comprises an anti-PD-L1 or an anti-PD-1 antibody.
94. The anti-PD-L1/TGF β Trap protein for use of claim 66 or any one of claims 67-75 or 93, which depends from claim 66, wherein the docetaxel is administered 75-100 mg/m² IV on day 1 every 21 days for four cycles.
- 25 95. The anti-PD-L1/TGF β Trap protein for use of claim 66 or any one of claims 67-75 or 93-94, which depends from claim 66, wherein the NSCLC is squamous NSCLC.

96. The anti-PD-L1/TGF β Trap protein for use of claim 66 or any one of claims 67-75 or 93-94, which depends from claim 66, wherein the NSCLC is non-squamous NSCLC.
97. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-96, wherein the treatment results in a disease response or improved survival of the patient.
- 5 98. The anti-PD-L1/TGF β Trap protein for use of claim 97, wherein the disease response is a complete response, a partial response, or a stable disease.
99. The anti-PD-L1/TGF β Trap protein for use of claim 97, wherein the survival is progression-free survival (PFS).
100. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-99, wherein the
10 protein is administered by intravenous administration.
101. The anti-PD-L1/TGF β Trap protein for use of claim 100, wherein the intravenous administration is performed with a prefilled bag, a prefilled pen, or a prefilled syringe comprising a formulation comprising the protein.
102. The anti-PD-L1/TGF β Trap protein for use of claim 101, wherein the bag is connected to
15 a channel comprising a tube and/or a needle.
103. The anti-PD-L1/TGF β Trap protein for use of any one of claims 64-102, wherein the second step is continued for 31 or more cycles.

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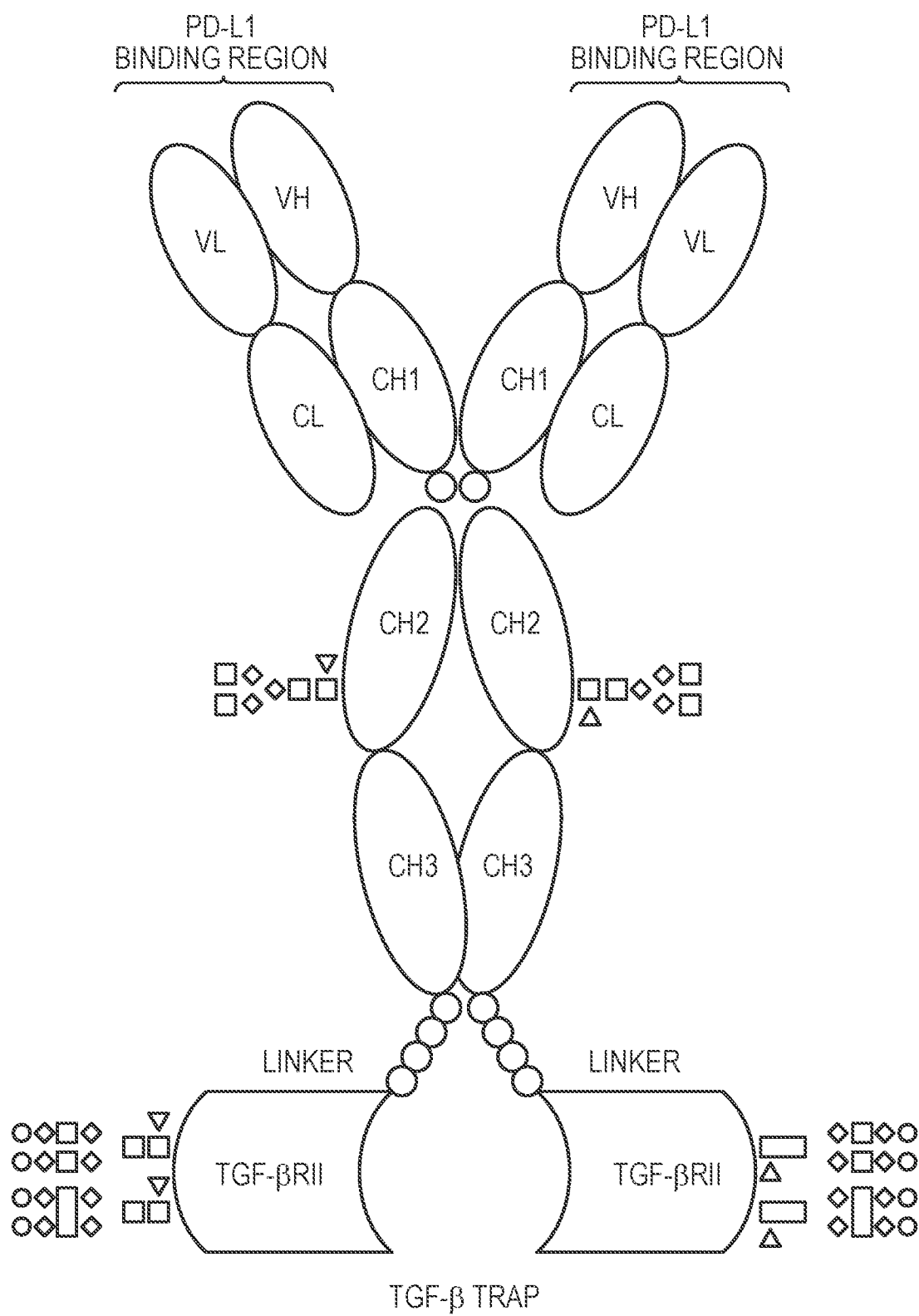


FIG. 1

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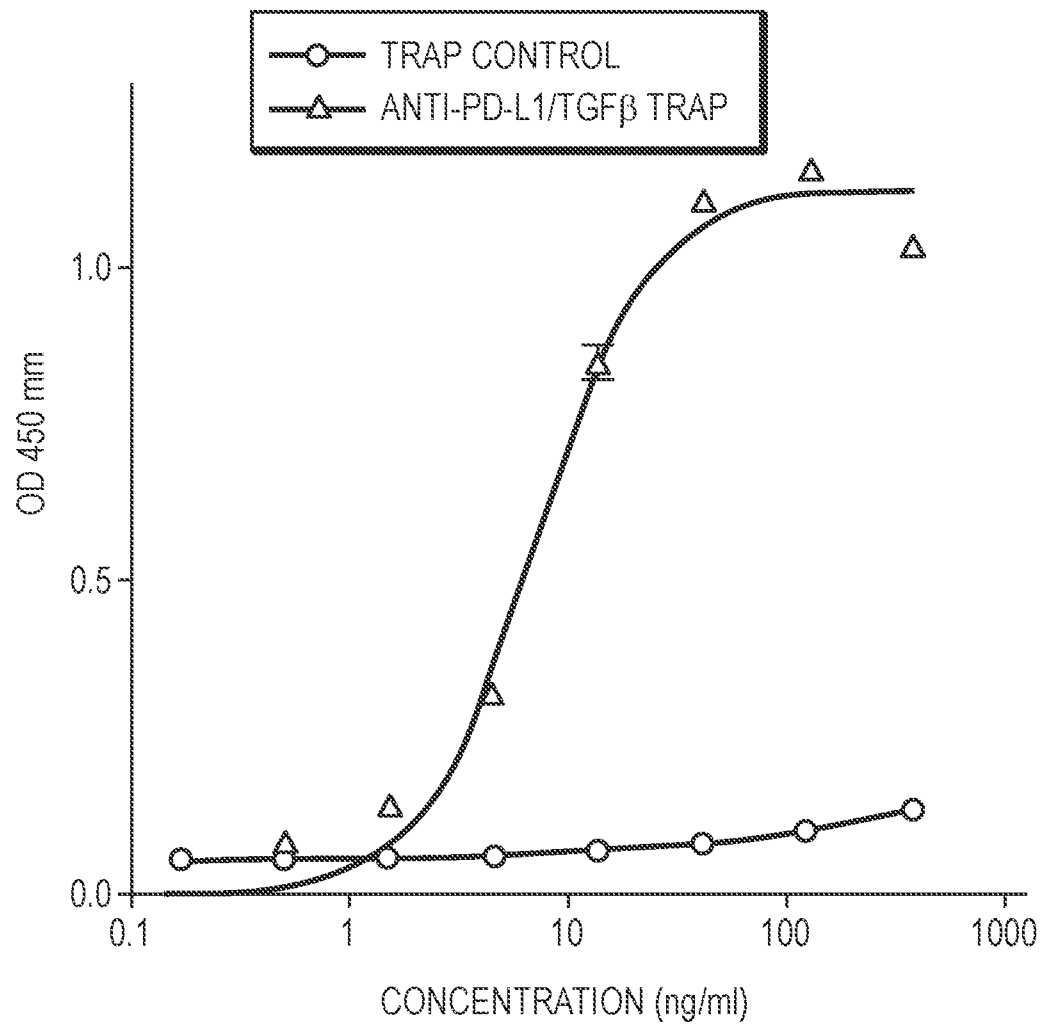


FIG. 2

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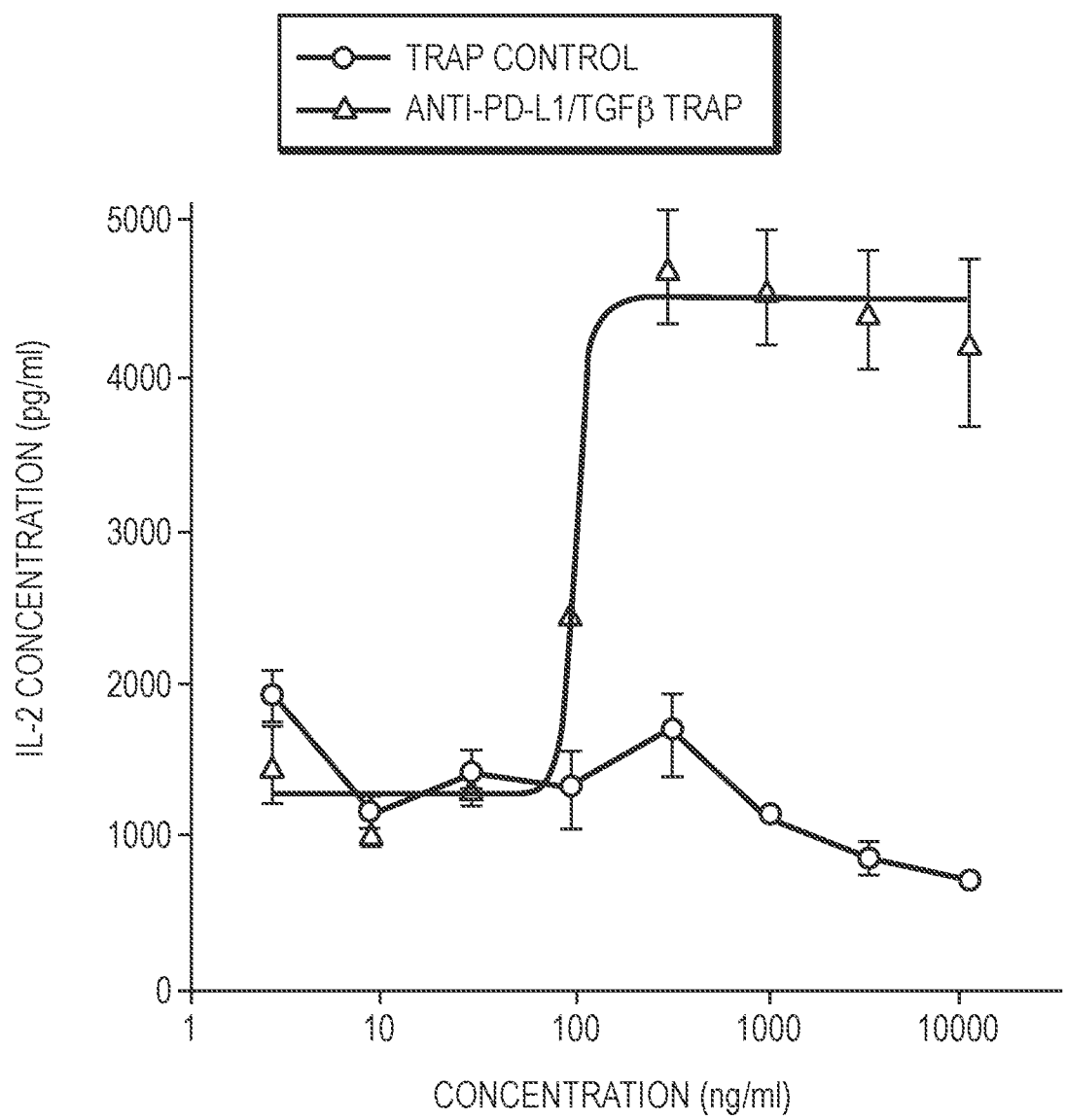


FIG. 3

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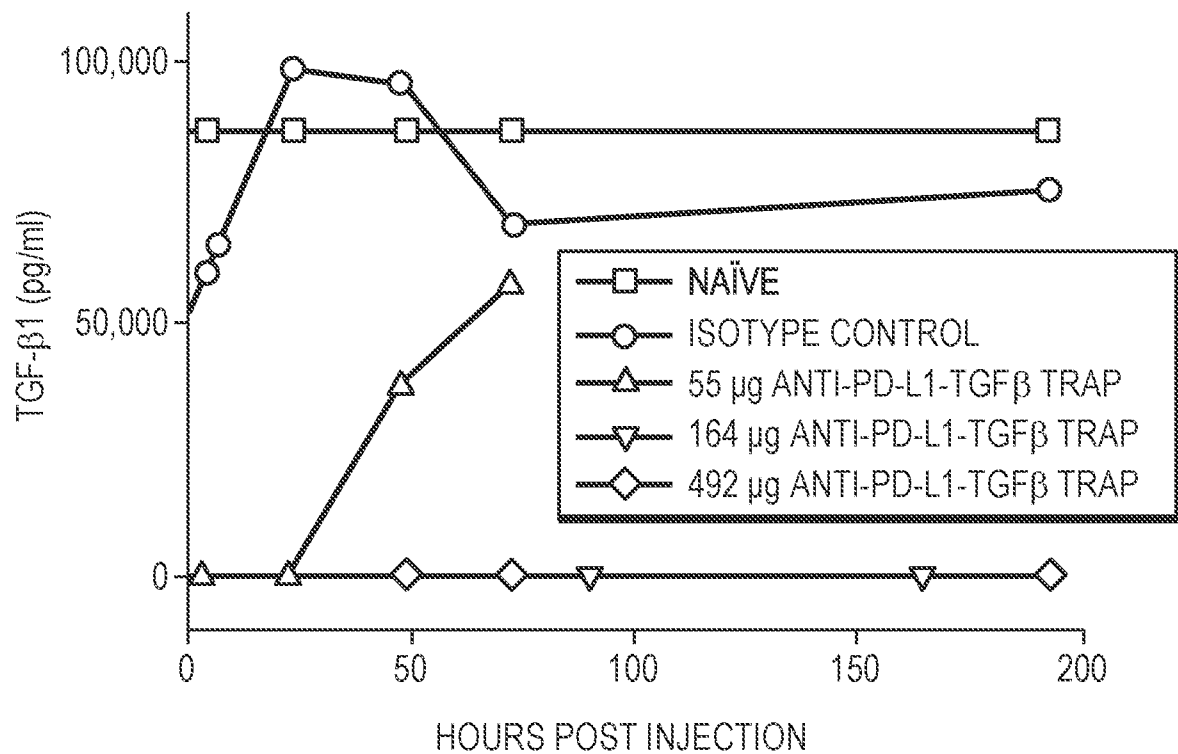


