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DESCRIPTION

Description

[0001] This application claims priority to U.S. Serial No. 15/906,481 filed on February 27, 2018. Serial No. 15/906,481 is a continuation-in-part of Serial No. 15/705,333 filed on September 15, 2017 and claims priority to U.S. Serial No. 62/395,195 filed on September 15, 2016.

[0002] This application incorporates by reference the contents of a 1.38 kb text file created on February 27, 2018 and named "00047900254sequencelisting.txt," which is the sequence listing for this application.

TECHNICAL FIELD

[0003] This disclosure relates generally to immunomodulatory peptides.

BRIEF DESCRIPTION OF THE DRAWINGS

[0004]

Figure 1. Graph showing saturatable binding of anti-human PD-1 antibody to Jurkat cells.

Figure 2. Graph showing saturatable binding of PD-L1 Fc to Jurkat cells.

Figures 3A-B. Graphs showing effect of peptide QP20 on binding of PD-L1 to PD-1. **Figure 3A**, MFI; **Figure 3B**, normalized mean fluorescence intensity (MFI).

Figures 4A-B. Graphs showing effect of peptide HD20 on binding of PD-L1 to PD-1. **Figure 4A**, MFI; **Figure 4B**, normalized MFI.

Figures 5A-B. Graphs showing effect of peptide WQ20 on binding of PD-L1 to PD-1. **Figure 5A**, MFI; **Figure 5B**, normalized MFI.

Figures 6A-B. Graphs showing effect of peptide SQ20 on binding of PD-L1 to PD-1. **Figure 6A**, MFI; **Figure 6B**, normalized MFI.

Figure 7A. Graph showing the effect of an anti-human PD-1 antibody on the interaction between PD-1-expressing Jurkat T cells and PD-L1-expressing CHO cells that results in inhibition of a PD-1 mediated suppression of luciferase reporter that is under the control of promoter containing IL-2, NFAT, and NF-kB response elements.

Figure 7B. Graph showing the effect of an anti-human PD-1 antibody on the interaction between PD-1-expressing Jurkat T cells and PD-L1-expressing CHO cells (data in 7B expressed as fold inhibition).

Figure 8A. Graph showing that PD-1 peptide inhibitors inhibit, in a dose-dependent manner, the interaction between PD-1-expressing Jurkat T cells and PD-L1-expressing CHO cells, which results in increased luciferase reporter expression,.

Figure 8B. Graph showing the effect of an anti-human PD-1 antibody on the interaction between PD-1-expressing Jurkat T cells and PD-L1-expressing CHO cells (data in 8B expressed as fold inhibition).

Figure 9. Graph showing IL-2 production by peripheral blood mononuclear cells (PBMCs) in a tetanus toxoid recall assay after culture with peptides QP20, HD20, WQ20, SQ20, or CQ-22.

Figure 10. Graph showing IL-4 production by PBMCs in a tetanus toxoid recall assay after culture with peptides QP20, HD20, WQ20, SQ20, or CQ-22.

Figure 11. Graph showing IL-6 production by PBMCs in a tetanus toxoid recall assay after culture with peptides QP20, HD20, WQ20, SQ20, or CQ-22.

Figure 12. Graph showing IL-10 production by PBMCs in a tetanus toxoid recall assay after culture with peptides QP20, HD20, WQ20, SQ20, or CQ-22.

Figure 13. Graph showing IL-17a production by PBMCs in a tetanus toxoid recall assay, after culture with peptides QP20, HD20, WQ20, SQ20, or CQ-22.

Figure 14. Graph showing IFN γ production by PBMCs in a tetanus toxoid recall assay, after culture with peptides QP20, HD20, WQ20, SQ20, or CQ-22.

Figure 15. Graph showing TNF α production by PBMCs in a tetanus toxoid recall assay, after culture with peptides QP20, HD20, WQ20, SQ20, or CQ-22.

Figure 16. Graph showing IL-2 production by PBMCs in a tetanus toxoid recall assay, after culture with various combinations of peptides QP20, HD20, WQ20, and SQ20.

Figure 17. Graph showing IL-4 production by PBMCs in a tetanus toxoid recall assay, after culture with various combinations of peptides QP20, HD20, WQ20, and SQ20.

Figure 18. Graph showing IL-6 production by PBMCs in a tetanus toxoid recall assay, after culture with various combinations of peptides QP20, HD20, WQ20, and SQ20.

Figure 19. Graph showing IL-10 production by PBMCs in a tetanus toxoid recall assay, after stimulation with various combinations of peptides QP20, HD20, WQ20, and SQ20.

Figure 20. Graph showing IL-17a production by PBMCs after stimulation with various combinations of peptides QP20, HD20, WQ20, and SQ20.

Figure 21. Graph showing IFN γ production by PBMCs after culture with various combinations

of peptides QP20, HD20, WQ20, and SQ20.

Figure 22. Graph showing TNF α production by PBMCs after culture with various combinations of peptides QP20, HD20, WQ20, and SQ20.

Figure 23A. Graph showing IL-2 production by PBMCs from donor A after culture with peptides QP20, HD20, WQ20, and SQ20, or CQ-22.

Figure 23B. Graph showing IL-2 production by PBMCs from donor B after culture with peptides QP20, HD20, WQ20, or SQ20 and combinations of these peptides.

Figure 24A. Graph showing IL-17a production by PBMCs from donor A after culture with peptides QP20, HD20, WQ20, and SQ20, or CQ-22.

Figure 24B. Graph showing IL-17a production by PBMCs from donor B after culture with peptides QP20, HD20, WQ20, or SQ20 and combinations of these peptides.

Figure 25. Graph showing number of surface metastases in mice bearing B 16-F10-LacZ tumor cells and treated with combinations of peptides.

Figure 26. Graph showing the average number $\pm$ standard deviation of *Plasmodium yoelii* circumsporozoite protein (PyCS)-specific, IFN γ -secreting CD8 T cells per 0.5×10^6 splenocytes for each cohort tested in Example 8.

Figure 27. Graph showing the effect of the combination of QP20, HD20, WQ20, and SQ20 peptides on the mean level of serum HBsAg (hepatitis B surface antigen) at weeks 2 and 3 post infection.

DETAILED DESCRIPTION

[0005] This disclosure provides four peptides:

peptide	amino acid sequence	SEQ ID NO:
QP20	QTRTVPMPKI HH PPWQNVVP	1
HD20	HHH QVYQVRSHWTGMHSGHD	2
WQ20	WNLPASFEIN HH IRPHEHEWIQ	3
SQ20	SSY HH FKMPELHFGKNTFHQ	4

[0006] These peptides share a core sequence of HH_, which is shown above in bold, and have a strong affinity for the checkpoint receptor "programmed death 1" (PD-1). These peptides block the interaction of PD-1 with its ligand PD-L1 and can therefore be used to inhibit the progression of a hyperproliferative disorder, including cancer, or to treat infectious diseases,

including persistent infections by agents such as HIV, hepatitis B virus (HBV), hepatitis C virus (HCV), and *Plasmodium falciparum*, by enhancing, stimulating, and/or increasing an individual's immune response.

[0007] In some embodiments, any of the disclosed peptides can be modified using chemical or recombinant methods to enhance stability or other pharmacokinetic properties. See, e.g., US 2017/0020956. Modifications include, but are not limited to, replacement of one or more L-amino acid with its corresponding D-form, acetylation on a C- and/or N-terminal residue, amidation on a C- and/or N-terminal residue, cyclization, esterification, glycosylation, acylation, attachment of myristic or palmitic acid, addition of an N-terminal glycine, addition of lipophilic moieties such as long fatty acid chains, and PEGylation.

[0008] In some embodiments, one or more of the disclosed peptides can be conjugated to various moieties, such as albumin and transthyretin, to enhance the plasma half-life of the peptide(s). Methods of preparing such conjugates are well known in the art (e.g., Penchala *et al.*, 2015; Kontermann, 2016; Zorzi *et al.*, 2017).

[0009] In some embodiments, any of the disclosed peptides can be conjugated to a partner molecule, such as a peptide or protein such as an antibody intended to increase the half-life *in vivo* and/or to provide specific delivery to a target tissue or cell. Conjugation may be direct or can be via a linker. In some of these embodiments, the peptide can be modified to substitute one or more amino acids with amino acids used to attach partner molecules, such as lysine, or by N-terminal extension of the peptide with, e.g., 1, 2, 3, or 4 glycine spacer molecules.

[0010] Peptides, or modified versions of the peptides as described above, can be made by any method known in the art, including synthetic methods, recombinant methods, or both. Synthetic methods include solid-phase and solution methods, and may include the use of protective groups. See, e.g., Bodanszky *et al.* (1976), McOmie (1973), Merrifield (1963), Neurath *et al.* (1976), Stuart & Young (1984).

[0011] Recombinant production of peptides can be carried out using any nucleotide sequence(s) encoding the peptides in any suitable expression system. Nucleic acid molecules encoding one or more of the disclosed peptides can be incorporated into an expression cassette that includes control elements operably linked to the coding sequences. Control elements include, but are not limited to, initiators, promoters (including inducible, repressible, and constitutive promoters), enhancers, and polyadenylation signals. Signal sequences can be included. The expression cassette can be provided in a vector that can be introduced into an appropriate host cell for production of the peptide(s). Methods of constructing expression cassettes and expression vectors are well known. Expression vectors can include one or more of the expression cassettes described in the paragraphs below.

[0012] In some embodiments, an expression cassette encodes a peptide comprising the amino acid sequence SEQ ID NO:1. In some embodiments, an expression cassette encodes a peptide consisting essentially of the amino acid sequence SEQ ID NO:1. In some

embodiments, an expression cassette encodes a peptide consisting of the amino acid sequence SEQ ID NO:1.

[0013] In some embodiments, an expression cassette encodes a peptide comprising the amino acid sequence SEQ ID NO:2. In some embodiments, an expression cassette encodes a peptide consisting essentially of the amino acid sequence SEQ ID NO:2. In some embodiments, an expression cassette encodes a peptide consisting of the amino acid sequence SEQ ID NO:2.

[0014] In some embodiments, an expression cassette encodes a peptide comprising the amino acid sequence SEQ ID NO:3. In some embodiments, an expression cassette encodes a peptide consisting essentially of the amino acid sequence SEQ ID NO:3. In some embodiments, an expression cassette encodes a peptide consisting of the amino acid sequence SEQ ID NO:3.

[0015] In some embodiments, an expression cassette encodes a peptide comprising the amino acid sequence SEQ ID NO:4. In some embodiments, an expression cassette encodes a peptide consisting essentially of the amino acid sequence SEQ ID NO:4. In some embodiments, an expression cassette encodes a peptide consisting of the amino acid sequence SEQ ID NO:4.

[0016] In some embodiments, an expression cassette encodes only two of the disclosed peptides; e.g., a peptide (a) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:1 and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:2; (b) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:1 and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:3; (c) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:1 and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:4; (d) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:2 and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:3; (e) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:2 and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:4; or (f) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:3 and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:4.

[0017] In some embodiments, an expression cassette encodes only three of the disclosed peptides; e.g., (a) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:1, a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:2, and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:3; (b) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:1, a peptide comprising,

consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:2, and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:4; (c) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:2, a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:3, and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:4; or (d) a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:1, a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:3, and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:4.

[0018] In some embodiments, an expression cassette encodes all four of the disclosed peptides; *i.e.*, a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO: 1, a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:2, a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:3, and a peptide comprising, consisting essentially of, or consisting of the amino acid sequence SEQ ID NO:4.