FIG. 4A

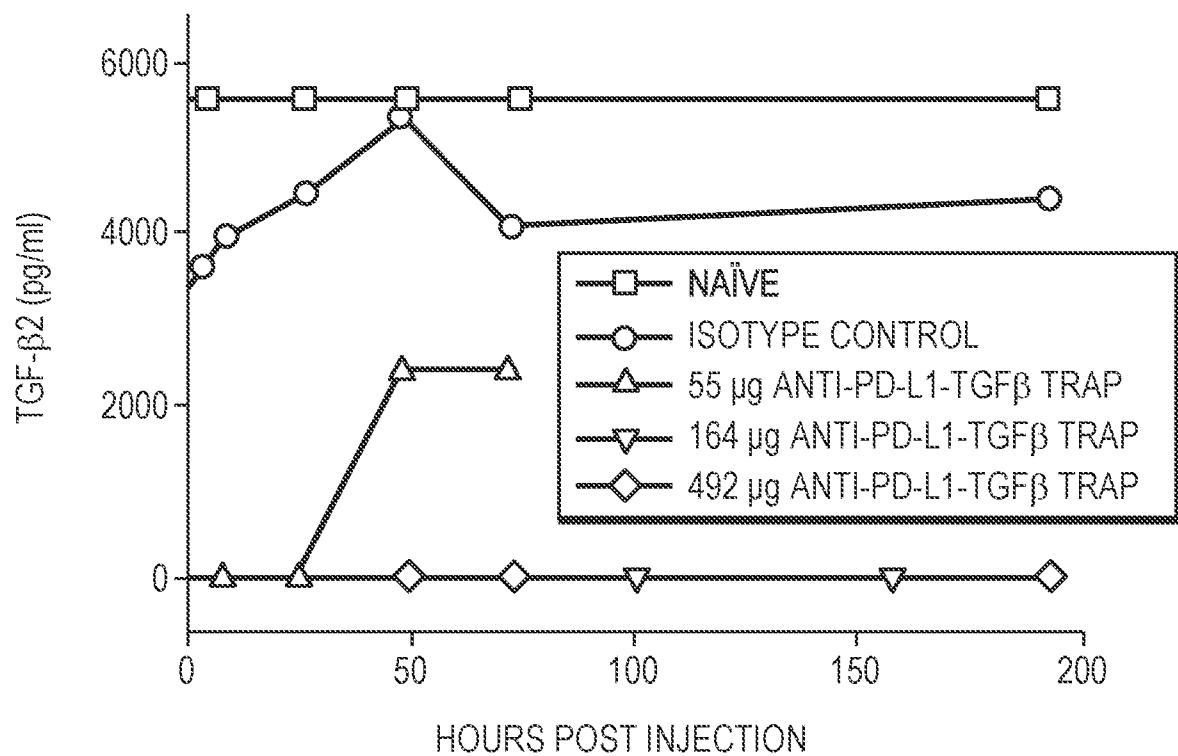


FIG. 4B

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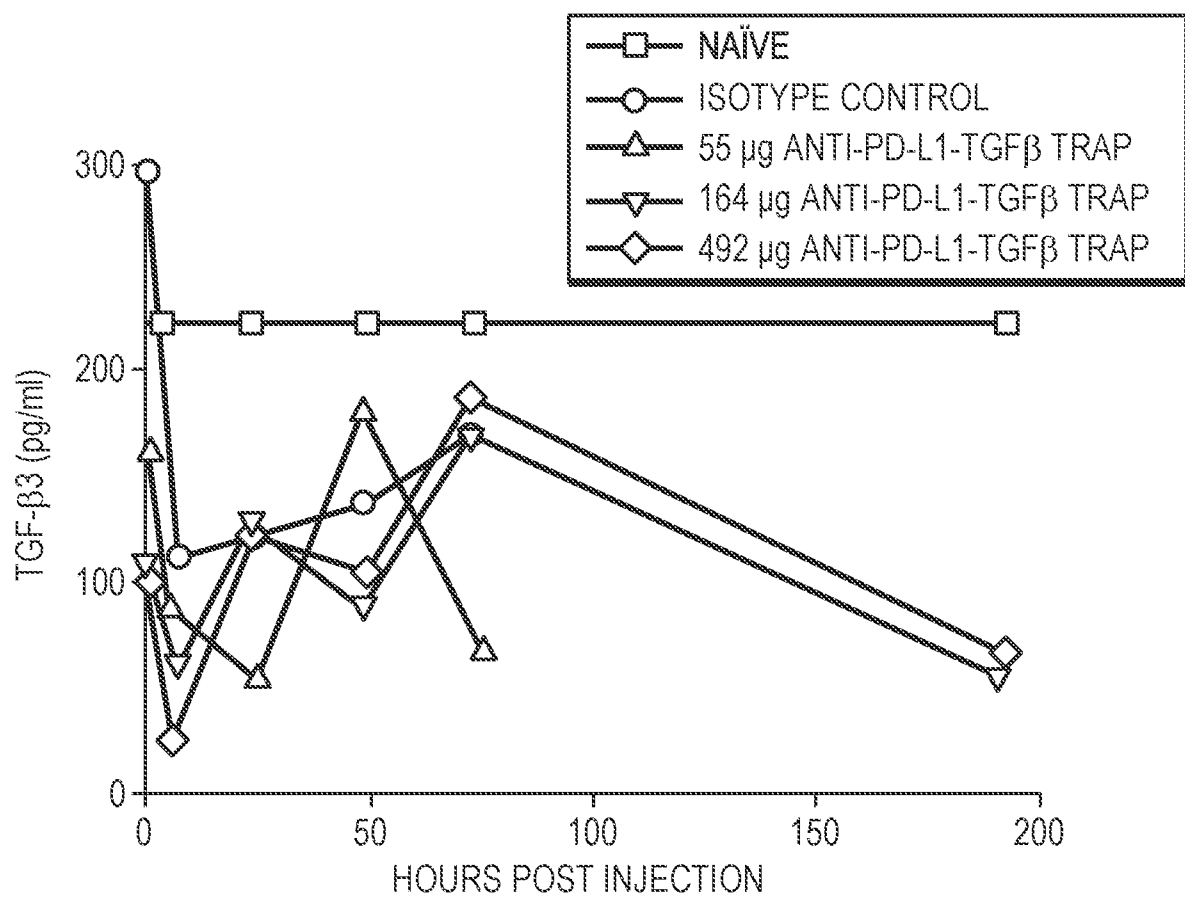


FIG. 4C

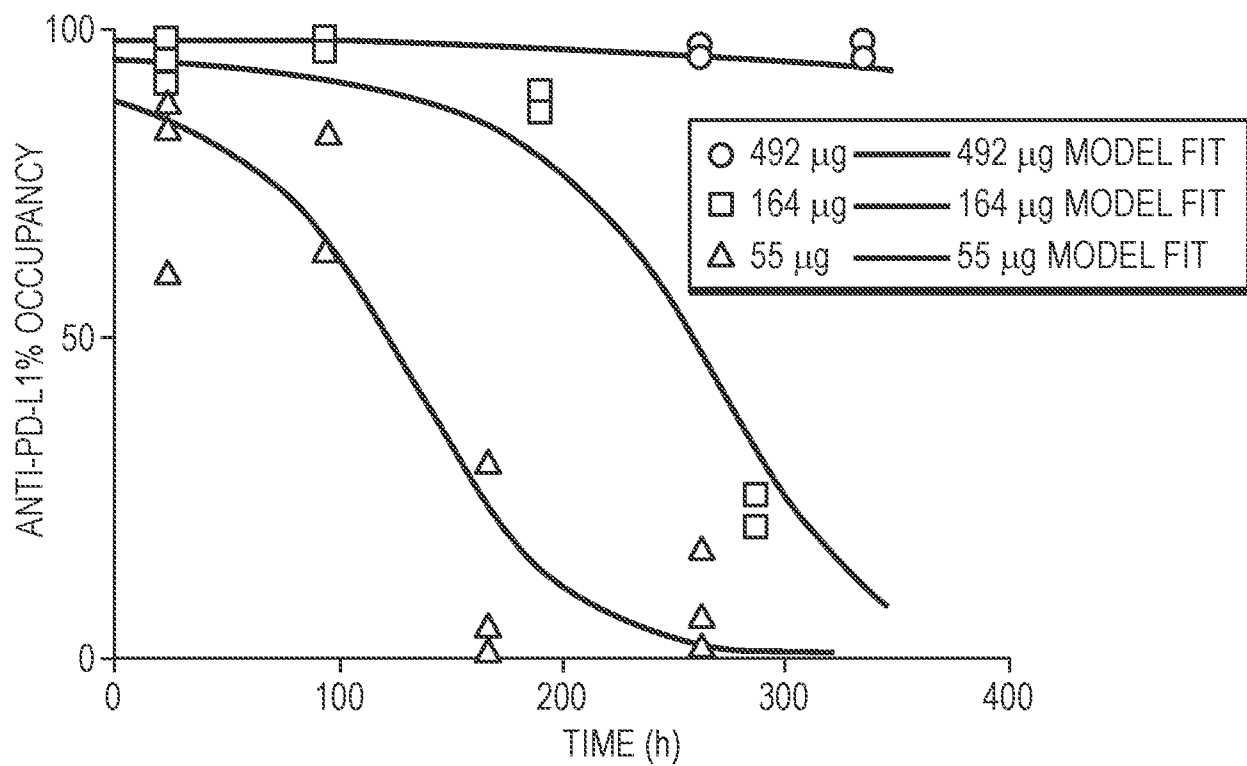


FIG. 4D

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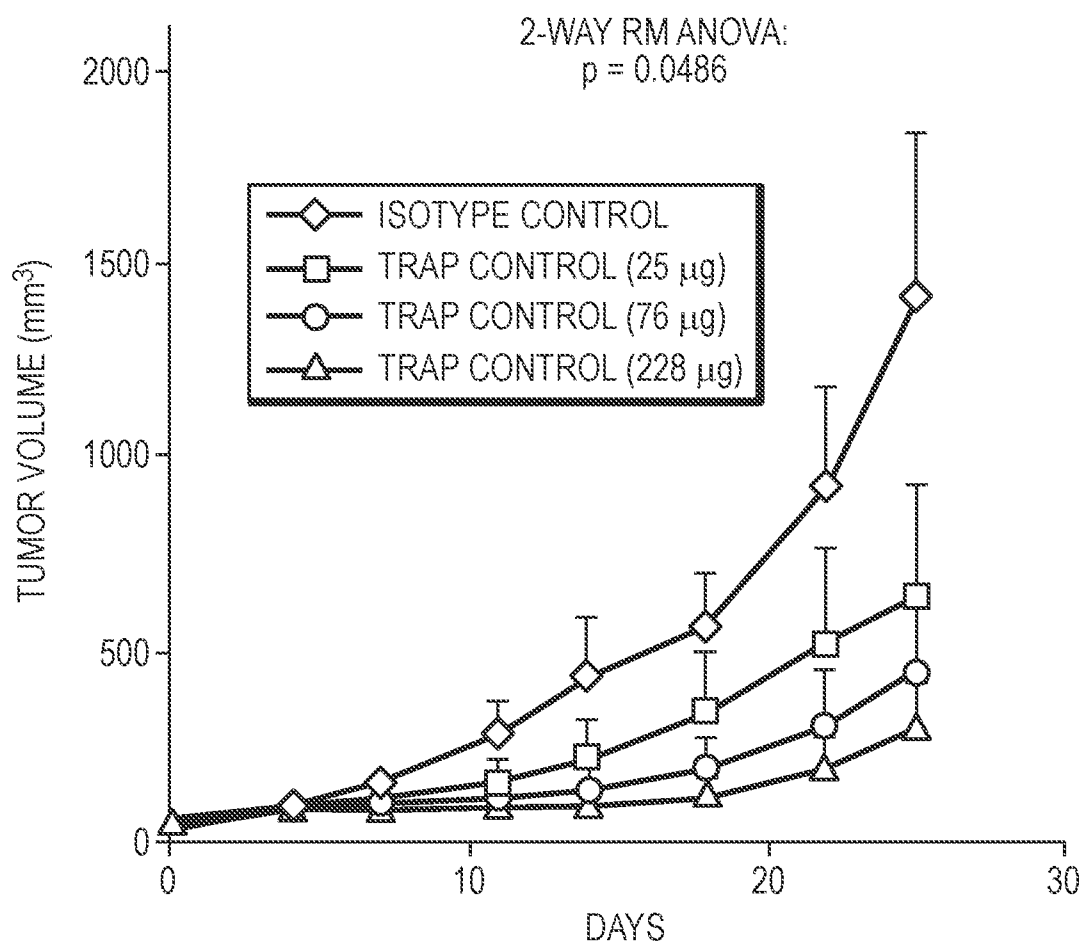


FIG. 5

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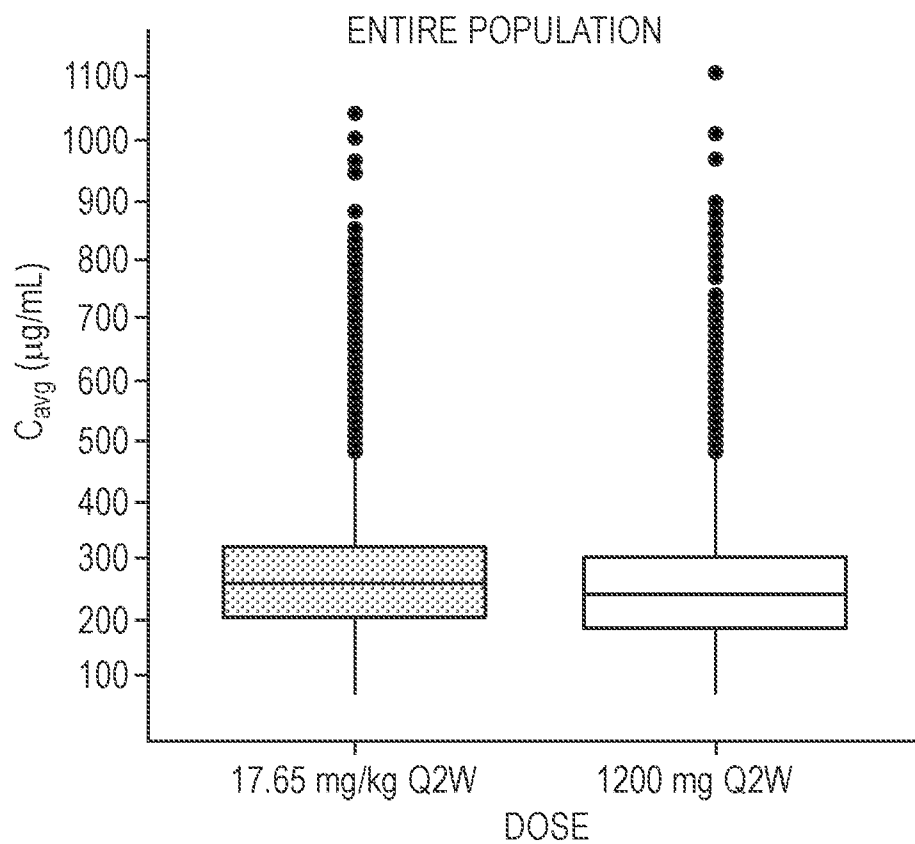


FIG. 6A

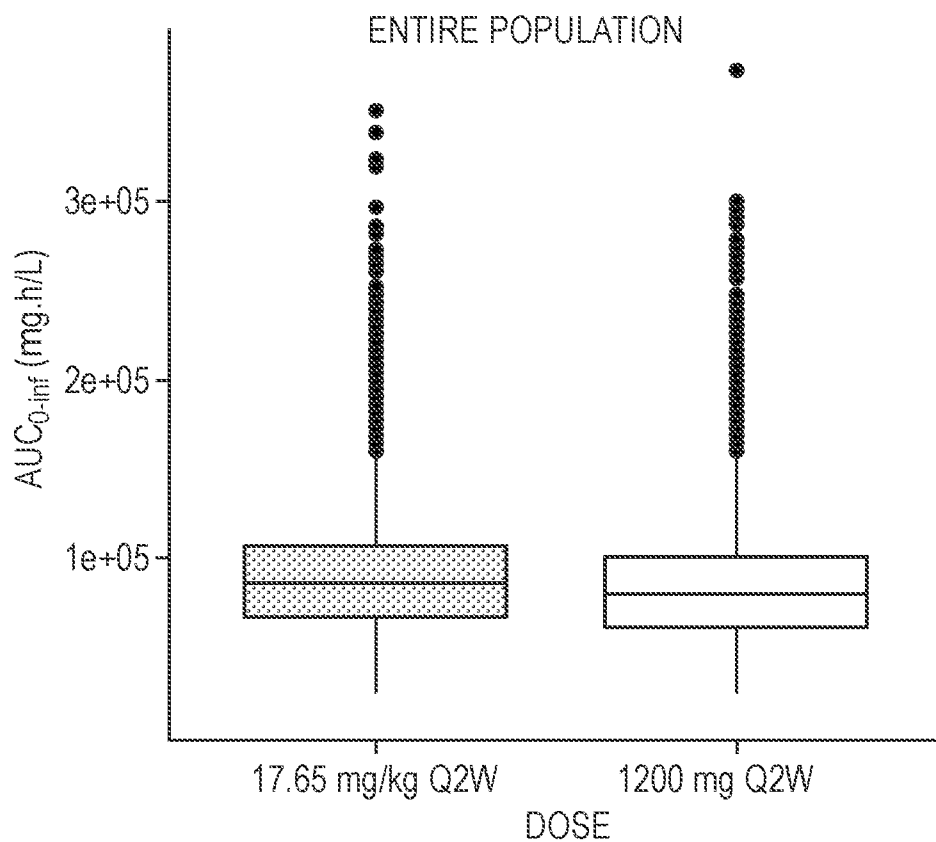


FIG. 6B

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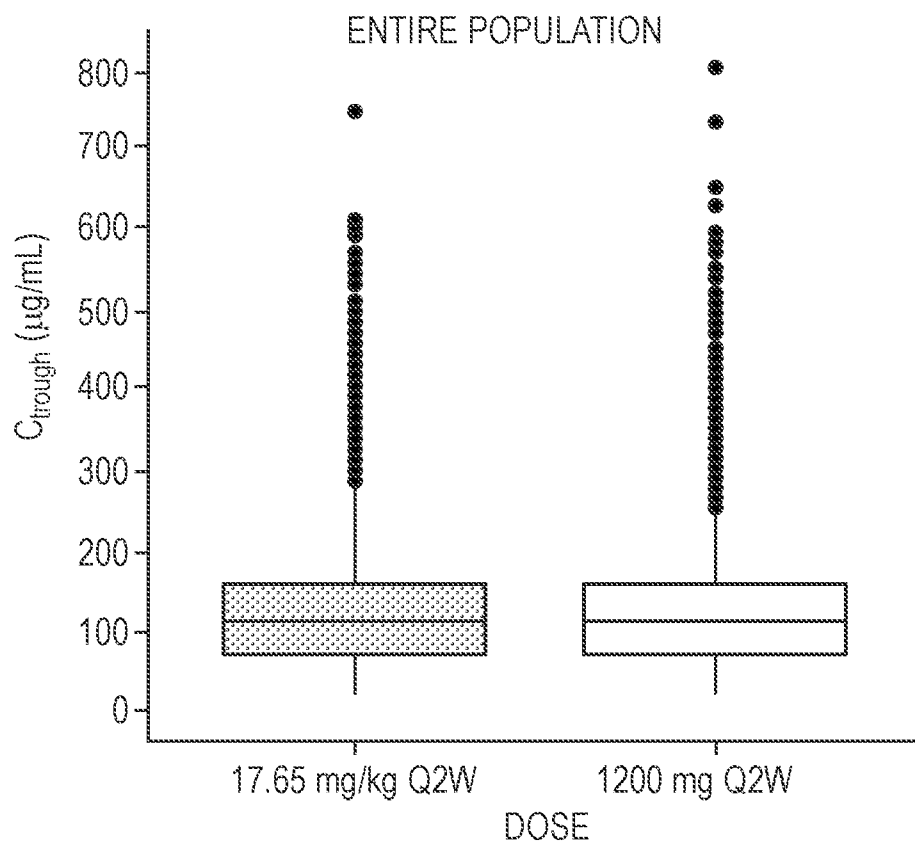


FIG. 6C

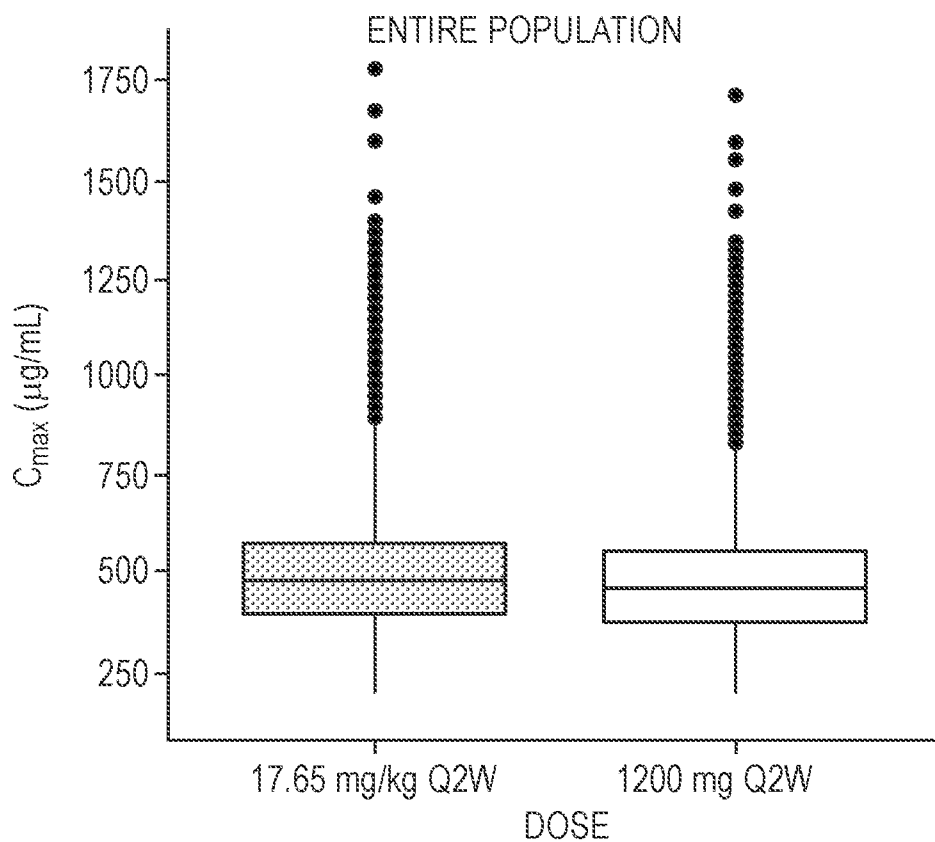


FIG. 6D

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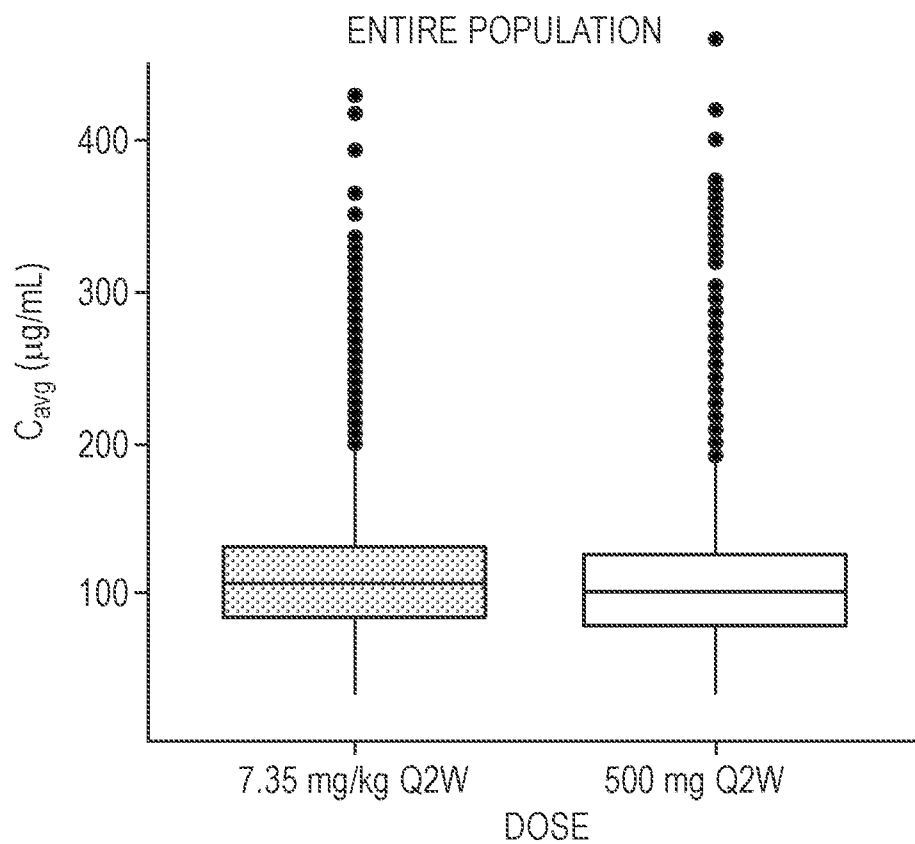


FIG. 6E

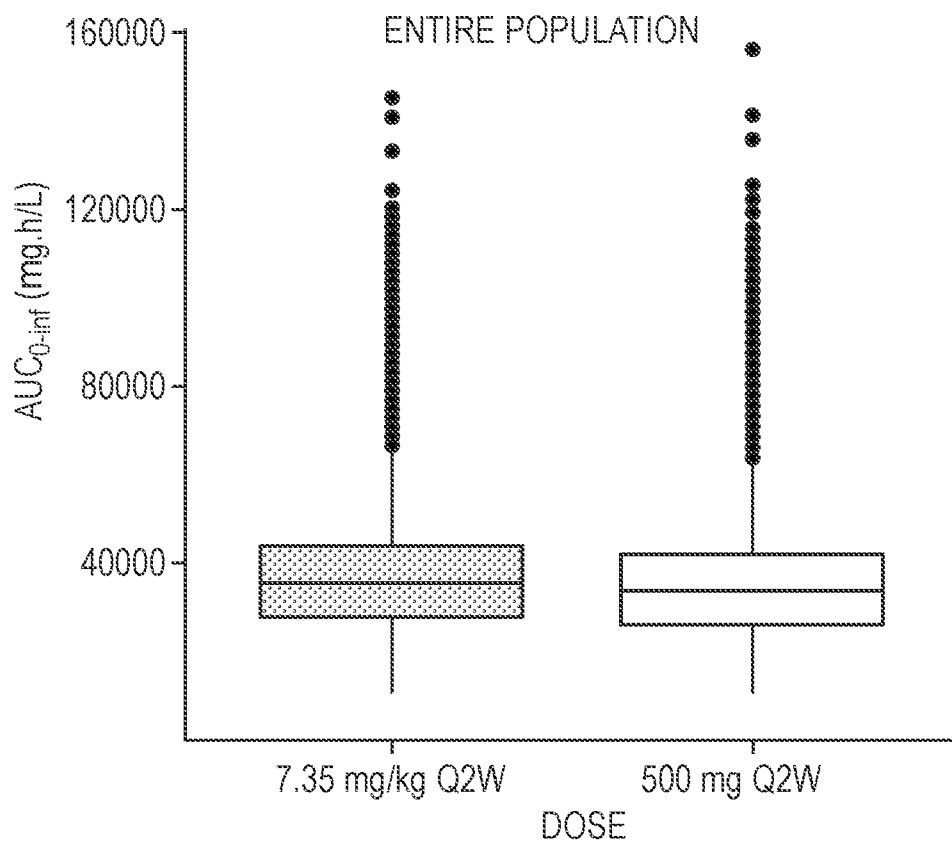


FIG. 6F

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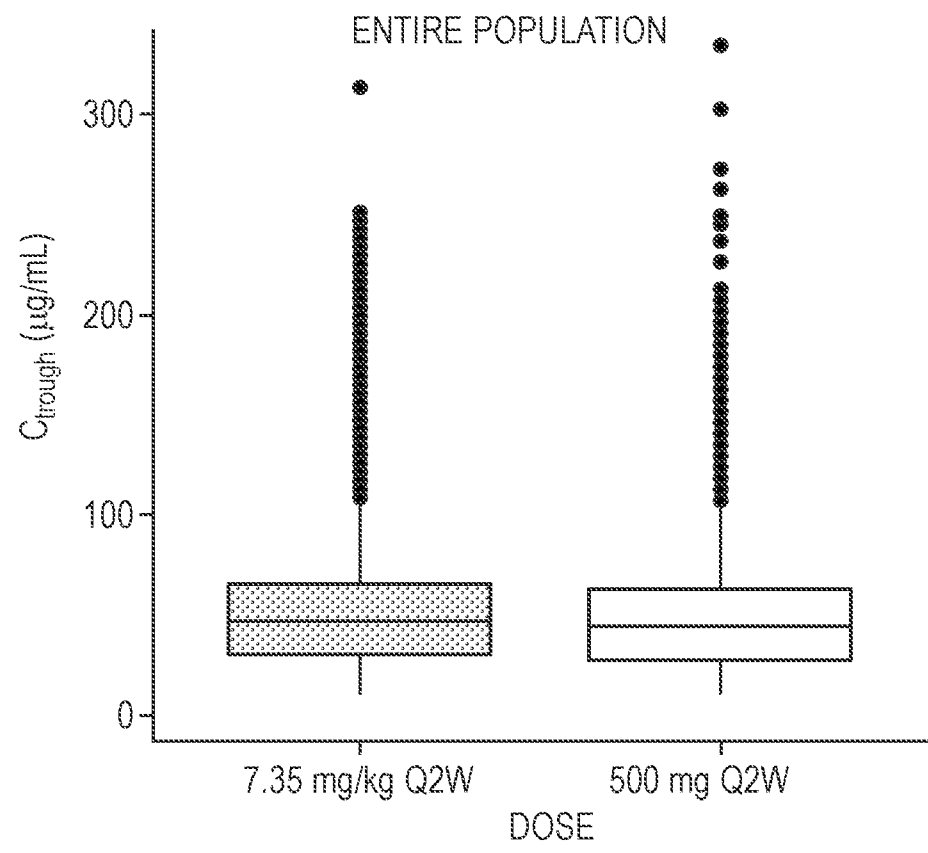