Therapeutic Uses

[0019] The disclosed peptides have a number of therapeutic applications and can be administered for a variety of purposes to both human and veterinary subjects. "Administer" as used herein includes direct administration of the disclosed peptides or modified versions thereof as well as indirect administration, *e.g.*, using a nucleic acid molecule encoding the peptides or modified versions of the peptides, as described below. In some embodiments, administration is carried out in conjunction with one or more other therapeutic moieties. "In conjunction with" includes administration together with, before, or after administration of the one or more other therapeutic moieties.

Treatment of Hyperproliferative Disorders, Including Cancer

[0020] In some embodiments, one or more of the disclosed peptides or modified versions thereof can be administered to inhibit the progression of a hyperproliferative disorder, such as cancer. Such inhibition may include, for example, reducing proliferation of neoplastic or pre-neoplastic cells; destroying neoplastic or pre-neoplastic cells; and inhibiting metastasis or decreasing the size of a tumor.

[0021] Examples of cancers that can be treated using one or more of the disclosed peptides or modified versions thereof include, but are not limited to, melanomas, lymphomas, sarcomas, and cancers of the colon, kidney, stomach, bladder, brain (*e.g.*, gliomas, glioblastomas, astrocytomas, medulloblastomas), prostate, bladder, rectum, esophagus, pancreas, liver, lung, breast, uterus, cervix, ovary, blood (*e.g.*, acute myeloid leukemia, acute lymphoid leukemia,

chronic myeloid leukemia, chronic lymphocytic leukemia, Burkitt's lymphoma, EBV-induced B-cell lymphoma).

Combination Cancer Therapies

[0022] In some embodiments, one or more of the disclosed peptides or modified versions thereof can be administered in conjunction with one or more therapies or immunotherapies, such as those described below.

[0023] In some embodiments, the second therapy comprises a second agent that reduces or blocks the activity of PD-1 (e.g., nivolumab, pembrolizumab, durvalumab) or

[0024] In some embodiments, the second therapy comprises an agent that reduces or blocks the activity of PD-L1 (e.g., atezolizumab).

[0025] In some embodiments, the second therapy comprises an agent that reduces or blocks the activity of other inhibitory checkpoint molecules and/or molecules that suppress the immune system. These molecules include, but are not limited to:

1. 1. Lymphocyte-activation gene-3 (LAG-3; see He et al., 2016; Triebel et al., 1990);
2. 2. cytotoxic T-lymphocyte-associated antigen 4 (CTLA-4);
3. 3. V-domain Immunoglobulin Suppressor of T cell Activation (VISTA, also known as c10orf54, PD-1H, DD1 α , Gi24, Dies1, and SISP1; see US 2017/0334990, US 2017/0112929, Gao et al., 2017, Wang et al., 2011; Liu et al., 2015);
4. 4. T-cell Immunoglobulin domain and Mucin domain 3 (TIM-3; see US 2017/0198041, US 2017/0029485, US 2014/0348842, Sakuishi et al., 2010);
5. 5. killer immunoglobulin-like receptors (KIRs; see US 2015/0290316);
6. 6. agents that inhibit indoleamine (2,3)-dioxygenase (IDO; see Mellemegaard et al., 2017);
7. 7. B and T Lymphocyte Attenuator (BTLA; see US 2016/09222114); and
8. 8. A2A adenosine receptor (A2AR; see Beavis et al., 2015; US 2013/0267515; US 2017/0166878; Leone et al., 2015; Mediavilla-Varela et al., 2017; Young et al., 2016).

[0026] Agents that reduce or block the activity of LAG-3 include, but are not limited to, BMS-986016, IMP321, and GSK2831781 (He et al., 2016).

[0027] Agents that reduce or block the activity of CTLA-4 include, but are not limited to, ipilimumab and tremelimumab.

[0028] Agents that reduce or block the activity of VISTA include, but are not limited to, small molecules, such as CA-170, and antibodies (e.g., Le Mercier et al., 2014).

[0029] Agents that reduce or block the activity of TIM-3 include, but are not limited to, antibodies such as MBG453 and TSR-022; see Dempke et al., 2017.

[0030] Agents that reduce or block the activity of KIRs include, but are not limited to, monoclonal antibodies such as IPH2101 and Lirilumab (BMS-986015, formerly IPH2102); see Benson & Caligiuri, 2014.

[0031] Agents that reduce or block the activity of IDO include, but are not limited to, epacadostat and agents disclosed in US 2017/0037125.

[0032] Agents that reduce or block the activity of BTLA include, but are not limited to, peptides (e.g., Spodzieja et al., 2017).

[0033] Agents that reduce or block the activity of A2AR include, but are not limited to, small molecules such as CPI-444 and vipadenant.

[0034] In some embodiments, the second therapy comprises a cytokine (e.g., interleukin 7).

[0035] In some embodiments, the second therapy comprises an agonist of a stimulatory checkpoint molecule. These molecules include, but are not limited to:

1. 1. CD40;
2. 2. OX40;
3. 3. glucocorticoid-induced tumor necrosis factor-related protein (GITR); and
4. 4. Inducible T-cell COStimulator (ICOS).

[0036] Agonists of CD40 include, but are not limited to, CD40 agonist monoclonal antibodies such as cp-870,893, ChiLob7/4, dacetuzumab, and lucatumumab. See, e.g., Vonderheide et al., 2007; Khubchandani et al., 2009; Johnson et al., 2010; Bensinger et al., 2012; Vonderheide and Glennie, 2013; Johnson et al., 2015.

[0037] Agonists of OX40 include, but are not limited to, OX40 agonist antibodies such as MOXR0916, MED16469, MED10562, PF-045618600, GSK3174998, and INCCAGN01949, and OX40L-Fc fusion proteins, such as MED16383. See, e.g., Huseni et al., 2014; Linch et al., 2015; Messenheimer et al., 2017. See also Shrimali et al., 2017.

[0038] Agonists of GITR include, but are not limited to, MEDI1873. See, e.g., Schaer et al., 2012; Tigue et al., 2017.

[0039] Agonists of ICOS include, but are not limited to, ICOS agonist antibodies JTX-2011 and GSK3359609. See, e.g., Harvey et al., 2015; Michaelson et al., 2016.

[0040] In other embodiments, the second therapy comprises a 4-1BB agonist (Shindo et al., 2015), such as urelumab; a 4-1BB antagonist (see US 2017/0174773); an inhibitor of anaplastic lymphoma kinase (ALK; Wang et al., 2014; US 2017/0274074), such as crizotinib, ceritinib, alectinib, PF-06463922, NVP-TAE684, AP26113, TSR-011, X-396, CEP-37440, RXDX-101; an inhibitor of histone deacetylase (HDAC; see US 2017/0327582); a VEGFR inhibitor, such as axitinib, sunitinib, sorafenib, tivozanib, bevacizumab; and/or an anti-CD27 antibody, such as varlilumab.

[0041] In some embodiments, the second therapy comprises a cancer vaccine (e.g., Duraiswamy et al., 2013). A "cancer vaccine" is an immunogenic composition intended to elicit an immune response against a particular antigen in the individual to which the cancer vaccine is administered. A cancer vaccine typically contains a tumor antigen which is able to induce or stimulate an immune response against the tumor antigen. A "tumor antigen" is an antigen that is present on the surface of a target tumor. A tumor antigen may be a molecule which is not expressed by a non-tumor cell or may be, for example, an altered version of a molecule expressed by a non-tumor cell (e.g., a protein that is misfolded, truncated, or otherwise mutated).

[0042] In some embodiments, the second therapy comprises a chimeric antigen receptor (CAR) T cell therapy. See, e.g., John et al., 2013; Chong et al., 2016.

Additional Therapeutic Uses

[0043] In some embodiments, one or more of the disclosed peptides or modified versions thereof can be administered to treat infectious diseases, including chronic infections, caused, e.g., by viruses, fungi, bacteria, and protozoa, and helminths.

[0044] Examples of viral agents include human immunodeficiency virus (HIV), Epstein Barr Virus (EBV), *Herpes simplex* (HSV, including HSV1 and HSV2), Human Papillomavirus (HPV), *Varicella zoster* (VSV) *Cytomegalovirus* (CMV), and hepatitis A, B, and C viruses.

[0045] Examples of fungal agents include *Aspergillus*, *Candida*, *Coccidioides*, *Cryptococcus*, and *Histoplasma capsulatum*.

[0046] Examples of bacterial agents include *Streptococcal* bacteria (e.g., *pyogenes*, *agalactiae*, *pneumoniae*), *Chlamydia*, *pneumoniae*, *Listeria monocytogenes*, and *Mycobacterium tuberculosis*.

[0047] Examples of protozoa include *Sarcodina* (e.g., *Entamoeba*), *Mastigophora* (e.g., *Giardia*), *Ciliophora* (e.g., *Balantidium*), and *Sporozoa* (e.g., *Plasmodium falciparum*, *Cryptosporidium*).

[0048] Examples of helminths include *Platyhelminths* (e.g., trematodes, cestodes),

Acanthocephalins, and *Nematodes*.

[0049] In some embodiments one or more of the disclosed peptides or modified versions thereof can be administered as a vaccine adjuvant in conjunction with a vaccine to enhance a response to vaccination (e.g., by increasing effector T cells and/or reducing T cell exhaustion). The vaccine can be, for example, an RNA vaccine (e.g., US 2016/0130345, US 2017/0182150), a DNA vaccine, a recombinant vector, a protein vaccine, or a peptide vaccine. Such vaccines can be delivered, for example, using virus--like particles, as is well known in the art.

[0050] In some embodiments one or more of the disclosed peptides or modified versions thereof can be administered to treat sepsis.

[0051] In some non-therapeutic methods one or more of the disclosed peptides or modified versions thereof can be administered to promote hair color re-pigmentation. In some embodiments one or more of the disclosed peptides or modified versions thereof can be administered to promote lightening of pigmented skin lesions.

Administration of Peptides

[0052] In some embodiments, one or more of the disclosed peptides themselves, or modified versions thereof, are administered. In some of these embodiments, a peptide carrier system is used. A number of peptide carrier systems are known in the art, including microparticles, polymeric nanoparticles, liposomes, solid lipid nanoparticles, hydrophilic mucoadhesive polymers, thiolated polymers, polymer matrices, nanoemulsions, and hydrogels. See Patel et al. (2014), Bruno et al. (2013), Feridooni et al. (2016). Any suitable system can be used.

[0053] In some embodiments, engineered T cell-based therapies that express and secrete a peptide or protein can be used to deliver PD-1 inhibition at the site of engagement of the T cell receptor with an antigen. The T cell-based therapy could be, for example, a CAR-T cell that expresses one or more of the disclosed peptides or modified versions thereof. Either inducible or constitutive expression can be used.

[0054] In other embodiments one or more of the disclosed peptides or modified versions thereof are delivered using one or more nucleic acids encoding the peptide(s) (e.g., DNA, cDNA, PNA, RNA or a combination thereof); See, e.g., US 2017/0165335. Nucleic acids encoding one or more peptides can be delivered using a variety of delivery systems known in the art. Nucleic acid delivery systems include, but are not limited to, gene-gun; cationic lipids and cationic polymers; encapsulation in liposomes, microparticles, or microcapsules; electroporation; virus-based, and bacterial-based delivery systems. Virus-based systems include, but are not limited to, modified viruses such as adenovirus, adeno-associated virus, herpes virus, retroviruses, vaccinia virus, or hybrid viruses containing elements of one or more viruses. US 2002/0111323 describes use of "naked DNA," i.e., a "non-infectious, non-

immunogenic, non-integrating DNA sequence," free from "transfection-facilitating proteins, viral particles, liposomal formulations, charged lipids and calcium phosphate precipitating agents," to administer a peptide. Bacterial-based delivery systems are disclosed, e.g., in Van Dessel et al. (2015) and Yang et al. (2007).