FIG. 6G

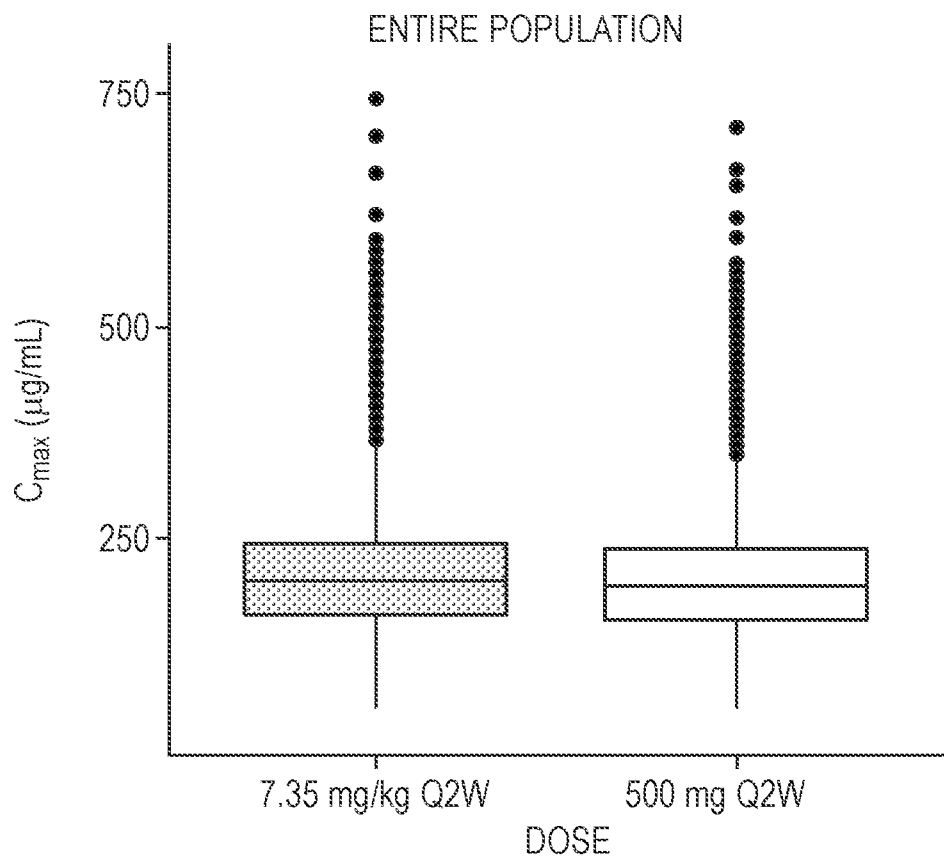


FIG. 6H

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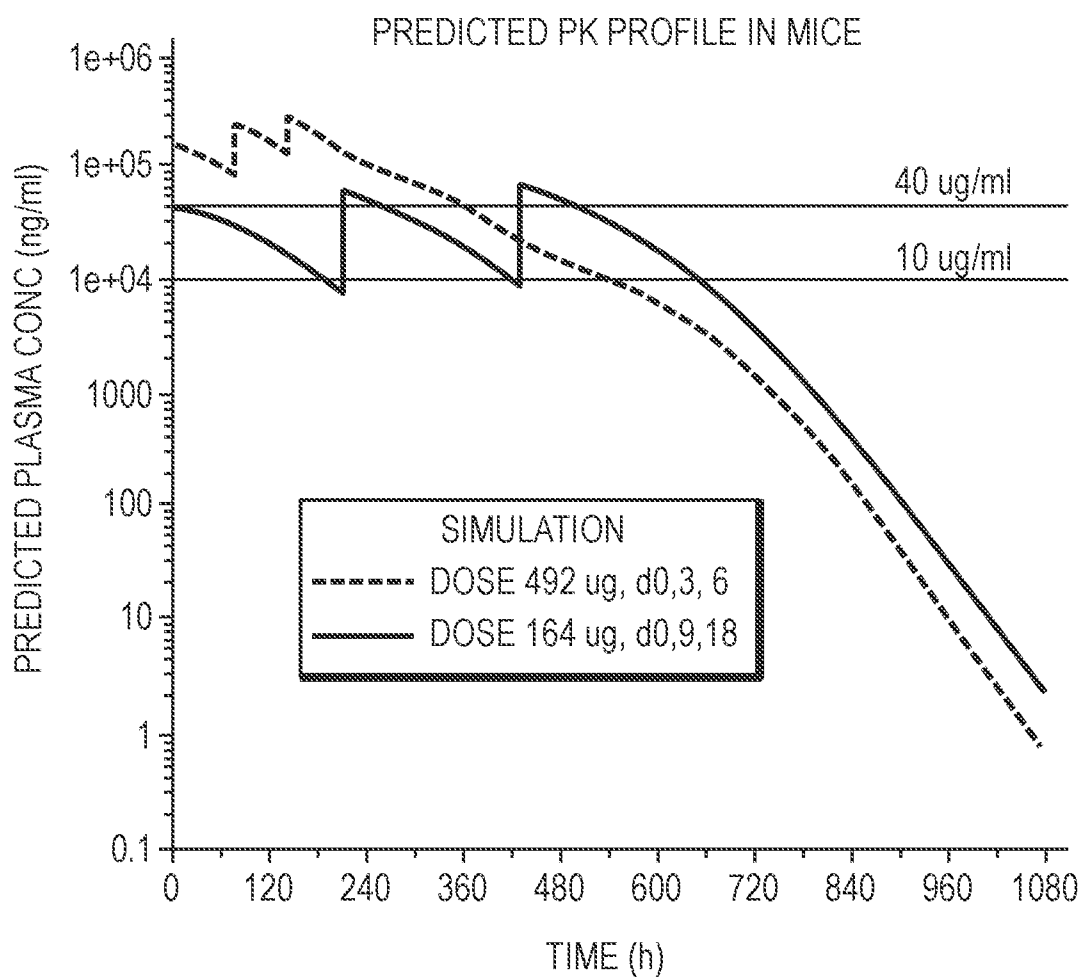


FIG. 7A

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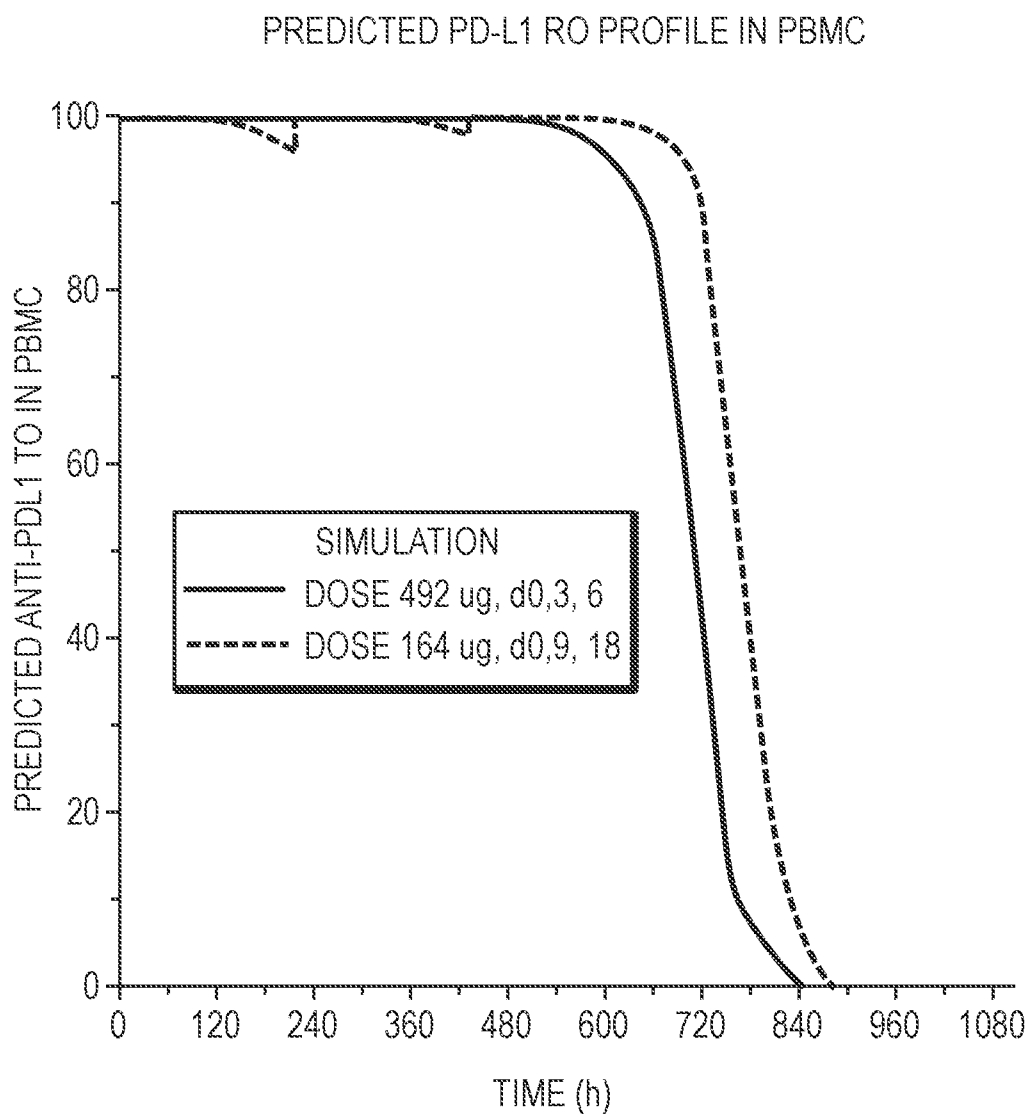


FIG. 7B

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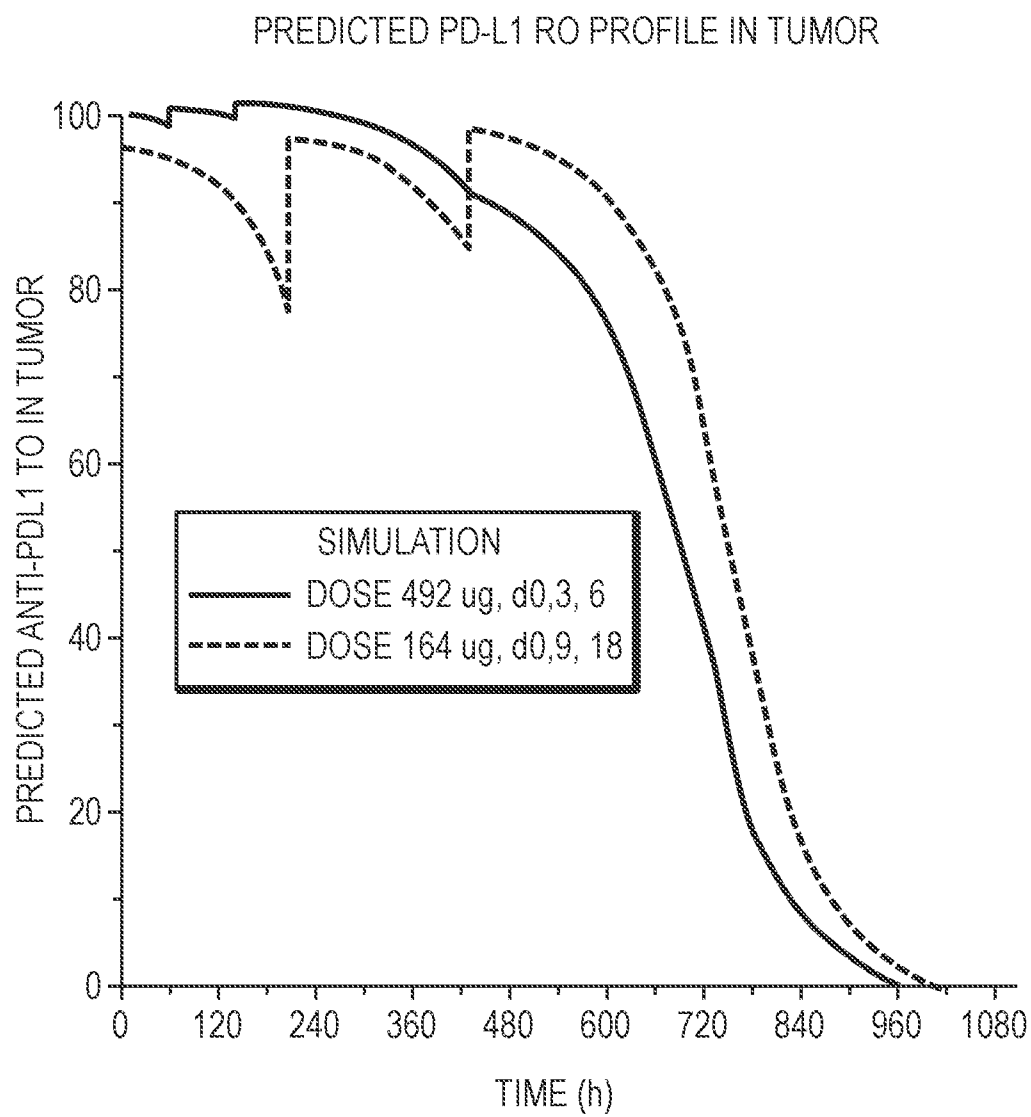
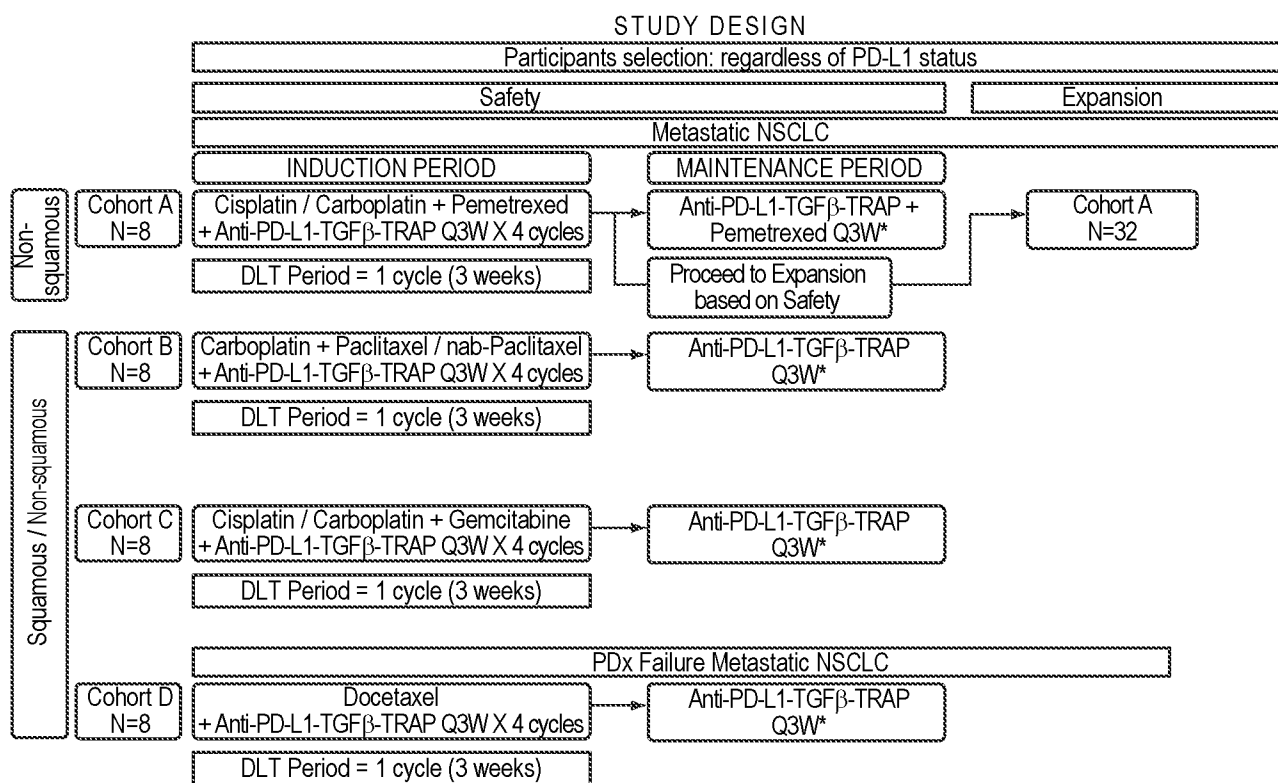


FIG. 7C

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* Up to 31 cycles (approximately 2 years), PD, or unacceptable toxicity

FIG. 8

SEQUENCE LISTING

<110> MERCK PATENT GMBH

<120> COMBINATION THERAPY WITH TARGETED TGF-B INHIBITION FOR
TREATMENT
OF ADVANCED NON-SMALL CELL LUNG CANCER

<130> EMD-011WO

<150> 62/693,042

<151> 2018-07-02

<150> 62/801,014

<151> 2019-02-04

<160> 62

<170> PatentIn version 3.5

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<211> 216

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<213> Artificial Sequence

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35 40 45

Met Ile Tyr Asp Val Ser Asn Arg Pro Ser Gly Val Ser Asn Arg Phe
50 55 60

Ser Gly Ser Lys Ser Gly Asn Thr Ala Ser Leu Thr Ile Ser Gly Leu
65 70 75 80

Gln Ala Glu Asp Glu Ala Asp Tyr Tyr Cys Ser Ser Tyr Thr Ser Ser
85 90 95

Ser Thr Arg Val Phe Gly Thr Gly Thr Lys Val Thr Val Leu Gly Gln
100 105 110

Pro Lys Ala Asn Pro Thr Val Thr Leu Phe Pro Pro Ser Ser Glu Glu
115 120 125

Leu Gln Ala Asn Lys Ala Thr Leu Val Cys Leu Ile Ser Asp Phe Tyr
130 135 140

Pro Gly Ala Val Thr Val Ala Trp Lys Ala Asp Gly Ser Pro Val Lys
145 150 155 160

Ala Gly Val Glu Thr Thr Lys Pro Ser Lys Gln Ser Asn Asn Lys Tyr
165 170 175

Ala Ala Ser Ser Tyr Leu Ser Leu Thr Pro Glu Gln Trp Lys Ser His
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20 25 30

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35 40 45

Ser Ser Ile Tyr Pro Ser Gly Gly Ile Thr Phe Tyr Ala Asp Thr Val
50 55 60

Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr
65 70 75 80

Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Ile Lys Leu Gly Thr Val Thr Thr Val Asp Tyr Trp Gly Gln
100 105 110

Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser Val
115 120 125

Phe Pro Leu Ala Pro Ser Ser Lys Ser Thr Ser Gly Gly Thr Ala Ala
130 135 140

Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser
145 150 155 160

Trp Asn Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val
165 170 175

Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro
180 185 190

Ser Ser Ser Leu Gly Thr Gln Thr Tyr Ile Cys Asn Val Asn His Lys
195 200 205

Pro Ser Asn Thr Lys Val Asp Lys Arg Val Glu Pro Lys Ser Cys Asp
210 215 220

Lys Thr His Thr Cys Pro Pro Cys Pro Ala Pro Glu Leu Leu Gly Gly
225 230 235 240

Pro Ser Val Phe Leu Phe Pro Pro Lys Pro Lys Asp Thr Leu Met Ile
245 250 255

Ser Arg Thr Pro Glu Val Thr Cys Val Val Val Asp Val Ser His Glu
260 265 270

Asp Pro Glu Val Lys Phe Asn Trp Tyr Val Asp Gly Val Glu Val His
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Asn Ala Lys Thr Lys Pro Arg Glu Glu Gln Tyr Asn Ser Thr Tyr Arg
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Val Val Ser Val Leu Thr Val Leu His Gln Asp Trp Leu Asn Gly Lys
305 310 315 320

Glu Tyr Lys Cys Lys Val Ser Asn Lys Ala Leu Pro Ala Pro Ile Glu
325 330 335

Lys Thr Ile Ser Lys Ala Lys Gly Gln Pro Arg Glu Pro Gln Val Tyr
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Thr Cys Leu Val Lys Gly Phe Tyr Pro Ser Asp Ile Ala Val Glu Trp
370 375 380

Glu Ser Asn Gly Gln Pro Glu Asn Asn Tyr Lys Thr Thr Pro Pro Val
385 390 395 400

Leu Asp Ser Asp Gly Ser Phe Phe Leu Tyr Ser Lys Leu Thr Val Asp
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Lys Ser Arg Trp Gln Gln Gly Asn Val Phe Ser Cys Ser Val Met His
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20 25 30

Ile Met Met Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val
35 40 45

Ser Ser Ile Tyr Pro Ser Gly Gly Ile Thr Phe Tyr Ala Asp Thr Val
50 55 60

Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr
65 70 75 80

Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Ile Lys Leu Gly Thr Val Thr Thr Val Asp Tyr Trp Gly Gln
100 105 110

Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser Val
115 120 125

Phe Pro Leu Ala Pro Ser Ser Lys Ser Thr Ser Gly Gly Thr Ala Ala
130 135 140

Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser
145 150 155 160

Trp Asn Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val
165 170 175

Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro
180 185 190

Ser Ser Ser Leu Gly Thr Gln Thr Tyr Ile Cys Asn Val Asn His Lys
 195 200 205

Pro Ser Asn Thr Lys Val Asp Lys Arg Val Glu Pro Lys Ser Cys Asp
 210 215 220

Lys Thr His Thr Cys Pro Pro Cys Pro Ala Pro Glu Leu Leu Gly Gly
 225 230 235 240

Pro Ser Val Phe Leu Phe Pro Pro Lys Pro Lys Asp Thr Leu Met Ile
 245 250 255

Ser Arg Thr Pro Glu Val Thr Cys Val Val Val Asp Val Ser His Glu
 260 265 270

Asp Pro Glu Val Lys Phe Asn Trp Tyr Val Asp Gly Val Glu Val His
 275 280 285

Asn Ala Lys Thr Lys Pro Arg Glu Glu Gln Tyr Asn Ser Thr Tyr Arg
 290 295 300

Val Val Ser Val Leu Thr Val Leu His Gln Asp Trp Leu Asn Gly Lys
 305 310 315 320

Glu Tyr Lys Cys Lys Val Ser Asn Lys Ala Leu Pro Ala Pro Ile Glu
 325 330 335

Lys Thr Ile Ser Lys Ala Lys Gly Gln Pro Arg Glu Pro Gln Val Tyr
 340 345 350

Thr Leu Pro Pro Ser Arg Glu Glu Met Thr Lys Asn Gln Val Ser Leu
 355 360 365

Thr Cys Leu Val Lys Gly Phe Tyr Pro Ser Asp Ile Ala Val Glu Trp

370

375

380

Glu Ser Asn Gly Gln Pro Glu Asn Asn Tyr Lys Thr Thr Pro Pro Val
 385 390 395 400

Leu Asp Ser Asp Gly Ser Phe Phe Leu Tyr Ser Lys Leu Thr Val Asp
 405 410 415

Lys Ser Arg Trp Gln Gln Gly Asn Val Phe Ser Cys Ser Val Met His
 420 425 430

Glu Ala Leu His Asn His Tyr Thr Gln Lys Ser Leu Ser Leu Ser Pro
 435 440 445

Gly Ala Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly Gly Gly Gly
 450 455 460

Ser Gly Gly Gly Gly Ser Gly Ile Pro Pro His Val Gln Lys Ser Val
 465 470 475 480

Asn Asn Asp Met Ile Val Thr Asp Asn Asn Gly Ala Val Lys Phe Pro
 485 490 495

Gln Leu Cys Lys Phe Cys Asp Val Arg Phe Ser Thr Cys Asp Asn Gln
 500 505 510

Lys Ser Cys Met Ser Asn Cys Ser Ile Thr Ser Ile Cys Glu Lys Pro
 515 520 525

Gln Glu Val Cys Val Ala Val Trp Arg Lys Asn Asp Glu Asn Ile Thr
 530 535 540

Leu Glu Thr Val Cys His Asp Pro Lys Leu Pro Tyr His Asp Phe Ile
 545 550 555 560

Leu Glu Asp Ala Ala Ser Pro Lys Cys Ile Met Lys Glu Lys Lys Lys
565 570 575

Pro Gly Glu Thr Phe Phe Met Cys Ser Cys Ser Ser Asp Glu Cys Asn
580 585 590

Asp Asn Ile Ile Phe Ser Glu Glu Tyr Asn Thr Ser Asn Pro Asp
595 600 605

<210> 4

<211> 711

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polynucleotide

<400> 4

atgagggccc tgctggctag actgctgctg tgcgtgctgg tcgtgtccga cagcaagggc
60

cagtccgccc tgaccagcc tgcctccgtg tctggctccc ctggccagtc catcaccatc
120

agctgcaccg gcacctccag cgacgtgggc ggctacaact acgtgtcctg gtatcagcag
180

caccccgcca aggccccaa gctgatgac tacgacgtgt ccaaccggcc ctccggcgtg
240

tccaacagat tctccggctc caagtccggc aacaccgcct ccctgaccat cagcggactg
300

caggcagagg acgaggccga ctactactgc tcctcctaca cctcctccag caccagagtg
360

ttcggcaccg gcacaaaagt gaccgtgctg ggccagccca aggccaaccc aaccgtgaca
420

ctgttcccc catcctccga ggaactgcag gccacaagg ccaccctggc ctgcctgac
480

tcagatttct atccaggcgc cgtgaccgtg gcctggaagg ctgatggctc cccagtgaag
540

gccggcgtgg aaaccaccaa gccctccaag cagtccaaca acaaatacgc cgcctcctcc
600

tacctgtccc tgacccccga gcagtggaag tcccaccggt cctacagctg ccaggtcaca
660

cacgagggct ccaccgtgga aaagaccgtc gccccaccg agtgctcatg a
711

<210> 5

<211> 1887

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polynucleotide

<400> 5

atggaacag acaccctgct gctgtgggtg ctgctgctgt gggtgcccgg ctccacaggc
60

gaggtgcagc tgctggaatc cggcggagga ctggtgcagc ctggcggctc cctgagactg
120

tcttgcccg cctccggctt caccttctcc agctacatca tgatgtgggt gcgacaggcc
180

cctggcaagg gcctggaatg ggtgtcctcc atctaccct cggcggcat caccttctac
240

gccgacaccg tgaagggccg gttcaccatc tcccgggaca actccaagaa caccctgtac
300

ctgcagatga actccctgcg ggccgaggac accgccgtgt actactgcgc ccggatcaag
360

ctgggcaccg tgaccaccgt ggactactgg ggccagggca ccctgggtgac agtgtcctcc
420

gctagcacca agggcccatc ggtcttcccc ctggcaccct cctccaagag cacctctggg
480

ggcacagcgg ccctgggctg cctgggtcaag gactacttcc ccgaaccggt gacgggtgtcg
540

tggaactcag gcgccctgac cagcggcgtg cacaccttcc cggctgtcct acagtcctca
600

ggactctact ccctcagcag cgtgggtgacc gtgccctcca gcagcttggg caccagacc
660

tacatctgca acgtgaatca caagcccagc aacaccaagg tggacaagag agttgagccc
720

aaatcttgtg acaaaactca cacatgccca ccgtgcccag cacctgaact cctgggggga
780

ccgtcagtct tcctcttccc cccaaaaccc aaggacaccc tcatgatctc ccggaccct
840

gaggtcacat gcgtgggtgg ggacgtgagc cacgaagacc ctgaggtcaa gttcaactgg
900

tacgtggacg gcgtggaggt gcataatgcc aagacaaagc cgcgggagga gcagtacaac
960

agcacgtacc gtgtgggtcag cgtcctcacc gtcctgcacc aggactggct gaatggcaag
1020

gagtacaagt gcaaggtctc caacaaagcc ctcccagccc ccatcgagaa aaccatctcc
1080

aaagccaaag ggcagccccg agaaccacag gtgtacaccc tgccccatc ccgggaggag
1140

atgaccaaga accaggtcag cctgacctgc ctgggtcaaag gcttctatcc cagcgacatc
1200

gccgtggagt gggagagcaa tgggcagccg gagaacaact acaagaccac gcctcccgtg
1260

ctggactccg acggctcctt cttcctctat agcaagctca ccgtggacaa gagcaggtgg
1320

cagcagggga acgtcttctc atgctccgtg atgcatgagg ctctgcacaa ccactacacg
1380

cagaagagcc tctccctgtc cccgggtgct ggcggcggag gaagcggagg aggtggcagc
1440

ggtggcgggtg gctccggcgg aggtggctcc ggaatccctc cccacgtgca gaagtccgtg
1500

aacaacgaca tgatcgtgac cgacaacaac ggcgccgtga agttccctca gctgtgcaag
1560

ttctgcgacg tgaggttcag cacctgcgac aaccagaagt cctgcatgag caactgcagc
1620

atcacaagca tctgcgagaa gccccaggag gtgtgtgtgg ccgtgtggag gaagaacgac
1680

gaaaacatca ccctcgagac cgtgtgccat gaccccaagc tgccctacca cgacttcac
1740

ctggaagacg ccgcctcccc caagtgcac atgaaggaga agaagaagcc cggcgagacc
1800

ttcttcatgt gcagctgcag cagcgacgag tgcaatgaca acatcatctt tagcgaggag
1860

t a c a a c a c c a g c a a c c c c g a c t g a t a a
1887

<210> 6

<211> 216

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 6

Gln Ser Ala Leu Thr Gln Pro Ala Ser Val Ser Gly Ser Pro Gly Gln
1 5 10 15

Ser Ile Thr Ile Ser Cys Thr Gly Thr Ser Ser Asp Val Gly Gly Tyr
20 25 30

Asn Tyr Val Ser Trp Tyr Gln Gln His Pro Gly Lys Ala Pro Lys Leu
35 40 45

Met Ile Tyr Glu Val Ser Asn Arg Pro Ser Gly Val Ser Asn Arg Phe
50 55 60

Ser Gly Ser Lys Ser Gly Asn Thr Ala Ser Leu Thr Ile Ser Gly Leu
65 70 75 80

Gln Ala Glu Asp Glu Ala Asp Tyr Tyr Cys Ser Ser Tyr Thr Ser Ser
85 90 95

Ser Thr Tyr Val Phe Gly Thr Gly Thr Lys Val Thr Val Leu Gly Gln
100 105 110

Pro Lys Ala Asn Pro Thr Val Thr Leu Phe Pro Pro Ser Ser Glu Glu
115 120 125

Leu Gln Ala Asn Lys Ala Thr Leu Val Cys Leu Ile Ser Asp Phe Tyr
130 135 140

Pro Gly Ala Val Thr Val Ala Trp Lys Ala Asp Gly Ser Pro Val Lys
145 150 155 160

Ala Gly Val Glu Thr Thr Lys Pro Ser Lys Gln Ser Asn Asn Lys Tyr
165 170 175

Ala Ala Ser Ser Tyr Leu Ser Leu Thr Pro Glu Gln Trp Lys Ser His
180 185 190

Arg Ser Tyr Ser Cys Gln Val Thr His Glu Gly Ser Thr Val Glu Lys
195 200 205

Thr Val Ala Pro Thr Glu Cys Ser
210 215

<210> 7

<211> 607

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 7

Glu Val Gln Leu Leu Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
1 5 10 15

Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Met Tyr
20 25 30

Met Met Met Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val
35 40 45

Ser Ser Ile Tyr Pro Ser Gly Gly Ile Thr Phe Tyr Ala Asp Ser Val
50 55 60

Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr
65 70 75 80

Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Ile Tyr Tyr Cys
85 90 95

Ala Arg Ile Lys Leu Gly Thr Val Thr Thr Val Asp Tyr Trp Gly Gln
100 105 110

Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser Val
115 120 125

Phe Pro Leu Ala Pro Ser Ser Lys Ser Thr Ser Gly Gly Thr Ala Ala
130 135 140

Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser
145 150 155 160

Trp Asn Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val
165 170 175

Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro
180 185 190

Ser Ser Ser Leu Gly Thr Gln Thr Tyr Ile Cys Asn Val Asn His Lys
195 200 205

Pro Ser Asn Thr Lys Val Asp Lys Arg Val Glu Pro Lys Ser Cys Asp
210 215 220

Lys Thr His Thr Cys Pro Pro Cys Pro Ala Pro Glu Leu Leu Gly Gly
225 230 235 240

Pro Ser Val Phe Leu Phe Pro Pro Lys Pro Lys Asp Thr Leu Met Ile
245 250 255

Ser Arg Thr Pro Glu Val Thr Cys Val Val Val Asp Val Ser His Glu
260 265 270

Asp Pro Glu Val Lys Phe Asn Trp Tyr Val Asp Gly Val Glu Val His
275 280 285

Asn Ala Lys Thr Lys Pro Arg Glu Glu Gln Tyr Asn Ser Thr Tyr Arg
290 295 300

Val Val Ser Val Leu Thr Val Leu His Gln Asp Trp Leu Asn Gly Lys
305 310 315 320

Glu Tyr Lys Cys Lys Val Ser Asn Lys Ala Leu Pro Ala Pro Ile Glu
325 330 335

Lys Thr Ile Ser Lys Ala Lys Gly Gln Pro Arg Glu Pro Gln Val Tyr
340 345 350

Thr Leu Pro Pro Ser Arg Glu Glu Met Thr Lys Asn Gln Val Ser Leu
355 360 365

Thr Cys Leu Val Lys Gly Phe Tyr Pro Ser Asp Ile Ala Val Glu Trp
370 375 380

Glu Ser Asn Gly Gln Pro Glu Asn Asn Tyr Lys Thr Thr Pro Pro Val
385 390 395 400

Leu Asp Ser Asp Gly Ser Phe Phe Leu Tyr Ser Lys Leu Thr Val Asp
405 410 415

Lys Ser Arg Trp Gln Gln Gly Asn Val Phe Ser Cys Ser Val Met His
420 425 430

Glu Ala Leu His Asn His Tyr Thr Gln Lys Ser Leu Ser Leu Ser Pro
435 440 445

Gly Ala Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly Gly Gly Gly
450 455 460

Ser Gly Gly Gly Gly Ser Gly Ile Pro Pro His Val Gln Lys Ser Val
465 470 475 480

Asn Asn Asp Met Ile Val Thr Asp Asn Asn Gly Ala Val Lys Phe Pro
 485 490 495

Gln Leu Cys Lys Phe Cys Asp Val Arg Phe Ser Thr Cys Asp Asn Gln
 500 505 510

Lys Ser Cys Met Ser Asn Cys Ser Ile Thr Ser Ile Cys Glu Lys Pro
 515 520 525

Gln Glu Val Cys Val Ala Val Trp Arg Lys Asn Asp Glu Asn Ile Thr
 530 535 540

Leu Glu Thr Val Cys His Asp Pro Lys Leu Pro Tyr His Asp Phe Ile
 545 550 555 560

Leu Glu Asp Ala Ala Ser Pro Lys Cys Ile Met Lys Glu Lys Lys Lys
 565 570 575

Pro Gly Glu Thr Phe Phe Met Cys Ser Cys Ser Ser Asp Glu Cys Asn
 580 585 590

Asp Asn Ile Ile Phe Ser Glu Glu Tyr Asn Thr Ser Asn Pro Asp
 595 600 605

<210> 8
 <211> 592
 <212> PRT
 <213> Homo sapiens

<400> 8

Met Gly Arg Gly Leu Leu Arg Gly Leu Trp Pro Leu His Ile Val Leu
 1 5 10 15

Trp Thr Arg Ile Ala Ser Thr Ile Pro Pro His Val Gln Lys Ser Asp
 20 25 30

Val Glu Met Glu Ala Gln Lys Asp Glu Ile Ile Cys Pro Ser Cys Asn
35 40 45

Arg Thr Ala His Pro Leu Arg His Ile Asn Asn Asp Met Ile Val Thr
50 55 60

Asp Asn Asn Gly Ala Val Lys Phe Pro Gln Leu Cys Lys Phe Cys Asp
65 70 75 80

Val Arg Phe Ser Thr Cys Asp Asn Gln Lys Ser Cys Met Ser Asn Cys
85 90 95

Ser Ile Thr Ser Ile Cys Glu Lys Pro Gln Glu Val Cys Val Ala Val
100 105 110

Trp Arg Lys Asn Asp Glu Asn Ile Thr Leu Glu Thr Val Cys His Asp
115 120 125

Pro Lys Leu Pro Tyr His Asp Phe Ile Leu Glu Asp Ala Ala Ser Pro
130 135 140

Lys Cys Ile Met Lys Glu Lys Lys Lys Pro Gly Glu Thr Phe Phe Met
145 150 155 160

Cys Ser Cys Ser Ser Asp Glu Cys Asn Asp Asn Ile Ile Phe Ser Glu
165 170 175

Glu Tyr Asn Thr Ser Asn Pro Asp Leu Leu Leu Val Ile Phe Gln Val
180 185 190

Thr Gly Ile Ser Leu Leu Pro Pro Leu Gly Val Ala Ile Ser Val Ile
195 200 205

Ile Ile Phe Tyr Cys Tyr Arg Val Asn Arg Gln Gln Lys Leu Ser Ser
210 215 220

Thr Trp Glu Thr Gly Lys Thr Arg Lys Leu Met Glu Phe Ser Glu His
225 230 235 240

Cys Ala Ile Ile Leu Glu Asp Asp Arg Ser Asp Ile Ser Ser Thr Cys
245 250 255

Ala Asn Asn Ile Asn His Asn Thr Glu Leu Leu Pro Ile Glu Leu Asp
260 265 270

Thr Leu Val Gly Lys Gly Arg Phe Ala Glu Val Tyr Lys Ala Lys Leu
275 280 285

Lys Gln Asn Thr Ser Glu Gln Phe Glu Thr Val Ala Val Lys Ile Phe
290 295 300

Pro Tyr Glu Glu Tyr Ala Ser Trp Lys Thr Glu Lys Asp Ile Phe Ser
305 310 315 320

Asp Ile Asn Leu Lys His Glu Asn Ile Leu Gln Phe Leu Thr Ala Glu
325 330 335

Glu Arg Lys Thr Glu Leu Gly Lys Gln Tyr Trp Leu Ile Thr Ala Phe
340 345 350

His Ala Lys Gly Asn Leu Gln Glu Tyr Leu Thr Arg His Val Ile Ser
355 360 365

Trp Glu Asp Leu Arg Lys Leu Gly Ser Ser Leu Ala Arg Gly Ile Ala
370 375 380

His Leu His Ser Asp His Thr Pro Cys Gly Arg Pro Lys Met Pro Ile
385 390 395 400

Val His Arg Asp Leu Lys Ser Ser Asn Ile Leu Val Lys Asn Asp Leu
405 410 415

Thr Cys Cys Leu Cys Asp Phe Gly Leu Ser Leu Arg Leu Asp Pro Thr
420 425 430

Leu Ser Val Asp Asp Leu Ala Asn Ser Gly Gln Val Gly Thr Ala Arg
435 440 445

Tyr Met Ala Pro Glu Val Leu Glu Ser Arg Met Asn Leu Glu Asn Val
450 455 460

Glu Ser Phe Lys Gln Thr Asp Val Tyr Ser Met Ala Leu Val Leu Trp
465 470 475 480

Glu Met Thr Ser Arg Cys Asn Ala Val Gly Glu Val Lys Asp Tyr Glu
485 490 495

Pro Pro Phe Gly Ser Lys Val Arg Glu His Pro Cys Val Glu Ser Met
500 505 510

Lys Asp Asn Val Leu Arg Asp Arg Gly Arg Pro Glu Ile Pro Ser Phe
515 520 525

Trp Leu Asn His Gln Gly Ile Gln Met Val Cys Glu Thr Leu Thr Glu
530 535 540

Cys Trp Asp His Asp Pro Glu Ala Arg Leu Thr Ala Gln Cys Val Ala
545 550 555 560

Glu Arg Phe Ser Glu Leu Glu His Leu Asp Arg Leu Ser Gly Arg Ser

565

570

575

Cys Ser Glu Glu Lys Ile Pro Glu Asp Gly Ser Leu Asn Thr Thr Lys
580 585 590

<210> 9

<211> 567

<212> PRT

<213> Homo sapiens

<400> 9

Met Gly Arg Gly Leu Leu Arg Gly Leu Trp Pro Leu His Ile Val Leu
1 5 10 15

Trp Thr Arg Ile Ala Ser Thr Ile Pro Pro His Val Gln Lys Ser Val
20 25 30

Asn Asn Asp Met Ile Val Thr Asp Asn Asn Gly Ala Val Lys Phe Pro
35 40 45

Gln Leu Cys Lys Phe Cys Asp Val Arg Phe Ser Thr Cys Asp Asn Gln
50 55 60

Lys Ser Cys Met Ser Asn Cys Ser Ile Thr Ser Ile Cys Glu Lys Pro
65 70 75 80

Gln Glu Val Cys Val Ala Val Trp Arg Lys Asn Asp Glu Asn Ile Thr
85 90 95

Leu Glu Thr Val Cys His Asp Pro Lys Leu Pro Tyr His Asp Phe Ile
100 105 110

Leu Glu Asp Ala Ala Ser Pro Lys Cys Ile Met Lys Glu Lys Lys Lys
115 120 125

Pro Gly Glu Thr Phe Phe Met Cys Ser Cys Ser Ser Asp Glu Cys Asn
130 135 140

Asp Asn Ile Ile Phe Ser Glu Glu Tyr Asn Thr Ser Asn Pro Asp Leu
145 150 155 160

Leu Leu Val Ile Phe Gln Val Thr Gly Ile Ser Leu Leu Pro Pro Leu
165 170 175

Gly Val Ala Ile Ser Val Ile Ile Ile Phe Tyr Cys Tyr Arg Val Asn
180 185 190

Arg Gln Gln Lys Leu Ser Ser Thr Trp Glu Thr Gly Lys Thr Arg Lys
195 200 205

Leu Met Glu Phe Ser Glu His Cys Ala Ile Ile Leu Glu Asp Asp Arg
210 215 220

Ser Asp Ile Ser Ser Thr Cys Ala Asn Asn Ile Asn His Asn Thr Glu
225 230 235 240

Leu Leu Pro Ile Glu Leu Asp Thr Leu Val Gly Lys Gly Arg Phe Ala
245 250 255

Glu Val Tyr Lys Ala Lys Leu Lys Gln Asn Thr Ser Glu Gln Phe Glu
260 265 270

Thr Val Ala Val Lys Ile Phe Pro Tyr Glu Glu Tyr Ala Ser Trp Lys
275 280 285

Thr Glu Lys Asp Ile Phe Ser Asp Ile Asn Leu Lys His Glu Asn Ile
290 295 300

Leu Gln Phe Leu Thr Ala Glu Glu Arg Lys Thr Glu Leu Gly Lys Gln
 305 310 315 320

Tyr Trp Leu Ile Thr Ala Phe His Ala Lys Gly Asn Leu Gln Glu Tyr
 325 330 335

Leu Thr Arg His Val Ile Ser Trp Glu Asp Leu Arg Lys Leu Gly Ser
 340 345 350

Ser Leu Ala Arg Gly Ile Ala His Leu His Ser Asp His Thr Pro Cys
 355 360 365

Gly Arg Pro Lys Met Pro Ile Val His Arg Asp Leu Lys Ser Ser Asn
 370 375 380

Ile Leu Val Lys Asn Asp Leu Thr Cys Cys Leu Cys Asp Phe Gly Leu
 385 390 395 400

Ser Leu Arg Leu Asp Pro Thr Leu Ser Val Asp Asp Leu Ala Asn Ser
 405 410 415

Gly Gln Val Gly Thr Ala Arg Tyr Met Ala Pro Glu Val Leu Glu Ser
 420 425 430

Arg Met Asn Leu Glu Asn Val Glu Ser Phe Lys Gln Thr Asp Val Tyr
 435 440 445

Ser Met Ala Leu Val Leu Trp Glu Met Thr Ser Arg Cys Asn Ala Val
 450 455 460

Gly Glu Val Lys Asp Tyr Glu Pro Pro Phe Gly Ser Lys Val Arg Glu
 465 470 475 480

His Pro Cys Val Glu Ser Met Lys Asp Asn Val Leu Arg Asp Arg Gly

495

Ser Ile Thr Ser Ile Cys Glu Lys Pro Gln Glu Val Cys Val Ala Val
50 55 60

Trp Arg Lys Asn Asp Glu Asn Ile Thr Leu Glu Thr Val Cys His Asp
65 70 75 80

Pro Lys Leu Pro Tyr His Asp Phe Ile Leu Glu Asp Ala Ala Ser Pro
85 90 95

Lys Cys Ile Met Lys Glu Lys Lys Lys Pro Gly Glu Thr Phe Phe Met
100 105 110

Cys Ser Cys Ser Ser Asp Glu Cys Asn Asp Asn Ile Ile Phe Ser Glu
115 120 125

Glu Tyr Asn Thr Ser Asn Pro Asp
130 135

<210> 11

<211> 21

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 11

Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly
1 5 10 15

Gly Gly Gly Ser Gly
20

<210> 12

<211> 118

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 12

Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
1 5 10 15

Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Asp Ser
20 25 30

Trp Ile His Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val
35 40 45

Ala Trp Ile Ser Pro Tyr Gly Gly Ser Thr Tyr Tyr Ala Asp Ser Val
50 55 60

Lys Gly Arg Phe Thr Ile Ser Ala Asp Thr Ser Lys Asn Thr Ala Tyr
65 70 75 80

Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Arg His Trp Pro Gly Gly Phe Asp Tyr Trp Gly Gln Gly Thr
100 105 110

Leu Val Thr Val Ser Ser
115

<210> 13

<211> 108

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 13

Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly
1 5 10 15

Asp Arg Val Thr Ile Thr Cys Arg Ala Ser Gln Asp Val Ser Thr Ala
20 25 30

Val Ala Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile
35 40 45

Tyr Ser Ala Ser Phe Leu Tyr Ser Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro
65 70 75 80

Glu Asp Phe Ala Thr Tyr Tyr Cys Gln Gln Tyr Leu Tyr His Pro Ala
85 90 95

Thr Phe Gly Gln Gly Thr Lys Val Glu Ile Lys Arg
100 105

<210> 14

<211> 118

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 14

Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
1 5 10 15

Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Asp Ser
20 25 30

Trp Ile His Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val
35 40 45

Ala Trp Ile Ser Pro Tyr Gly Gly Ser Thr Tyr Tyr Ala Asp Ser Val
50 55 60

Lys Gly Arg Phe Thr Ile Ser Ala Asp Thr Ser Lys Asn Thr Ala Tyr
65 70 75 80

Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Arg His Trp Pro Gly Gly Phe Asp Tyr Trp Gly Gln Gly Thr
100 105 110