[0055] In some embodiments, a peptide is administered via an RNA molecule encoding the peptide. In some embodiments, the RNA molecule is encapsulated in a nanoparticle. In some embodiments, the nanoparticle comprises a cationic polymer (e.g., poly-L-lysine, polyamidoamine, polyethyleneimine, chitosan, poly(β --amino esters). In some embodiments, the nanoparticle comprises a cationic lipid or an ionizable lipid. In some embodiments, the RNA molecule is conjugated to a bioactive ligand (e.g., N-acetylgalactosamine (GalNAc), cholesterol, vitamin E, antibodies, cell-penetrating peptides). See, e.g., Akinc et al. (2008), Akinc et al. (2009), Anderson et al. (2003), Behr (1997), Boussif et al. (1995), Chen et al. (2012), Dalilman et al. (2014), Desigaux et al. (2007), Dong et al. (2014), Dosta et al. (2015), Fenton et al. (2016), Guo et al. (2012), Howard et al. (2006), Kaczmarek et al. (2016), Kanasty et al. (2013), Kauffman et al. (2015), Kozielski et al. (2013), Leus et al. (2014), Lorenz et al. (2004), Love et al. (2010), Lynn & Langer (2000), Moschos et al. (2007), Nair et al. (2014), Nishina et al. (2008), Pack et al. (2005), Rehnian et al. (2013), Schroeder et al. (2010), Tsutsumi et al. (2007), Tzeng et al. (2012), Won et al. (2009), Xia et al. (2009), Yu et al. (2016).

[0056] In some embodiments, an RNA molecule can be modified to reduce its chances of degradation or recognition by the immune system. The ribose sugar, the phosphate linkage, and/or individual bases can be modified. See, e.g., Behlke (2008), Bramsen (2009), Chiu (2003), Judge & MacLachlan (2008), Kauffman (2016), Li (2016), Morrissey (2005), Prakash (2005), Pratt & MacRae (2009), Sahin (2014), Soutschek (2004), Wittrup & Lieberman (2015). In some embodiments, the modification is one or more of a ribo-difluorotoluyll nucleotide, a 4'-thio modified RNA, a boranophosphate linkage, a phosphorothioate linkage, a 2'-O-methyl (2'-OMe) sugar substitution, a 2'-fluoro (2'-F), a 2'-O-methoxyethyl (2'--MOE) sugar substitution, a locked nucleic acid (LNA), and an L-RNA.

Routes of Administration, Pharmaceutical Compositions, and Devices

[0057] Pharmaceutical compositions comprising an effective amount of any of the active agents described in the paragraphs above include a pharmaceutically acceptable vehicle. The "pharmaceutically acceptable vehicle" may comprise one or more substances which do not affect the biological activity of the active agent(s) and, when administered to a patient, does not cause an adverse reaction. Pharmaceutical compositions may be liquid or may be lyophilized. Lyophilized compositions may be provided in a kit with a suitable liquid, typically water for injection (WFI) for use in reconstituting the composition. Other suitable forms of pharmaceutical compositions include suspensions, emulsions, and tablets.

[0058] Routes of administration include injection or infusion (e.g., epidural, intradermal,

intramuscular, intraperitoneal, intravenous, sub-cutaneous), transdermal (e.g., US 2017/0281672), mucosal (e.g., intranasal or oral), pulmonary, and topical (e.g., US 2017/0274010) administration. See, e.g., US 2017/0101474.

[0059] Administration can be systemic or local. In addition to local infusions and injections, implants can be used to achieve a local administration. Examples of suitable materials include, but are not limited to, sialastic membranes, polymers, fibrous matrices, and collagen matrices.

[0060] Topical administration can be by way of a cream, ointment, lotion, transdermal patch (such as a microneedle patch), or other suitable forms well known in the art.

[0061] Administration can also be by controlled release, for example, using a microneedle patch, pump and/or suitable polymeric materials. Examples of suitable materials include, but are not limited to, poly(2-hydroxy ethyl methacrylate), poly(methyl methacrylate), poly(acrylic acid), poly(ethylene-co-vinyl acetate), poly(methacrylic acid), polyglycolides (PLG), polyanhydrides, poly(N-vinyl pyrrolidone), poly(vinyl alcohol), polyacrylamide, poly(ethylene glycol), polylactides (PLA), poly(lactide-co-glycolides) (PLGA), and polyorthoesters.

[0062] Devices comprising any of the active agents described above include, but are not limited to, syringes, pumps, transdermal patches, spray devices, vaginal rings, and pessaries.

EXAMPLE 1. Peptide Library Screening

[0063] The TriCo-20[™] (TRICO-20[™]) and TriCo-16[™] (TRICO-16[™]) Phage Display Peptide Libraries (Creative Biolabs, 45-1 Ramsey Road, Shirley, NY 11967) were screened to identify binders of soluble recombinant human PD-1 receptor. After the fourth round of panning, obvious enrichment for specific binders was observed, and individual peptides were confirmed as weakly specific binders in a clonal phage ELISA. A fifth round of panning led to greater enrichment. Table 1 lists four peptides which showed strong specific binding in the clonal phage ELISA.

Table 1

Clone	Clonal Phase ELISA		peptide sequence	SEQ ID NO:
	coated signal	uncoated signal		
QP20	0.851	0.446	QTRTVPMPKIHPPWQNVVP	1
HD20	0.281	0.109	HHHQVYQVRSHWTGMHSGHD	2
WQ20	0.275	0.115	WNLPASFHNHHIRPHEHEWIQ	3
SQ20	0.284	0.159	SSYHHFKMPELHFGKNTFHQ	4

EXAMPLE 2. Competitive PD-1:PD-L1 Binding Inhibition Assay

[0064] Briefly, detection of cell surface PD-1 on Jurkat cells was accomplished by incubating cells with the human PD-L1-Fc fusion protein, followed by detection of the recombinant molecule with a fluorescently labeled anti-human Fc antibody. Flow cytometry was performed to detect binding between PD-1 and the PD-L1 recombinant protein. Quantitative binding measurement was then determined by mean fluorescence intensity (MFI).

[0065] Jurkat Cell-surface expression of PD1 and binding of PD-L1 to these cells were verified as shown in Figures 1 and 2. The results are shown in Figures 3A-B, 4A-B, 5A-B, and 6A-B.

EXAMPLE 3. Cell-Based Reporter Assay

[0066] A cell-based reporter assay was used to assess whether binding of the four peptides identified above was sufficient to block the interaction with PD-1 and its ligand PD-L1. The components of the assay include a Jurkat T cell line that stably expresses human PD-1 and a luciferase reporter, a CHO cell line that stably expressed human PD-L1, and a positive control anti-PD-1 antibody that blocks the interaction of PD-1 and PD-L1, resulting in a measurable effect in the assay. The luciferase reporter in the Jurkat T cell line is triggered by IL-1, NFAT, or NF- κ B response elements in the promoter region. The Jurkat T cells are pre-treated with CD3 and immediately cryopreserved for use in the assay. Interaction of the Jurkat T cells with the PD-L1 expressing cell line inhibits the intracellular mechanism by which the luciferase construct is activated, thereby preventing luciferase expression. A molecule that binds to either PD-1 on the Jurkat T cells or to PD-L1 on the CHO cells sufficiently to prevent their interaction permits the Jurkat T cells to produce luciferase. CellTiter-Glo[®] (CELLTITER-GLO[®], Promega) was used to measure luciferase expression.

[0067] The results of positive control assays using the anti-PD-1 control antibody are shown in Figures 7A-B. These results demonstrate that the control antibody restores luciferase expression in a dose-dependent manner, with peak-fold inhibition of approximately 8 at an antibody concentration of 20 μ M.

[0068] The results of assays of the peptides identified above are shown in Figures 8A-B. These results demonstrate that each of the four peptides restores luciferase expression in a dose-dependent manner, with peak-fold inhibition of approximately 1.5 at a concentration of approximately 25 μ M.

EXAMPLE 4. Tetanus Toxoid Recall Assay Using Individual Peptides

[0069] Peptides 1-4 were tested in a human PBMC-based tetanus antigen recall assay. "Peptide CQ-22" was used as a negative control.

[0070] PBMCs were obtained from plasma of human donors and tested *in vitro* for recall of tetanus toxoid. Suitable PBMCs were cryopreserved until needed, then thawed and cultured in a 96-wellplate. Tetanus toxoid was added to the cultures in the presence or absence of peptides 1 - 4, and the production of cytokines and cell surface T cell activation markers were examined.

[0071] The results of these assays are shown in Figures 9-15 and summarized qualitatively in Table 2. In the table, "x" indicates no effect, "-" indicates a possible low effect, "+" indicates some effect, and "++" indicates a definite effect.

Table 2.

peptide	IL-2	IL-4	IL-6	IL-10	IL-17a	IFN γ	TNF α
QP20	x	-	x	x	x	x	x
HD20	-	x	++	x	++	++	++
WQ20	-	++	++	x	++	++	++
SQ20	+	-	++	+	++	++	+

[0072] The results demonstrated a trend towards modest enhancement of IL-6, IL-17 α , IFN γ , and TNF α production at the highest concentrations of peptides. No significant enhancement of IL-2 production was detected.

EXAMPLE 5. Tetanus Toxoid Recall Assay Using Combinations of Peptides

[0073] Combinations of peptides were tested in the antigen recall assay described above, using a different PBMC donor and a different lot number of tetanus toxoid. The results are shown in Figures 16, 17, 18, 19, 20, 21, and 22. These results demonstrated that the combination of the four peptides combination of the four peptides QP20, HD20, WQ20, and SQ20 result in increased IL-2 production and reduced IL-17a production.

[0074] The effect of peptides QP20, HD20, WQ20, and QP20 on the production of IL-2 and IL-17a appears to be donor-specific, as shown in Figures 23A-B and 24A-B.

EXAMPLE 6. BIACORE® Assays

[0075] BIACORE® assays were carried out using a BIACORE® T-200 at 25°C. The assay and regeneration buffers contained 10 mM HEPES (pH 7.4), 150 mM NaCl, 3mM EDTA, and 0.05% P20. The immobilization buffer was 10mM sodium acetate, pH 5.0. The flow rate used for immobilizing the ligand was 5 μ l/min. The flow rate for kinetics analysis was 30 μ l/min.

[0076] Scouting. 12,000 response units (RU) of human and 6000 RU of mouse PD-1 receptors were directly immobilized on flow cell 2 and flow cell 4 of the CM5 chip by amine coupling method (EDC/NHS). The un-occupied sites were blocked with 1M ethanol amine. Scouting was performed at a single analyte concentration of 25 μ M to confirm yes/no binding. Flow cell 1 was kept blank and used for reference subtraction. Binding of analyte to the ligand was monitored in real time.

[0077] Full Kinetics. Based on the scouting results, full kinetics were performed by immobilizing higher RU of the ligand to a new chip and analyte concentration at 25 μ M, followed by serial dilution to 12.5, 6.25, 3.125, 1.562, 0.78 and 0 μ M concentration or as indicated. Due to fast on rate and off rate, KD was determined by steady state equilibrium kinetics.

[0078] Chi square (χ^2) analysis was carried out between the actual sensorgram and a sensorgram generated from the BIANALYSIS[®] software (black line) to determine the accuracy of the analysis. A χ^2 value within 1- 2 is considered significant (accurate) and below 1 is highly significant (highly accurate). The results are summarized in Table 3.