Leu Val Thr Val Ser Ala
115

<210> 15

<211> 4

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 15

Gln Phe Asn Ser

1

<210> 16

<211> 4

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 16

Gln Ala Gln Ser

1

<210> 17

<211> 6

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 17

Pro Lys Ser Cys Asp Lys

1

5

<210> 18

<211> 6

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 18

Pro Lys Ser Ser Asp Lys

1

5

<210> 19

<211> 4

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 19

Leu Ser Leu Ser
1

<210> 20

<211> 4

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 20

Ala Thr Ala Thr
1

<210> 21

<211> 5

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<220>

<221> MOD_RES

<222> (1)..(1)

<223> Lys, Arg, Thr, Gln, Gly, Ala, Trp, Met, Ile or Ser

<220>

<221> MOD_RES

<222> (3)..(3)

<223> Val, Arg, Lys, Leu, Met or Ile

<220>

<221> MOD_RES

<222> (5)..(5)

<223> His, Thr, Asn, Gln, Ala, Val, Tyr, Trp, Phe or Met

<400> 21

Xaa Tyr Xaa Met Xaa
1 5

<210> 22
<211> 17
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic peptide

<220>
<221> MOD_RES
<222> (8)..(8)
<223> Phe or Ile

<220>
<221> MOD_RES
<222> (14)..(14)
<223> Ser or Thr

<400> 22

Ser Ile Tyr Pro Ser Gly Gly Xaa Thr Phe Tyr Ala Asp Xaa Val Lys
1 5 10 15

Gly

<210> 23
<211> 11
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic peptide

<220>
<221> MOD_RES

<222> (10)..(10)

<223> Glu or Asp

<400> 23

Ile Lys Leu Gly Thr Val Thr Thr Val Xaa Tyr
1 5 10

<210> 24

<211> 30

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 24

Glu Val Gln Leu Leu Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
1 5 10 15

Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser
20 25 30

<210> 25

<211> 14

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 25

Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val Ser
1 5 10

<210> 26

<211> 32

<212> PRT

<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 26

Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr Leu Gln
1 5 10 15

Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys Ala Arg
20 25 30

<210> 27
<211> 11
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic peptide

<400> 27

Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser
1 5 10

<210> 28
<211> 14
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic peptide

<220>
<221> MOD_RES
<222> (4)..(4)
<223> Asn or Ser

<220>
<221> MOD_RES
<222> (5)..(5)

<223> Thr, Arg or Ser

<220>

<221> MOD_RES

<222> (9)..(9)

<223> Ala or Gly

<400> 28

Thr	Gly	Thr	Xaa	Xaa	Asp	Val	Gly	Xaa	Tyr	Asn	Tyr	Val	Ser
1				5					10				

<210> 29

<211> 7

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<220>

<221> MOD_RES

<222> (1)..(1)

<223> Glu or Asp

<220>

<221> MOD_RES

<222> (3)..(3)

<223> Ile, Asn or Ser

<220>

<221> MOD_RES

<222> (4)..(4)

<223> Asp, His or Asn

<400> 29

Xaa	Val	Xaa	Xaa	Arg	Pro	Ser
1				5		

<210> 30

<211> 10

<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic peptide

<220>
<221> MOD_RES
<222> (3)..(3)
<223> Phe or Tyr

<220>
<221> MOD_RES
<222> (5)..(5)
<223> Asn or Ser

<220>
<221> MOD_RES
<222> (6)..(6)
<223> Arg, Thr or Ser

<220>
<221> MOD_RES
<222> (7)..(7)
<223> Gly or Ser

<220>
<221> MOD_RES
<222> (8)..(8)
<223> Ile or Thr

<400> 30

Ser Ser Xaa Thr Xaa Xaa Xaa Xaa Arg Val
1 5 10

<210> 31
<211> 22
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic peptide

<400> 31

Gln Ser Ala Leu Thr Gln Pro Ala Ser Val Ser Gly Ser Pro Gly Gln
1 5 10 15

Ser Ile Thr Ile Ser Cys
20

<210> 32

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 32

Trp Tyr Gln Gln His Pro Gly Lys Ala Pro Lys Leu Met Ile Tyr
1 5 10 15

<210> 33

<211> 32

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 33

Gly Val Ser Asn Arg Phe Ser Gly Ser Lys Ser Gly Asn Thr Ala Ser
1 5 10 15

Leu Thr Ile Ser Gly Leu Gln Ala Glu Asp Glu Ala Asp Tyr Tyr Cys
20 25 30

<210> 34

<211> 10

<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic peptide

<400> 34

Phe Gly Thr Gly Thr Lys Val Thr Val Leu
1 5 10

<210> 35
<211> 5
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic peptide

<400> 35

Ser Tyr Ile Met Met
1 5

<210> 36
<211> 17
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic peptide

<400> 36

Ser Ile Tyr Pro Ser Gly Gly Ile Thr Phe Tyr Ala Asp Thr Val Lys
1 5 10 15

Gly

<210> 37

<211> 11
<212> PRT
<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 37

Ile Lys Leu Gly Thr Val Thr Thr Val Asp Tyr
1 5 10

<210> 38
<211> 14
<212> PRT
<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 38

Thr Gly Thr Ser Ser Asp Val Gly Gly Tyr Asn Tyr Val Ser
1 5 10

<210> 39
<211> 7
<212> PRT
<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 39

Asp Val Ser Asn Arg Pro Ser
1 5

<210> 40
<211> 10
<212> PRT
<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 40

Ser Ser Tyr Thr Ser Ser Ser Thr Arg Val
1 5 10

<210> 41

<211> 5

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 41

Met Tyr Met Met Met
1 5

<210> 42

<211> 17

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 42

Ser Ile Tyr Pro Ser Gly Gly Ile Thr Phe Tyr Ala Asp Ser Val Lys
1 5 10 15

Gly

<210> 43

<211> 14

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic peptide

<400> 43

Thr Gly Thr Ser Ser Asp Val Gly Ala Tyr Asn Tyr Val Ser
1 5 10

<210> 44

<211> 119

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 44

Glu Val Gln Leu Leu Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
1 5 10 15

Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Ser Tyr
20 25 30

Ile Met Met Val Trp Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val
35 40 45

Ser Ser Ile Tyr Pro Ser Gly Gly Ile Thr Phe Tyr Ala Asp Trp Lys
50 55 60

Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr Leu
65 70 75 80

Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys Ala
85 90 95

Arg Ile Lys Leu Gly Thr Val Thr Thr Val Asp Tyr Trp Gly Gln Gly
100 105 110

Thr Leu Val Thr Val Ser Ser
115

<210> 45

<211> 110

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 45

Gln Ser Ala Leu Thr Gln Pro Ala Ser Val Ser Gly Ser Pro Gly Gln
1 5 10 15

Ser Ile Thr Ile Ser Cys Thr Gly Thr Ser Ser Asp Val Gly Gly Tyr
20 25 30

Asn Tyr Val Ser Trp Tyr Gln Gln His Pro Gly Lys Ala Pro Lys Leu
35 40 45

Met Ile Tyr Asp Val Ser Asn Arg Pro Ser Gly Val Ser Asn Arg Phe
50 55 60

Ser Gly Ser Lys Ser Gly Asn Thr Ala Ser Leu Thr Ile Ser Gly Leu
65 70 75 80

Gln Ala Glu Asp Glu Ala Asp Tyr Tyr Cys Ser Ser Tyr Thr Ser Ser
85 90 95

Ser Thr Arg Val Phe Gly Thr Gly Thr Lys Val Thr Val Leu
100 105 110

<210> 46
<211> 120
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 46

Glu Val Gln Leu Leu Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
1 5 10 15

Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Met Tyr
20 25 30

Met Met Met Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Val Trp
35 40 45

Ser Ser Ile Tyr Pro Ser Gly Gly Ile Thr Phe Tyr Ala Asp Ser Val
50 55 60

Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr
65 70 75 80

Leu Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Ile Tyr Tyr Cys
85 90 95

Ala Arg Ile Lys Leu Gly Thr Val Thr Thr Val Asp Tyr Trp Gly Gln
100 105 110

Gly Thr Leu Val Thr Val Ser Ser
115 120

<210> 47
<211> 110

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 47

Gln Ser Ala Leu Thr Gln Pro Ala Ser Val Ser Gly Ser Pro Gly Gln
1 5 10 15

Ser Ile Thr Ile Ser Cys Thr Gly Thr Ser Ser Asp Val Gly Ala Tyr
20 25 30

Asn Tyr Val Ser Trp Tyr Gln Gln His Pro Gly Lys Ala Pro Lys Leu
35 40 45

Met Ile Tyr Asp Val Ser Asn Arg Pro Ser Gly Val Ser Asn Arg Phe
50 55 60

Ser Gly Ser Lys Ser Gly Asn Thr Ala Ser Leu Thr Ile Ser Gly Leu
65 70 75 80

Gln Ala Glu Asp Glu Ala Asp Tyr Tyr Cys Ser Ser Tyr Thr Ser Ser
85 90 95

Ser Thr Arg Val Phe Gly Thr Gly Thr Lys Val Thr Val Leu
100 105 110

<210> 48

<211> 1407

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polynucleotide
from
human Fab library

<400> 48

atggagttgc ctgttaggct gttgggtgctg atgtttctgga ttcctgctag ctccagcgag
60

gtgcagctgc tggaatccgg cggaggactg gtgcagcctg gcggctccct gagactgtct
120

tgcgccgcct ccggcttcac cttctccagc tacatcatga tgtgggtgcg acaggcccct
180

ggcaagggcc tggaatgggt gtcctccatc taccctccg gcggcatcac cttctacgcc
240

gacaccgtga agggccggtt caccatctcc cgggacaact ccaagaacac cctgtacctg
300

cagatgaact ccctgcgggc cgaggacacc gccgtgtact actgcgcccg gatcaagctg
360

ggcaccgtga ccaccgtgga ctactggggc cagggcaccc tgggtgacagt gtcctccgcc
420

tccaccaagg gcccatcggt cttccccctg gcaccctcct ccaagagcac ctctgggggc
480

acagcggccc tgggctgcct ggtcaaggac tacttccccg aaccggtgac ggtgtcgtgg
540

aactcaggcg ccctgaccag cggcgtgcac accttccccg ctgtcctaca gtcctcagga
600

ctctactccc tcagcagcgt ggtgaccgtg ccctccagca gcttgggcac ccagacctac
660

atctgcaacg tgaatcacia gccagcaac accaagggtgg acaagaaagt tgagcccaaa
720

tcttgtgaca aaactcacac atgcccaccg tgcccagcac ctgaactcct ggggggaccg
780

tcagtcttcc tcttcccccc aaaaccaag gacaccctca tgatctcccg gaccctgag
840

gtcacatgcg tgggtggtgga cgtgagccac gaagaccctg aggtcaagtt caactggtac
900

gtggacggcg tggaggtgca taatgccaag acaaagccgc gggaggagca gtacaacagc
960

acgtaccgtg tggtcagcgt cctcaccgtc ctgcaccagg actggctgaa tggcaaggag
1020

tacaagtgca aggtctccaa caaagccctc ccagccccc a tcgagaaaac catctccaaa
1080

gccaaagggc agccccgaga accacaggtg tacaccctgc ccccatcacg ggatgagctg
1140

accaagaacc aggtcagcct gacctgcctg gtcaaaggct tctatcccag cgacatcgcc
1200

gtggagtggg agagcaatgg gcagccggag aacaactaca agaccacgcc tcccgtgctg
1260

gactccgacg gctccttctt cctctatagc aagctcaccg tggacaagag caggtggcag
1320

caggggaacg tcttctcatg ctccgtgatg catgaggctc tgcacaacca ctacacgcag
1380

a a g a g c c t c t c c c t g t c c c c g g g t a a a
1407

<210> 49

<211> 705

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polynucleotide
from
human Fab library

<400> 49

atggagttagc ctgtaggct gttgggtgctg atgttctgga ttcctgcttc ctttaagccag
60

tccgccctga cccagcctgc ctccgtgtct ggctcccctg gccagtccat caccatcagc
120

tgcaccggca cctccagcga cgtgggcggc tacaactacg tgtcctggta tcagcagcac
180

cccggcaagg cccccaagct gatgatctac gacgtgtcca accggccctc cggcgtgtcc
240

aacagattct ccggctccaa gtccggcaac accgcctccc tgaccatcag cggactgcag
300

gcagaggacg aggccgacta ctactgctcc tcctacacct cctccagcac cagagtgttc
360

ggcaccggca caaaagtgac cgtgctgggc cagcccaagg ccaacccaac cgtgacactg
420

ttccccccat cctccgagga actgcaggcc aacaaggcca ccctgggtctg cctgatctca
480

gatttctatc caggcgccgt gaccgtggcc tggaaggctg atggctcccc agtgaaggcc
540

ggcgtggaaa ccaccaagcc ctccaagcag tccaacaaca aatacgccgc ctctcctac
600

ctgtccctga ccccgagca gtggaagtcc caccggctct acagctgcca ggtcacacac
660

gagggctcca ccgtggaaaa gaccgtcgcc cccaccgagt gctca
705

<210> 50

<211> 117

<212> PRT

<213> Homo sapiens

<400> 50

Gly	Ala	Val	Lys	Phe	Pro	Gln	Leu	Cys	Lys	Phe	Cys	Asp	Val	Arg	Phe
1				5					10					15	

Ser	Thr	Cys	Asp	Asn	Gln	Lys	Ser	Cys	Met	Ser	Asn	Cys	Ser	Ile	Thr
			20					25					30		