Table 3

Ligand 10,000 RU	Analyte	Rmax (RU)	KA(1/M)	KD (M)	Conc. (μ M)	χ^2
mouse PD-1	WQ-21	270	1.31×10^3	7.61×10^{-4}	0-25	0.0203
mouse PD-1	QP-20	13.4	1.80×10^4	5.54×10^{-5}	0-25	0.0446
mouse PD-1	HD-20	76	4.25×10^3	2.35×10^{-4}	0-25	0.11
mouse PD-1	SQ-20	12.8	2.14×10^4	4.68×10^{-5}	0-25	0.039
human PD-1	WQ-21	84.7	3.28×10^3	3.05×10^{-4}	0-25	0.0309
human PD-1	QP-20	3.83	9.36×10^4	1.07×10^{-5}	0-25	0.0569
human PD-1	HD-20	3.35	3.18×10^5	3.41×10^{-6}	0-12.5	0.0733
human PD-1	SQ-20	4.05	1.94×10^5	5.16×10^{-6}	0-25	0.111
mouse PD-1	Mouse PD-L1	259	2.75×10^6	3.64×10^{-7}	0-50	0.105
human PD-1	Human PD-L1	213	6.92×10^6	1.44×10^{-7}	0-50	2.44

[0079] These results indicate that each of the four peptides bind both human and mouse PD-1. QP20 and SQ20 showed the highest affinity towards mouse PD-1. HD20 and SQ20 showed the highest affinity towards human PD-1.

EXAMPLE 7. Experimental Metastasis Model

[0080] Efficacy of the peptides was evaluated in a B16-F10-LacZ experimental metastasis model. In this model, B 16-F10-LacZ cells, transfected to express the LacZ gene that encodes β -galactoside, an intracellular enzyme, are injected into the tail vein of syngeneic mice. The cells travel through the circulation, settle in the lungs, and form tumors. Mice are terminated 2 weeks after implant. When the enzyme cleaves its substrate, X-gal, the products dimerize and change color and can be detected *ex vivo*. The number of metastatic tumors on the surface of the lung is then quantified by manual counting of tumors under a dissecting microscope.

[0081] Briefly, mice (N=7) were implanted on study day 0 with B16-F10-LacZ tumor cells (5×10^5 or 1×10^6 cells per mouse) by intravenous injection in the tail vein. Mice received a treatment of the peptide combination (200 μ g, 20 μ g, or 2 μ g, each peptide per dose) intravenously by tail vein injection on study days 2, 5, 7, 9 and 12. Detailed clinical examinations and body weights were recorded regularly during treatment. Mice were terminated on study day 14, and their lungs were removed and stained. The number of tumor metastases were counted. Treatment groups are described in Table 4.

Table 4

Group	N	Implant	Treatment	Dose	Route	Treatment Days
1	7	5×10^5	QP-20, SQ-20, HD-20, WQ-20	200 μ g	IV	SD 2, 5, 7, 9, 12
2	7	5×10^5	QP-20, SQ-20, HD-20, WQ-20	20 μ g	IV	SD 2, 5, 7, 9, 12
3	7	5×10^5	QP-20, SQ-20, HD-20, WQ-20	2 μ g	IV	SD 2, 5, 7, 9, 12
4	7	5×10^5	Untreated	-	-	-
5	7	1×10^6	QP-20, SQ-20, HD-20, WQ-20	200 μ g	IV	SD 2, 5, 7, 9, 12
6	7	1×10^6	Untreated	-	-	-

The results are shown in Figure 25. A good dose response was observed when mice were implanted at both cell concentrations. Mice treated with the highest dose of peptide mixture (200 μ g) had the fewest tumors (average 97), and mice treated with the lowest dose of peptide mixture (2 μ g) had the most tumors (average 205). Similarly, in the two groups that were implanted with high tumor numbers, the untreated group had significantly more tumors. This indicates that the 4 peptides in combination showed a dose-dependent efficacy on B16-F10-LacZ tumor growth *in vivo*. Moreover, the peptide combination was well tolerated by the mice and did not have any acute adverse effects on animal health.

EXAMPLE 8. Effect of Peptide Combination on the Immunogenicity of a Malaria Vaccine

[0082] Immunogenicity of the peptide combination as a prophylactic vaccine adjuvant was

assessed in a mouse model of malaria. Balb/c mice immunized with an adenovirus-based malaria vaccine expressing the *Plasmodium yoelli* circumsporozoite protein (AdPyCS) were given 200µg of the peptide combination, anti-PD-1 mAb, anti-PDL1 mAb, or the negative control peptide ovalbumin (OVA) on days 1, 3, 5, and 7 after immunization with AdPyCS (Table 5). Note that no additional adjuvant was added to the AdPyCS antigen. Spleens were collected 12 days after immunization, and the number of splenic PyCS-specific, IFN γ -secreting CD8⁺ T cells was determined via ELISpot assay. Note that for the ELISpot assay, splenocytes were stimulated with the SYVPSAEQI peptide (SEQ ID NO:5), an H-2Kd-restricted CD8⁺ T cell epitope of PyCS.

Table 5

Cohort	Test Sample	# Mice	Route	Treatment days
1	AdPyCS only	5	-	-
2	AdPyCS + control OVA peptide (200 µg)	5	i.p.	0, 1, 3, 5, 7
3	AdPyCS + peptide combo (200 µg)	5	i.p.	0, 1, 3, 5, 7
4	AdPyCS + anti-PD-1 antibody (200 µg)	5	i.p.	0, 1, 3, 5, 7
5	AdPyCS + anti-PDL1 antibody (200 µg)	5	i.p.	0, 1, 3, 5, 7

[0083] Figure 26 shows the average number + standard deviation of CSP-specific, IFN γ -secreting CD8⁺ T cells per 0.5×10^6 splenocytes for each cohort. Significant differences in the average number $\pm$ standard deviation of CSP-specific, IFN γ -secreting CD8⁺ T cells per 0.5×10^6 splenocytes between the AdPyCS alone (Cohort 1) and the peptide combination (Cohort 3), anti-PD-1 antibody (Cohort 4) or anti-PD-L1 antibody (Cohort 5) were detected using the one-way ANOVA test (** $p < 0.001$, and * $p < 0.05$). These results demonstrate that the peptide combination (Cohort 3) is functionally active *in vivo*, increasing the number of CSP-specific, IFN γ -secreting CD8⁺ T cells ~1.6-fold relative to AdPyCS alone (Cohort 1), which was similar to changes with anti-PD-1 or -PD-L1 antibody (Cohort 4 and 5).

EXAMPLE 9. Effect of Peptide Combination on Survival in a Model of Sepsis

[0084] Sepsis can negatively alter T cell function and survival, however this can be reversed when the PD-1:PDL1 interaction is blocked, which results in improved survival. Thus the efficacy of the peptide combination was assessed in a representative, clinically relevant model of sepsis where CD1 mice are subjected to cecal ligation and puncture (CLP) to induce intra-abdominal peritonitis. For this study, 200µg of either the peptide combination or anti-PD-1 antibody were administered i.v. at 2, 24, 48, 72 and 96 hours after surgery. A vehicle control

group was also included. Six mice were in each group. All mice were checked twice daily for signs of morbidity and mortality. Administration of the peptide combination conferred an enhanced survival advantage over the vehicle control group where the peptide combination showed a 2-fold higher survival rate (Table 6). Moreover, survival in the peptide combination group was slightly above treatment with anti-PD-1 antibody.

Table 6

Group	% Survival
Vehicle Control	50%
Anti-PD-1 antibody	83%
PD-1 Peptide Combo	100%

EXAMPLE 10. Effect of Peptide Combination on Serum HBsAg Levels in HBV-Infected Mice

[0085] The combination of QP20, HD20, WQ20, and SQ20 peptides was assessed in a hepatitis B virus (HBV) mouse model where the role of PD-1 in T cell exhaustion and immunotolerance is documented (Tzeng et al., 2012; Ye et al., 2015). PD-1 is elevated in the hepatic T cells of mice with persistent HBV infection but not in animals that have cleared the infection. In this model, it has been shown that inhibition of the PD-1/PD-L1 interaction with an anti-PD-1 mAb both increases antigen-specific IFN γ production by hepatic T cells and reverses HBV persistence (Tzeng et al., 2012). This mouse model of persistent HBV presented an opportunity to test whether the combination of QP20, HD20, WQ20, and SQ20 peptides can reverse T cell exhaustion *in vivo* and aid the immune system in controlling viral infection.

[0086] Mice infected with HBV were treated with saline (negative control), 200 μ g of QP20, HD20, WQ20, and SQ20 peptides combined, or 200 μ g anti-PD-1 mAb at 9 time points, 2 days prior to infection and days 1, 3, 6, 9, 12, 14, 17 and 20 post infection. The level of serum HB surface antigen (HBsAg) was monitored by ELISA on days 7, 14, and 21 to follow the infection (higher levels of serum HBsAg are reflective of higher viral titer) and detect the immune enhancement activity of the combination of QP20, HD20, WQ20, and SQ20 peptides. The group treated with the combination of QP20, HD20, WQ20, and SQ20 peptides showed significantly lower mean level of serum HBsAg at weeks 2 and 3 post infection ($p < 0.05$, 1-way ANOVA, Tukey's Multiple Comparison Test) compared to the saline negative control (Figure 27).

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P a t e n t k r a v

1. Sammensætning til terapeutisk anvendelse til inhibering af progressionen af en hyperproliferativ lidelse, behandling af en infektionssygdom, forstærkning af et respons på vaccination eller behandling af sepsis, hvor sammensætningen omfatter en farmaceutisk acceptabel vehikel og:

(a) en nukleinsyre, der koder for mindst ét peptid;

(b) et peptidbærersystem, der omfatter det mindst ene peptid, hvor peptidbærersystemet er valgt fra gruppen bestående af en mikropartikel, en polymernanopartikel, et liposom, en fast lipidnanopartikel, en hydrofil mucoadhæsiv polymer, en thioleret polymer, en polymermatrix, en nanoemulsion og en hydrogel; eller

(c) en CAR-T-celle, der udtrykker det mindst ene peptid;

hvor det mindst ene peptid er valgt fra gruppen bestående af (i) et peptid omfattende aminosyresekvensen SEQ ID NO: 1; (ii) et peptid omfattende aminosyresekvensen SEQ ID NO:2; (iii) et peptid omfattende aminosyresekvensen SEQ ID NO:3; og (iv) et peptid omfattende aminosyresekvensen SEQ ID NO:4.

2. Ikke-terapeutisk fremgangsmåde til at fremme genpigmentering eller fremme oplysning af en pigmenteret hudlæsion, hvor fremgangsmåden omfatter indgivelse til et individ med behov derfor af en effektiv mængde af en sammensætning, der omfatter:

(a) en nukleinsyre, der koder for mindst ét peptid;

(b) et peptidbærersystem, der omfatter det mindst ene peptid, hvor peptidbærersystemet er valgt fra gruppen bestående af en mikropartikel, en polymernanopartikel, et liposom, en fast lipidnanopartikel, en hydrofil mucoadhæsiv polymer, en thioleret polymer, en polymermatrix, en nanoemulsion og en hydrogel; eller

(c) en CAR-T-celle, der udtrykker det mindst ene peptid;

hvor det mindst ene peptid er valgt fra gruppen bestående af (i) et peptid omfattende aminosyresekvensen SEQ ID NO: 1; (ii) et peptid omfattende aminosyresekvensen SEQ ID NO:2; (iii) et peptid omfattende aminosyresekvensen SEQ ID NO:3; og (iv) et peptid omfattende aminosyresekvensen SEQ ID NO:4.

5

3. Sammensætning til anvendelse ifølge krav 1 eller fremgangsmåde ifølge krav 2, hvor sammensætningen omfatter nukleinsyren, der koder for det mindst ene peptid, hvor nukleinsyren er valgt fra gruppen bestående af DNA, cDNA, PNA og RNA

10

4. Sammensætning til anvendelse ifølge krav 1 eller fremgangsmåde ifølge krav 2, hvor sammensætningen omfatter peptidbærersystemet, der omfatter det mindst ene peptid.

15

5. Sammensætning til anvendelse ifølge krav 1, hvor det mindst ene peptid indgives for at inhibere progression af den hyperproliferative lidelse, hvor den hyperproliferative lidelse fortrinsvis er en cancer, og hvor canceren fortrinsvis er et melanom.

20

6. Sammensætning til anvendelse ifølge krav 5, hvor når den hyperproliferative lidelse er en cancer, er sammensætningen beregnet til indgivelse med en anden terapi til patienten.

25

7. Sammensætning til anvendelse ifølge krav 6, hvor den anden terapi omfatter:

en cancervaccine;

en kimær antigenreceptor (CAR)-T-celleterapi;

reducerende eller blokerende aktivitet af et molekyle valgt fra gruppen bestående af PD-1, PD-L1, lymfocytaktiveringsgen-3 (LAG-3), cytotoksisk T-lymfocyt-associeret antigen 4 (CTLA-4), V-domæne immunoglobulinunder-

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trykker af T-celleaktivering (VISTA), T-celle-immunoglobulin-domæne og Mucin-domæne 3 (TIM-3), en dræber-immunoglobulinlignende receptor (KIR), indolamin-(2,3)-dioxygenase (IDO), B- og T-Lymfocytattenuator (BTLA), A2A-adenosinreceptor (A2AR);

5 et cytokin;

en agonist af et molekyle valgt fra gruppen bestående af CD40, OX40, glukokortikoid-induceret tumornekrosefaktor-relateret protein (GITR) og inducerbar T-celle Costimulator (ICOS); eller

10 et terapeutisk middel valgt fra gruppen bestående af en 4-1BB-agonist, en 4-1BB-antagonist, en inhibitor af anaplastisk lymfomkinase (ALK), en inhibitor af histon-deacetylase (HDAC) og en inhibitor af VEGFR.

8. Sammensætning til anvendelse ifølge krav 1, hvor det mindst ene peptid indgives til behandling af en infektionssygdom.

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9. Sammensætning til anvendelse ifølge krav 8, hvor den smitsomme sygdom er malaria eller hepatitis B.

10. Sammensætning til anvendelse ifølge krav 8, hvor det mindst ene peptid indgives som en vaccineadjuvans til en vaccine mod den infektionssygdom.

20

11. Sammensætning til anvendelse ifølge krav 1, hvor det mindst ene peptid indgives til behandling af sepsis.

12. Ekspressionskonstruktion, der koder for op til fire peptider valgt fra gruppen bestående af (i) et peptid bestående af aminosyresekvensen SEQ ID NO:1; (ii) et peptid bestående af aminosyresekvensen SEQ ID NO:2; (iii) et peptid bestående af aminosyresekvensen SEQ ID NO:3; og (iv) et peptid bestående af aminosyresekvensen SEQ ID NO:4.

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13. RNA-molekyle, der koder for op til fire peptider valgt fra gruppen bestående af (i) et peptid bestående af aminosyresekvensen SEQ ID NO:1; (ii) et peptid bestående af aminosyresekvensen SEQ ID NO:2; (iii) et peptid bestående af aminosyresekvensen SEQ ID NO:3; og (iv) et peptid bestående af aminosyresekvensen SEQ ID NO:4, hvor RNA-molekylet omfatter mindst én modifikation valgt fra gruppen bestående af (i) modifikation af et ribosesukker, (ii) modifikation af en phosphatbinding og (iii) modifikation af en base.

14. RNA-molekyle ifølge krav 13, hvor den mindst ene modifikation er valgt fra gruppen bestående af et ribo-difluortoluylnukleotid, et 4'-thio-modificeret RNA, en boranophosphat-binding, en phosphorthioat-binding, en 2'-O-methyl-(2'-OMe)-sukkersubstitution, en 2'-fluor-(2'-F), en 2'-O-methoxyethyl-(2'-MOE)-sukkersubstitution, en låst nukleinsyre (LNA) og en L-RNA.

15. Farmaceutisk sammensætning, der omfatter:

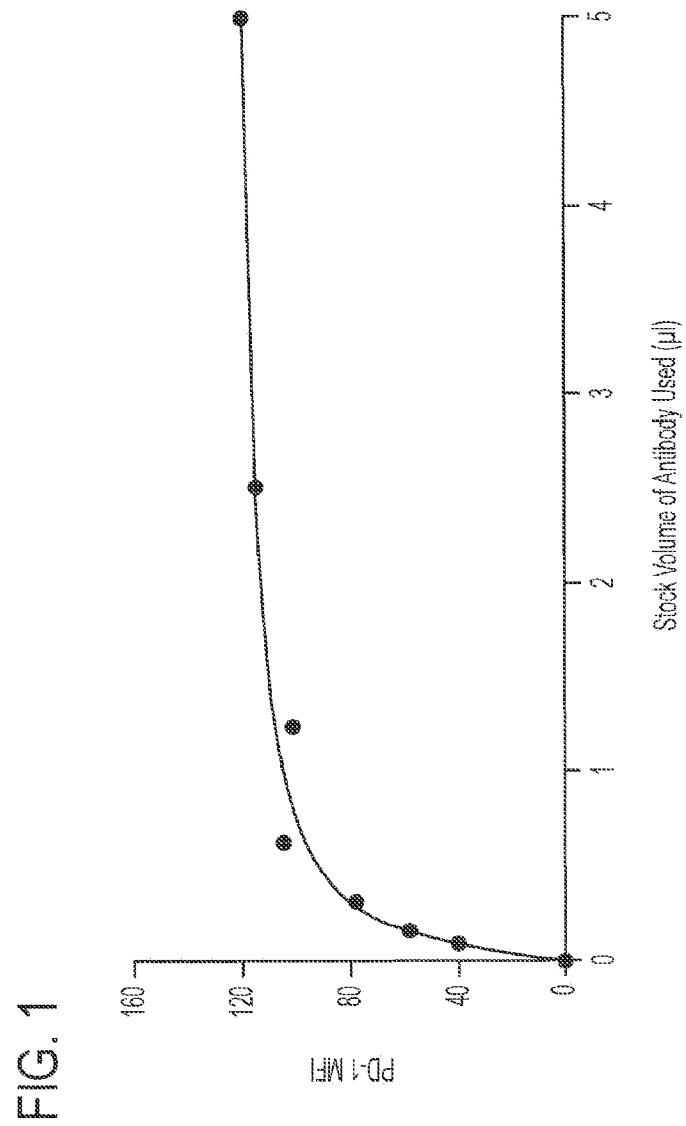
(a) et nukleinsyremolekyle, der koder for op til fire peptider valgt fra gruppen bestående af (i) et peptid bestående af aminosyresekvensen SEQ ID NO: 1; (ii) et peptid bestående af aminosyresekvensen SEQ ID NO:2; (iii) et peptid bestående af aminosyresekvensen SEQ ID NO:3; og (iv) et peptid bestående af aminosyresekvensen SEQ ID NO:4; og

(b) en farmaceutisk acceptabel bærer,

hvor nukleinsyremolekylet fortrinsvis er et RNA-molekyle.