Ser Ile Cys Glu Lys Pro Gln Glu Val Cys Val Ala Val Trp Arg Lys
35 40 45

Asn Asp Glu Asn Ile Thr Leu Glu Thr Val Cys His Asp Pro Lys Leu
50 55 60

Pro Tyr His Asp Phe Ile Leu Glu Asp Ala Ala Ser Pro Lys Cys Ile
65 70 75 80

Met Lys Glu Lys Lys Lys Pro Gly Glu Thr Phe Phe Met Cys Ser Cys
85 90 95

Ser Ser Asp Glu Cys Asn Asp Asn Ile Ile Phe Ser Glu Glu Tyr Asn
100 105 110

Thr Ser Asn Pro Asp
115

<210> 51
<211> 115
<212> PRT
<213> Homo sapiens

<400> 51

Val Lys Phe Pro Gln Leu Cys Lys Phe Cys Asp Val Arg Phe Ser Thr
1 5 10 15

Cys Asp Asn Gln Lys Ser Cys Met Ser Asn Cys Ser Ile Thr Ser Ile
20 25 30

Cys Glu Lys Pro Gln Glu Val Cys Val Ala Val Trp Arg Lys Asn Asp
35 40 45

Glu Asn Ile Thr Leu Glu Thr Val Cys His Asp Pro Lys Leu Pro Tyr
50 55 60

His Asp Phe Ile Leu Glu Asp Ala Ala Ser Pro Lys Cys Ile Met Lys
65 70 75 80

Glu Lys Lys Lys Pro Gly Glu Thr Phe Phe Met Cys Ser Cys Ser Ser
85 90 95

Asp Glu Cys Asn Asp Asn Ile Ile Phe Ser Glu Glu Tyr Asn Thr Ser
100 105 110

Asn Pro Asp
115

<210> 52
<211> 122
<212> PRT
<213> Homo sapiens

<400> 52

Val Thr Asp Asn Asn Gly Ala Val Lys Phe Pro Gln Leu Cys Lys Phe
1 5 10 15

Cys Asp Val Arg Phe Ser Thr Cys Asp Asn Gln Lys Ser Cys Met Ser
20 25 30

Asn Cys Ser Ile Thr Ser Ile Cys Glu Lys Pro Gln Glu Val Cys Val
35 40 45

Ala Val Trp Arg Lys Asn Asp Glu Asn Ile Thr Leu Glu Thr Val Cys
50 55 60

His Asp Pro Lys Leu Pro Tyr His Asp Phe Ile Leu Glu Asp Ala Ala
65 70 75 80

Ser Pro Lys Cys Ile Met Lys Glu Lys Lys Lys Pro Gly Glu Thr Phe
85 90 95

Phe Met Cys Ser Cys Ser Ser Asp Glu Cys Asn Asp Asn Ile Ile Phe
100 105 110

Ser Glu Glu Tyr Asn Thr Ser Asn Pro Asp
115 120

<210> 53

<211> 110

<212> PRT

<213> Homo sapiens

<400> 53

Leu Cys Lys Phe Cys Asp Val Arg Phe Ser Thr Cys Asp Asn Gln Lys
1 5 10 15

Ser Cys Met Ser Asn Cys Ser Ile Thr Ser Ile Cys Glu Lys Pro Gln
20 25 30

Glu Val Cys Val Ala Val Trp Arg Lys Asn Asp Glu Asn Ile Thr Leu
35 40 45

Glu Thr Val Cys His Asp Pro Lys Leu Pro Tyr His Asp Phe Ile Leu
50 55 60

Glu Asp Ala Ala Ser Pro Lys Cys Ile Met Lys Glu Lys Lys Lys Pro
65 70 75 80

Gly Glu Thr Phe Phe Met Cys Ser Cys Ser Ser Asp Glu Cys Asn Asp
85 90 95

Asn Ile Ile Phe Ser Glu Glu Tyr Asn Thr Ser Asn Pro Asp
100 105 110

<210> 54

<211> 122

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 54

Val Thr Asp Asn Ala Gly Ala Val Lys Phe Pro Gln Leu Cys Lys Phe
1 5 10 15

Cys Asp Val Arg Phe Ser Thr Cys Asp Asn Gln Lys Ser Cys Met Ser
20 25 30

Asn Cys Ser Ile Thr Ser Ile Cys Glu Lys Pro Gln Glu Val Cys Val
35 40 45

Ala Val Trp Arg Lys Asn Asp Glu Asn Ile Thr Leu Glu Thr Val Cys
50 55 60

His Asp Pro Lys Leu Pro Tyr His Asp Phe Ile Leu Glu Asp Ala Ala
65 70 75 80

Ser Pro Lys Cys Ile Met Lys Glu Lys Lys Lys Pro Gly Glu Thr Phe
85 90 95

Phe Met Cys Ser Cys Ser Ser Asp Glu Cys Asn Asp Asn Ile Ile Phe
100 105 110

Ser Glu Glu Tyr Asn Thr Ser Asn Pro Asp
115 120

<210> 55
<211> 118
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 55

Gln Val Gln Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Thr Val Ser Gly Gly Ser Ile Ser Asn Asp
20 25 30

Tyr Trp Thr Trp Ile Arg Gln His Pro Gly Lys Gly Leu Glu Tyr Ile
35 40 45

Gly Tyr Ile Ser Tyr Thr Gly Ser Thr Tyr Tyr Asn Pro Ser Leu Lys
50 55 60

Ser Arg Val Thr Ile Ser Arg Asp Thr Ser Lys Asn Gln Phe Ser Leu
65 70 75 80

Lys Leu Ser Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr Cys Ala
85 90 95

Arg Ser Gly Gly Trp Leu Ala Pro Phe Asp Tyr Trp Gly Arg Gly Thr
100 105 110

Leu Val Thr Val Ser Ser
115

<210> 56
<211> 113

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 56

Asp Ile Val Met Thr Gln Ser Pro Asp Ser Leu Ala Val Ser Leu Gly
1 5 10 15

Glu Arg Ala Thr Ile Asn Cys Lys Ser Ser Gln Ser Leu Phe Tyr His
20 25 30

Ser Asn Gln Lys His Ser Leu Ala Trp Tyr Gln Gln Lys Pro Gly Gln
35 40 45

Pro Pro Lys Leu Leu Ile Tyr Gly Ala Ser Thr Arg Glu Ser Gly Val
50 55 60

Pro Asp Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr
65 70 75 80

Ile Ser Ser Leu Gln Ala Glu Asp Val Ala Val Tyr Tyr Cys Gln Gln
85 90 95

Tyr Tyr Gly Tyr Pro Tyr Thr Phe Gly Gly Gly Thr Lys Val Glu Ile
100 105 110

Lys

<210> 57

<211> 119

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 57

Gln Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Val Ser Cys Lys Ala Ser Gly Tyr Thr Phe Thr Ser Tyr
20 25 30

Trp Met His Trp Val Arg Gln Ala Pro Gly Gln Gly Leu Glu Trp Met
35 40 45

Gly Arg Ile Gly Pro Asn Ser Gly Phe Thr Ser Tyr Asn Glu Lys Phe
50 55 60

Lys Asn Arg Val Thr Met Thr Arg Asp Thr Ser Thr Ser Thr Val Tyr
65 70 75 80

Met Glu Leu Ser Ser Leu Arg Ser Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Gly Gly Ser Ser Tyr Asp Tyr Phe Asp Tyr Trp Gly Gln Gly
100 105 110

Thr Thr Val Thr Val Ser Ser
115

<210> 58

<211> 111

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 58

Asp Ile Val Leu Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Pro Gly
1 5 10 15

Gln Arg Ala Thr Ile Thr Cys Arg Ala Ser Glu Ser Val Ser Ile His
20 25 30

Gly Thr His Leu Met His Trp Tyr Gln Gln Lys Pro Gly Gln Pro Pro
35 40 45

Lys Leu Leu Ile Tyr Ala Ala Ser Asn Leu Glu Ser Gly Val Pro Ala
50 55 60

Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile Asn
65 70 75 80

Pro Val Glu Ala Glu Asp Thr Ala Asn Tyr Tyr Cys Gln Gln Ser Phe
85 90 95

Glu Asp Pro Leu Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys
100 105 110

<210> 59

<211> 445

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 59

Gln Val Gln Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln
1 5 10 15

Thr Leu Ser Leu Thr Cys Thr Val Ser Gly Gly Ser Ile Ser Asn Asp

20

25

30

Tyr Trp Thr Trp Ile Arg Gln His Pro Gly Lys Gly Leu Glu Tyr Ile
 35 40 45

Gly Tyr Ile Ser Tyr Thr Gly Ser Thr Tyr Tyr Asn Pro Ser Leu Lys
 50 55 60

Ser Arg Val Thr Ile Ser Arg Asp Thr Ser Lys Asn Gln Phe Ser Leu
 65 70 75 80

Lys Leu Ser Ser Val Thr Ala Ala Asp Thr Ala Val Tyr Tyr Cys Ala
 85 90 95

Arg Ser Gly Gly Trp Leu Ala Pro Phe Asp Tyr Trp Gly Arg Gly Thr
 100 105 110

Leu Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser Val Phe Pro
 115 120 125

Leu Ala Pro Cys Ser Arg Ser Thr Ser Glu Ser Thr Ala Ala Leu Gly
 130 135 140

Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser Trp Asn
 145 150 155 160

Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val Leu Gln
 165 170 175

Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro Ser Ser
 180 185 190

Ser Leu Gly Thr Lys Thr Tyr Thr Cys Asn Val Asp His Lys Pro Ser
 195 200 205

Asn Thr Lys Val Asp Lys Arg Val Glu Ser Lys Tyr Gly Pro Pro Cys
210 215 220

Pro Pro Cys Pro Ala Pro Glu Ala Ala Gly Gly Pro Ser Val Phe Leu
225 230 235 240

Phe Pro Pro Lys Pro Lys Asp Thr Leu Met Ile Ser Arg Thr Pro Glu
245 250 255

Val Thr Cys Val Val Val Asp Val Ser Gln Glu Asp Pro Glu Val Gln
260 265 270

Phe Asn Trp Tyr Val Asp Gly Val Glu Val His Asn Ala Lys Thr Lys
275 280 285

Pro Arg Glu Glu Gln Phe Asn Ser Thr Tyr Arg Val Val Ser Val Leu
290 295 300

Thr Val Leu His Gln Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys
305 310 315 320

Val Ser Asn Lys Gly Leu Pro Ser Ser Ile Glu Lys Thr Ile Ser Lys
325 330 335

Ala Lys Gly Gln Pro Arg Glu Pro Gln Val Tyr Thr Leu Pro Pro Ser
340 345 350

Gln Glu Glu Met Thr Lys Asn Gln Val Ser Leu Thr Cys Leu Val Lys
355 360 365

Gly Phe Tyr Pro Ser Asp Ile Ala Val Glu Trp Glu Ser Asn Gly Gln
370 375 380

Pro Glu Asn Asn Tyr Lys Thr Thr Pro Pro Val Leu Asp Ser Asp Gly
385 390 395 400

Ser Phe Phe Leu Tyr Ser Arg Leu Thr Val Asp Lys Ser Arg Trp Gln
405 410 415

Glu Gly Asn Val Phe Ser Cys Ser Val Met His Glu Ala Leu His Asn
420 425 430

His Tyr Thr Gln Lys Ser Leu Ser Leu Ser Leu Gly Lys
435 440 445

<210> 60

<211> 220

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 60

Asp Ile Val Met Thr Gln Ser Pro Asp Ser Leu Ala Val Ser Leu Gly
1 5 10 15

Glu Arg Ala Thr Ile Asn Cys Lys Ser Ser Gln Ser Leu Phe Tyr His
20 25 30

Ser Asn Gln Lys His Ser Leu Ala Trp Tyr Gln Gln Lys Pro Gly Gln
35 40 45

Pro Pro Lys Leu Leu Ile Tyr Gly Ala Ser Thr Arg Glu Ser Gly Val
50 55 60

Pro Asp Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr
65 70 75 80

Ile Ser Ser Leu Gln Ala Glu Asp Val Ala Val Tyr Tyr Cys Gln Gln
85 90 95

Tyr Tyr Gly Tyr Pro Tyr Thr Phe Gly Gly Gly Thr Lys Val Glu Ile
100 105 110

Lys Arg Thr Val Ala Ala Pro Ser Val Phe Ile Phe Pro Pro Ser Asp
115 120 125

Glu Gln Leu Lys Ser Gly Thr Ala Ser Val Val Cys Leu Leu Asn Asn
130 135 140

Phe Tyr Pro Arg Glu Ala Lys Val Gln Trp Lys Val Asp Asn Ala Leu
145 150 155 160

Gln Ser Gly Asn Ser Gln Glu Ser Val Thr Glu Gln Asp Ser Lys Asp
165 170 175

Ser Thr Tyr Ser Leu Ser Ser Thr Leu Thr Leu Ser Lys Ala Asp Tyr
180 185 190

Glu Lys His Lys Val Tyr Ala Cys Glu Val Thr His Gln Gly Leu Ser
195 200 205

Ser Pro Val Thr Lys Ser Phe Asn Arg Gly Glu Cys
210 215 220

<210> 61

<211> 446

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 61

Gln Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Val Ser Cys Lys Ala Ser Gly Tyr Thr Phe Thr Ser Tyr
20 25 30

Trp Met His Trp Val Arg Gln Ala Pro Gly Gln Gly Leu Glu Trp Met
35 40 45

Gly Arg Ile Gly Pro Asn Ser Gly Phe Thr Ser Tyr Asn Glu Lys Phe
50 55 60

Lys Asn Arg Val Thr Met Thr Arg Asp Thr Ser Thr Ser Thr Val Tyr
65 70 75 80

Met Glu Leu Ser Ser Leu Arg Ser Glu Asp Thr Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Gly Gly Ser Ser Tyr Asp Tyr Phe Asp Tyr Trp Gly Gln Gly
100 105 110

Thr Thr Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser Val Phe
115 120 125

Pro Leu Ala Pro Cys Ser Arg Ser Thr Ser Glu Ser Thr Ala Ala Leu
130 135 140

Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser Trp
145 150 155 160

Asn Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val Leu
165 170 175

Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro Ser
180 185 190

Ser Ser Leu Gly Thr Lys Thr Tyr Thr Cys Asn Val Asp His Lys Pro
195 200 205

Ser Asn Thr Lys Val Asp Lys Arg Val Glu Ser Lys Tyr Gly Pro Pro
210 215 220

Cys Pro Pro Cys Pro Ala Pro Glu Ala Ala Gly Gly Pro Ser Val Phe
225 230 235 240

Leu Phe Pro Pro Lys Pro Lys Asp Thr Leu Met Ile Ser Arg Thr Pro
245 250 255

Glu Val Thr Cys Val Val Val Asp Val Ser Gln Glu Asp Pro Glu Val
260 265 270

Gln Phe Asn Trp Tyr Val Asp Gly Val Glu Val His Asn Ala Lys Thr
275 280 285

Lys Pro Arg Glu Glu Gln Phe Asn Ser Thr Tyr Arg Val Val Ser Val
290 295 300

Leu Thr Val Leu His Gln Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys
305 310 315 320

Lys Val Ser Asn Lys Gly Leu Pro Ser Ser Ile Glu Lys Thr Ile Ser
325 330 335

Lys Ala Lys Gly Gln Pro Arg Glu Pro Gln Val Tyr Thr Leu Pro Pro
340 345 350

Ser Gln Glu Glu Met Thr Lys Asn Gln Val Ser Leu Thr Cys Leu Val
355 360 365

Lys Gly Phe Tyr Pro Ser Asp Ile Ala Val Glu Trp Glu Ser Asn Gly
370 375 380

Gln Pro Glu Asn Asn Tyr Lys Thr Thr Pro Pro Val Leu Asp Ser Asp
385 390 395 400

Gly Ser Phe Phe Leu Tyr Ser Arg Leu Thr Val Asp Lys Ser Arg Trp
405 410 415

Gln Glu Gly Asn Val Phe Ser Cys Ser Val Met His Glu Ala Leu His
420 425 430

Asn His Tyr Thr Gln Lys Ser Leu Ser Leu Ser Leu Gly Ala
435 440 445

<210> 62

<211> 218

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic polypeptide

<400> 62

Asp Ile Val Leu Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Pro Gly
1 5 10 15

Gln Arg Ala Thr Ile Thr Cys Arg Ala Ser Glu Ser Val Ser Ile His
20 25 30

Gly Thr His Leu Met His Trp Tyr Gln Gln Lys Pro Gly Gln Pro Pro
35 40 45

Lys Leu Leu Ile Tyr Ala Ala Ser Asn Leu Glu Ser Gly Val Pro Ala
50 55 60

Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile Asn
65 70 75 80

Pro Val Glu Ala Glu Asp Thr Ala Asn Tyr Tyr Cys Gln Gln Ser Phe
85 90 95

Glu Asp Pro Leu Thr Phe Gly Gln Gly Thr Lys Leu Glu Ile Lys Arg
100 105 110

Thr Val Ala Ala Pro Ser Val Phe Ile Phe Pro Pro Ser Asp Glu Gln
115 120 125

Leu Lys Ser Gly Thr Ala Ser Val Val Cys Leu Leu Asn Asn Phe Tyr
130 135 140

Pro Arg Glu Ala Lys Val Gln Trp Lys Val Asp Asn Ala Leu Gln Ser
145 150 155 160

Gly Asn Ser Gln Glu Ser Val Thr Glu Gln Asp Ser Lys Asp Ser Thr
165 170 175

Tyr Ser Leu Ser Ser Thr Leu Thr Leu Ser Lys Ala Asp Tyr Glu Lys
180 185 190

His Lys Val Tyr Ala Cys Glu Val Thr His Gln Gly Leu Ser Ser Pro
195 200 205

Val Thr Lys Ser Phe Asn Arg Gly Glu Cys
210 215