DRAWINGS

Drawing



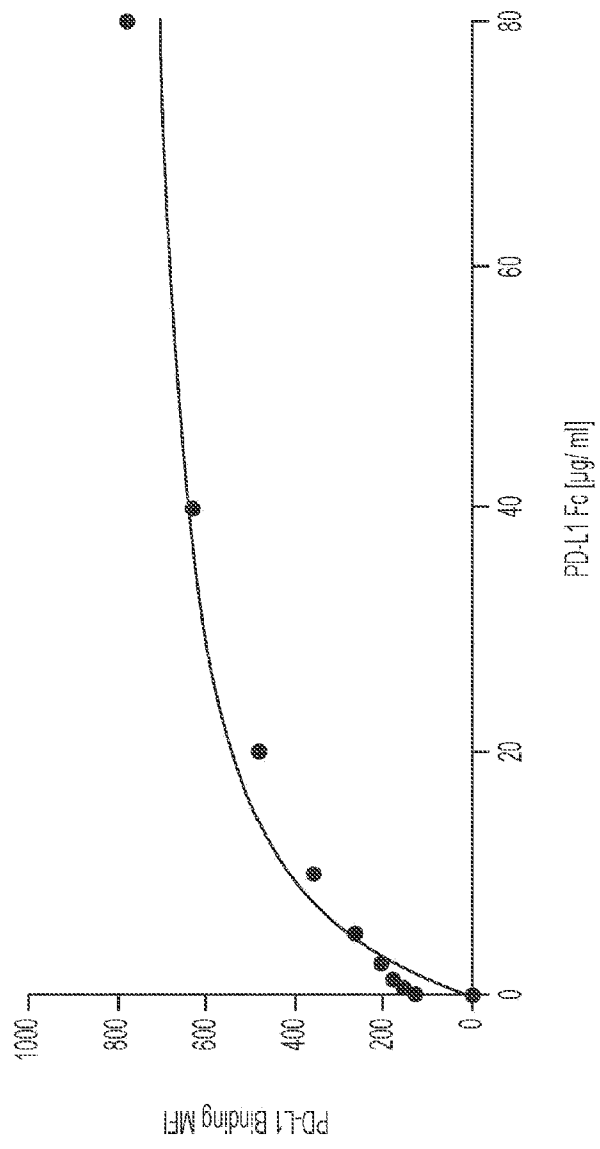


FIG. 2

FIG. 3A

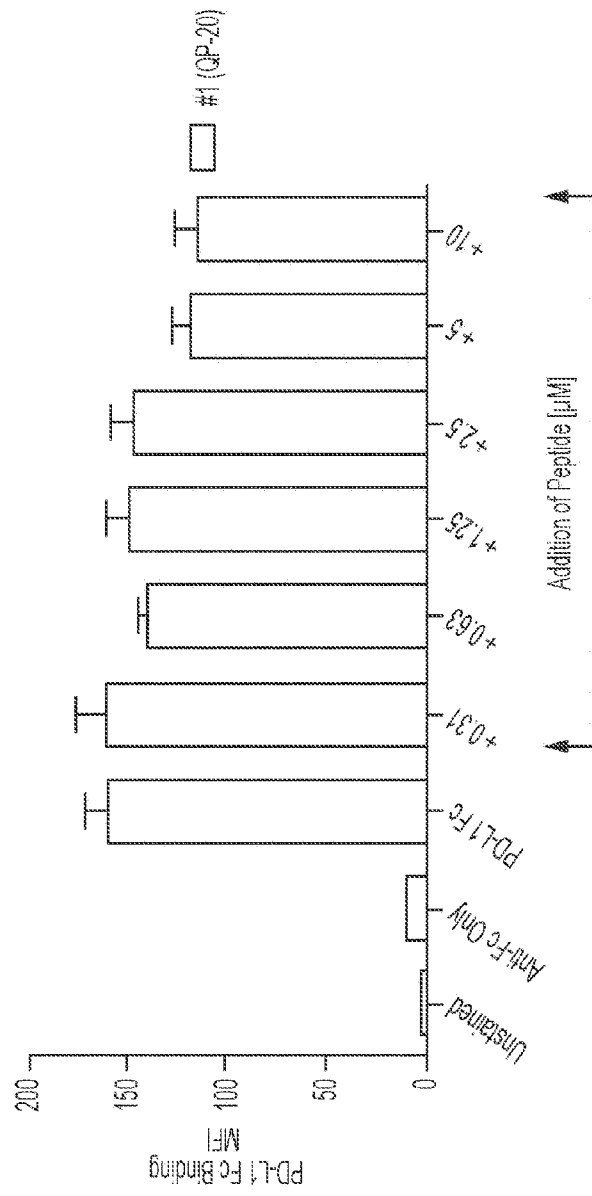


FIG. 3B

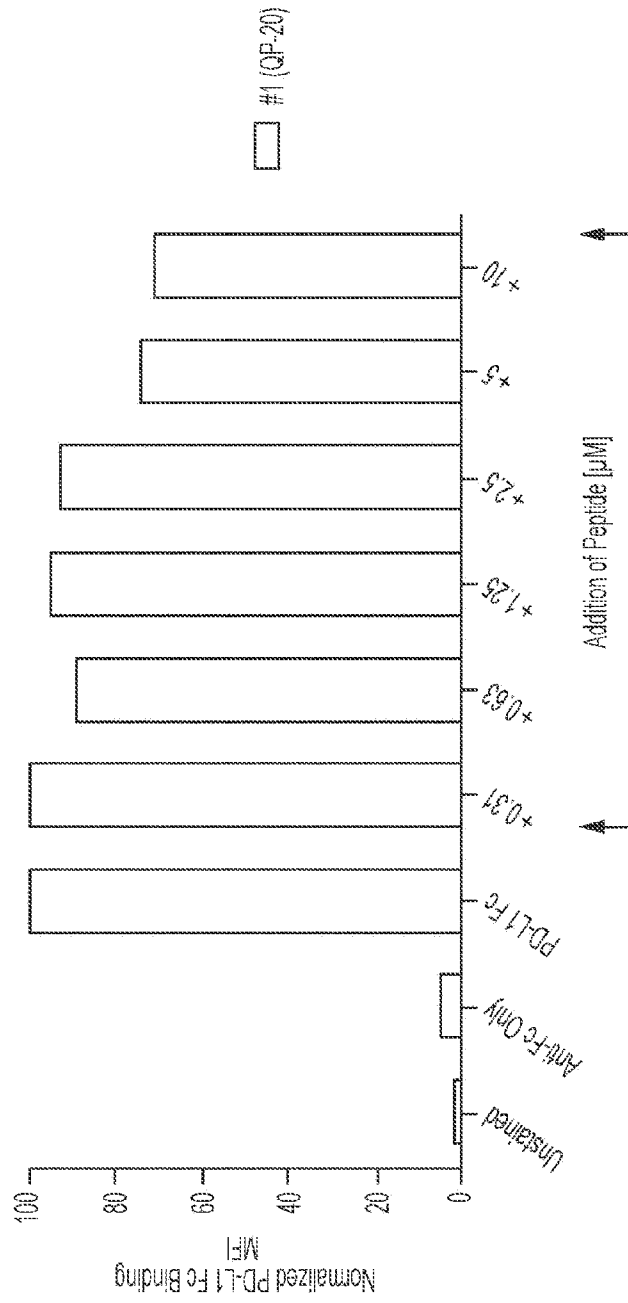


FIG. 4A

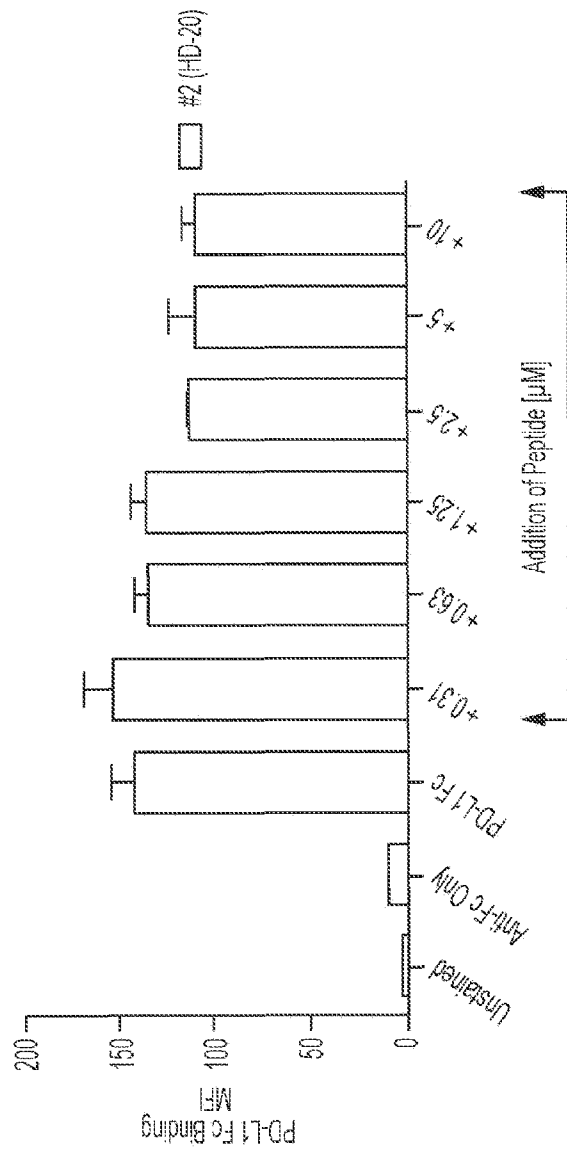


FIG. 4B

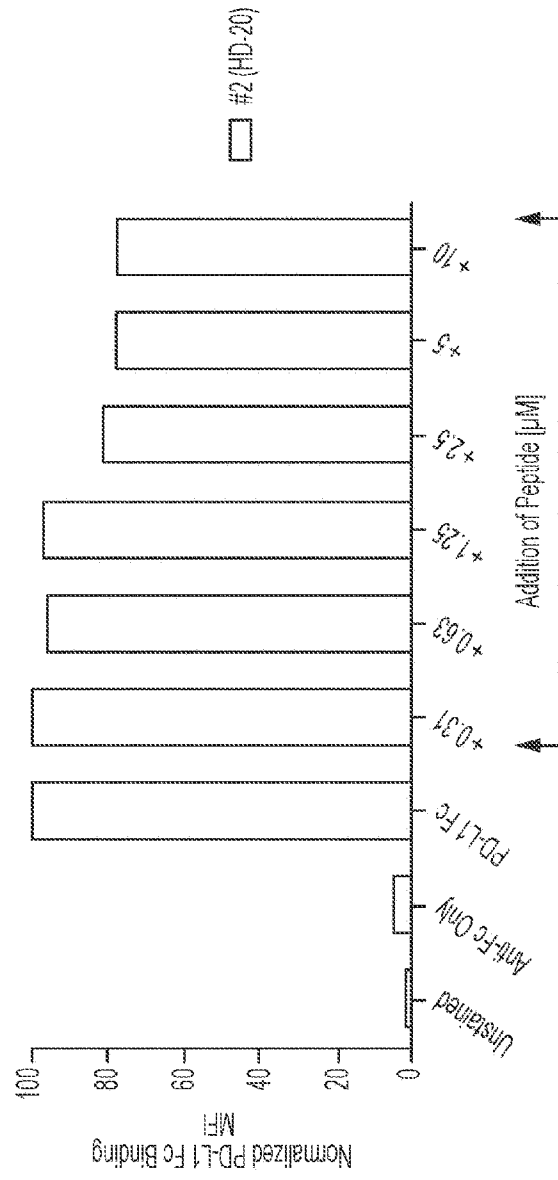


FIG. 5A

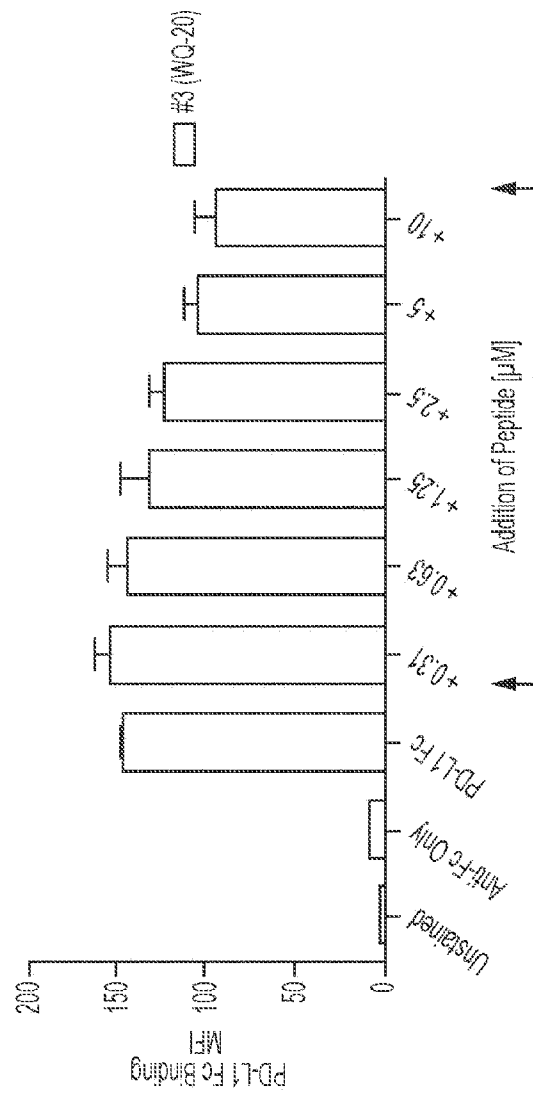


FIG. 5B

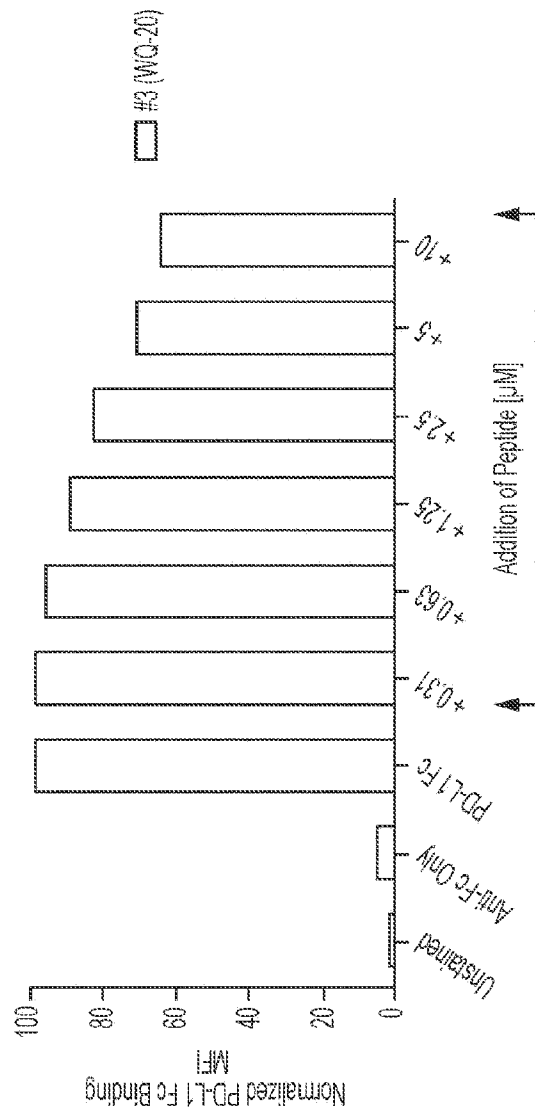


FIG. 6A

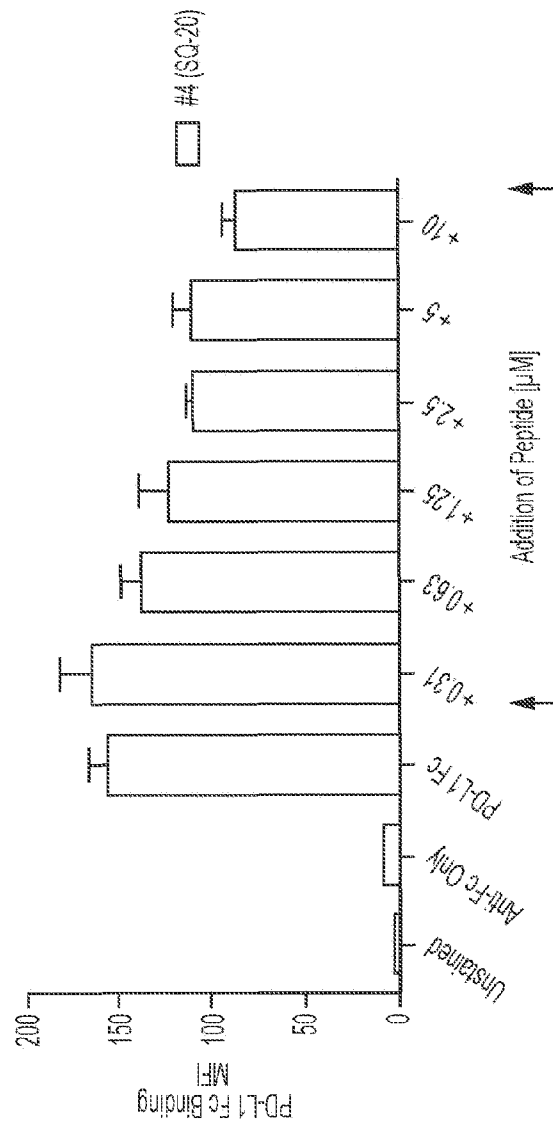


FIG. 6B

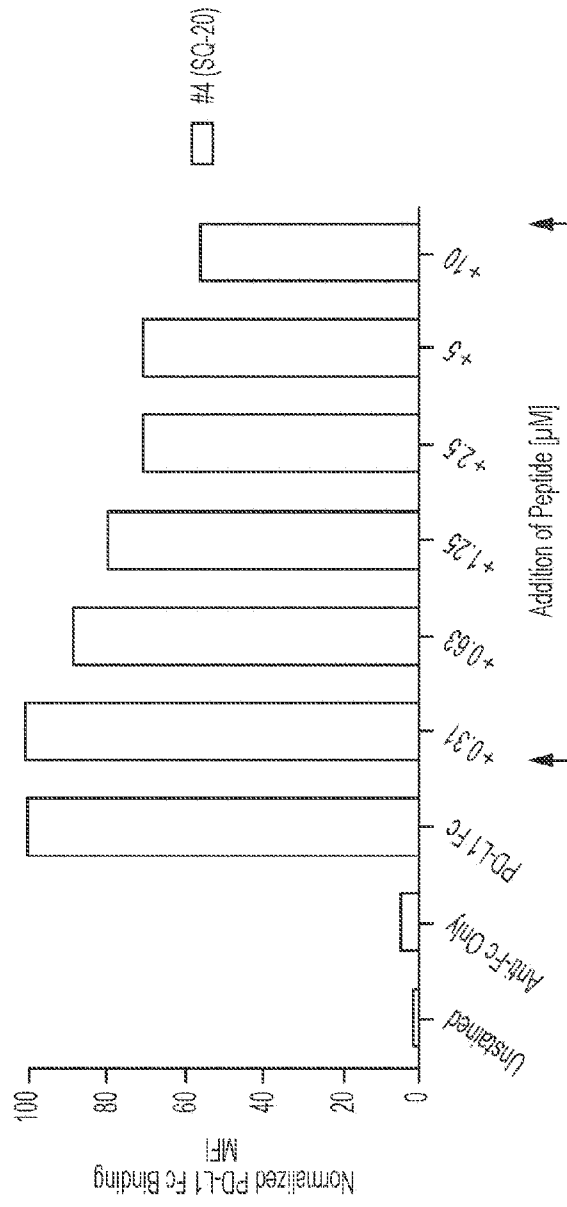


FIG. 7A

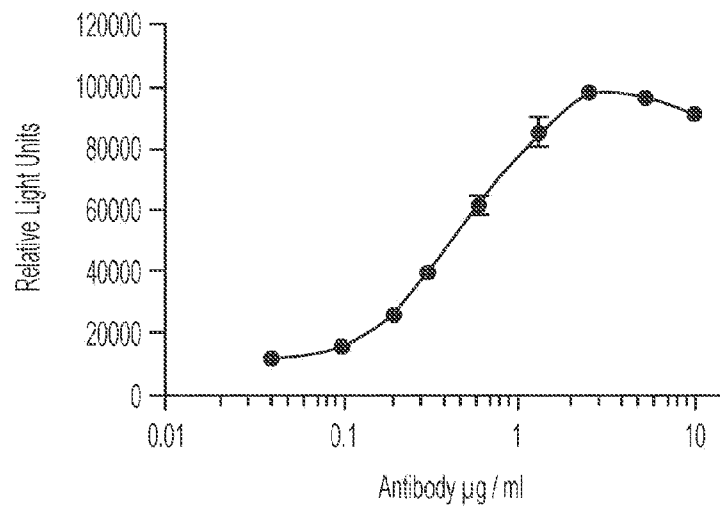


FIG. 7B

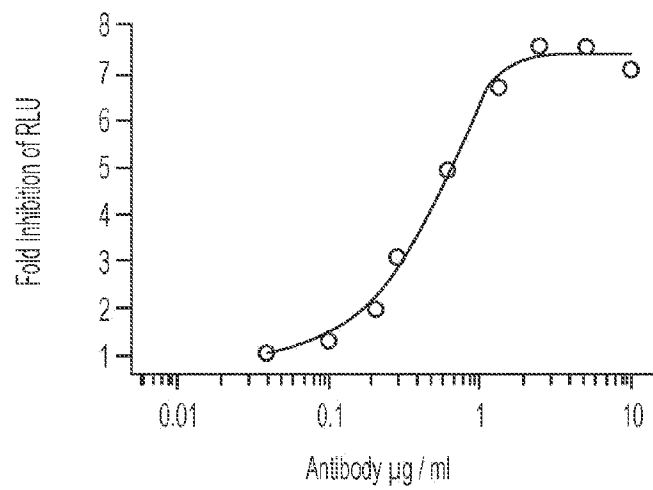


FIG. 8A

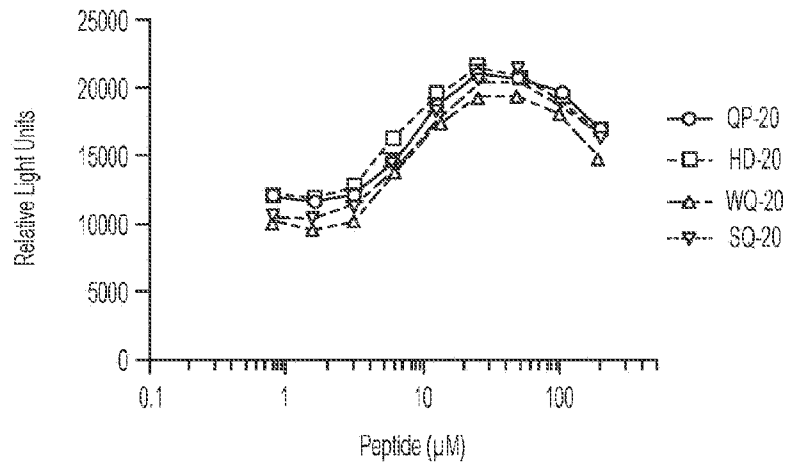
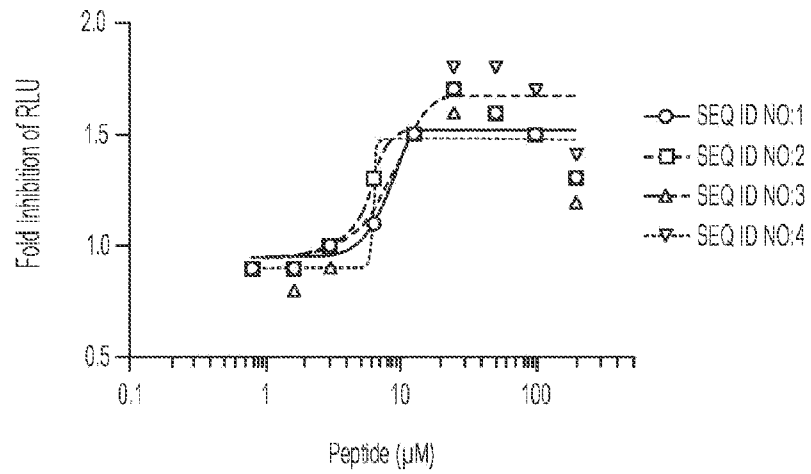
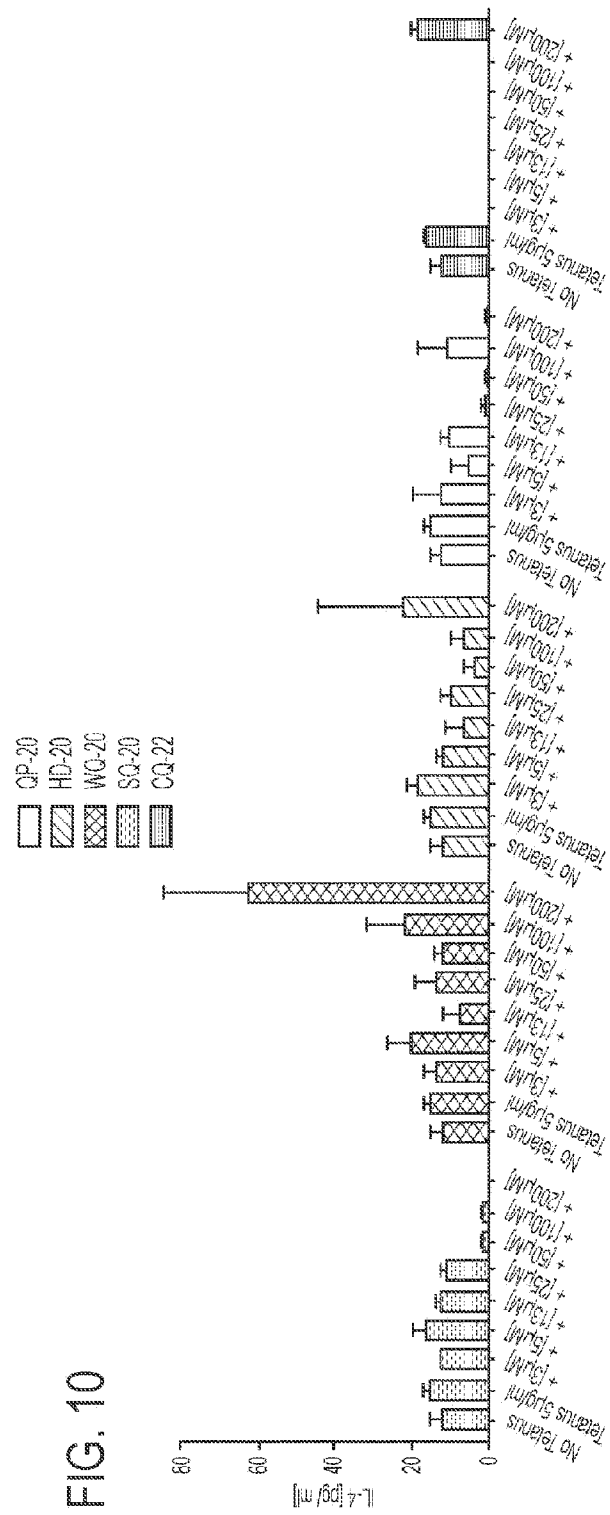


FIG. 8B







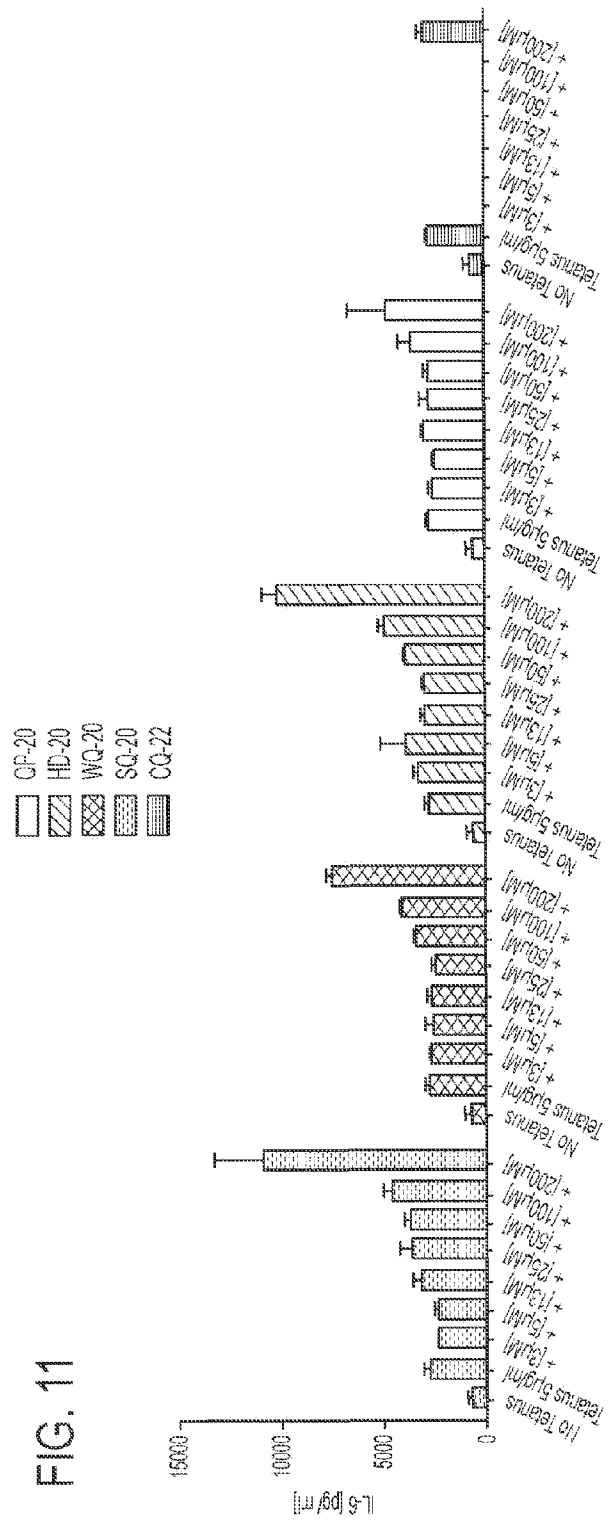
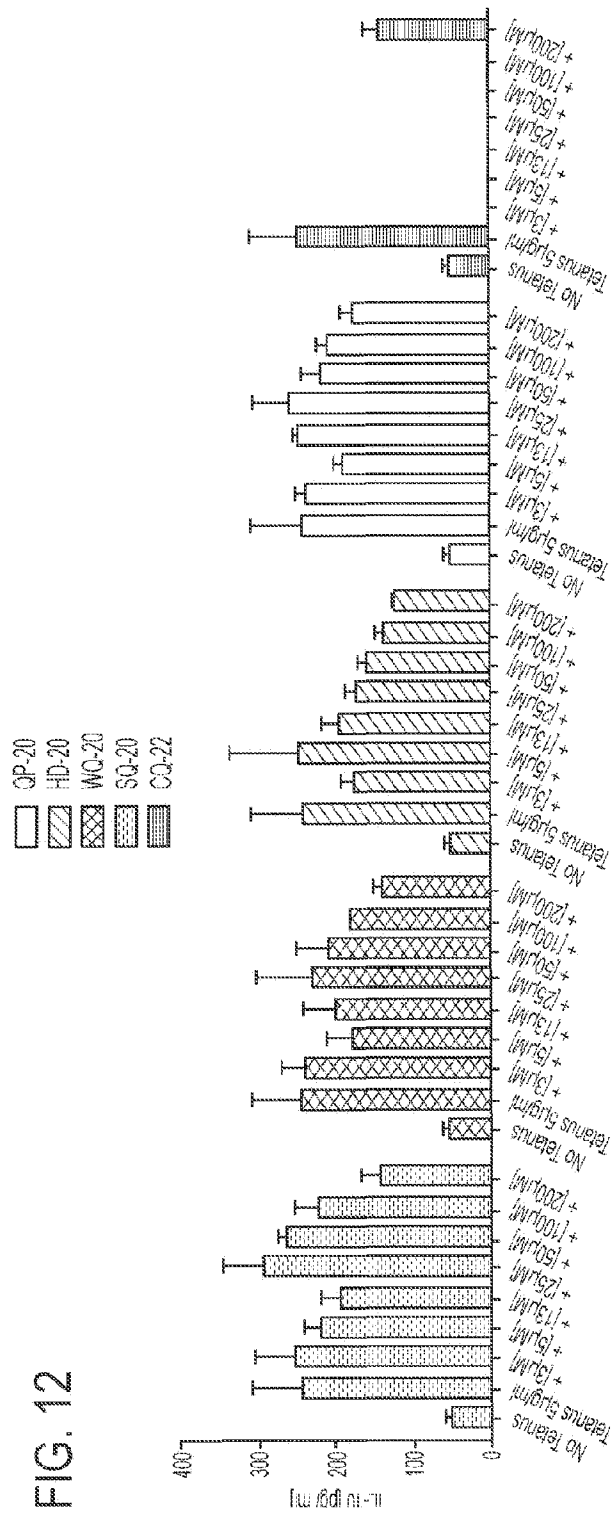
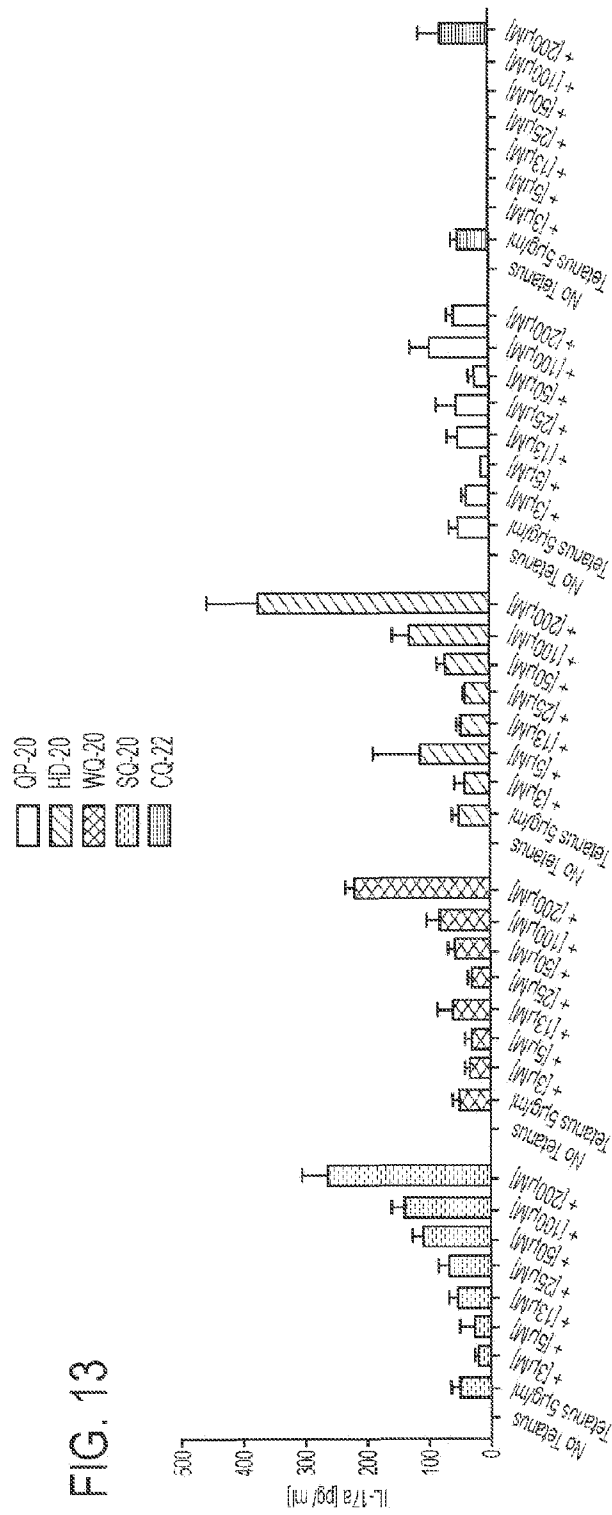


FIG. 12





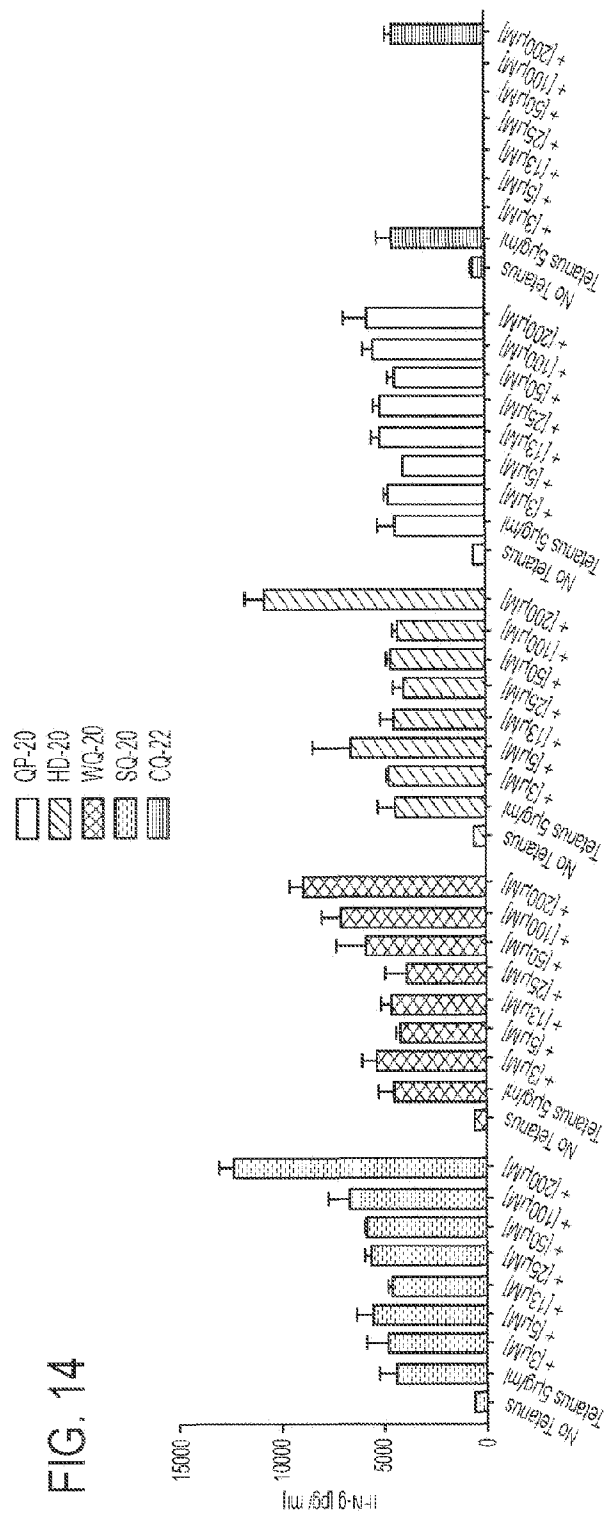


FIG. 15

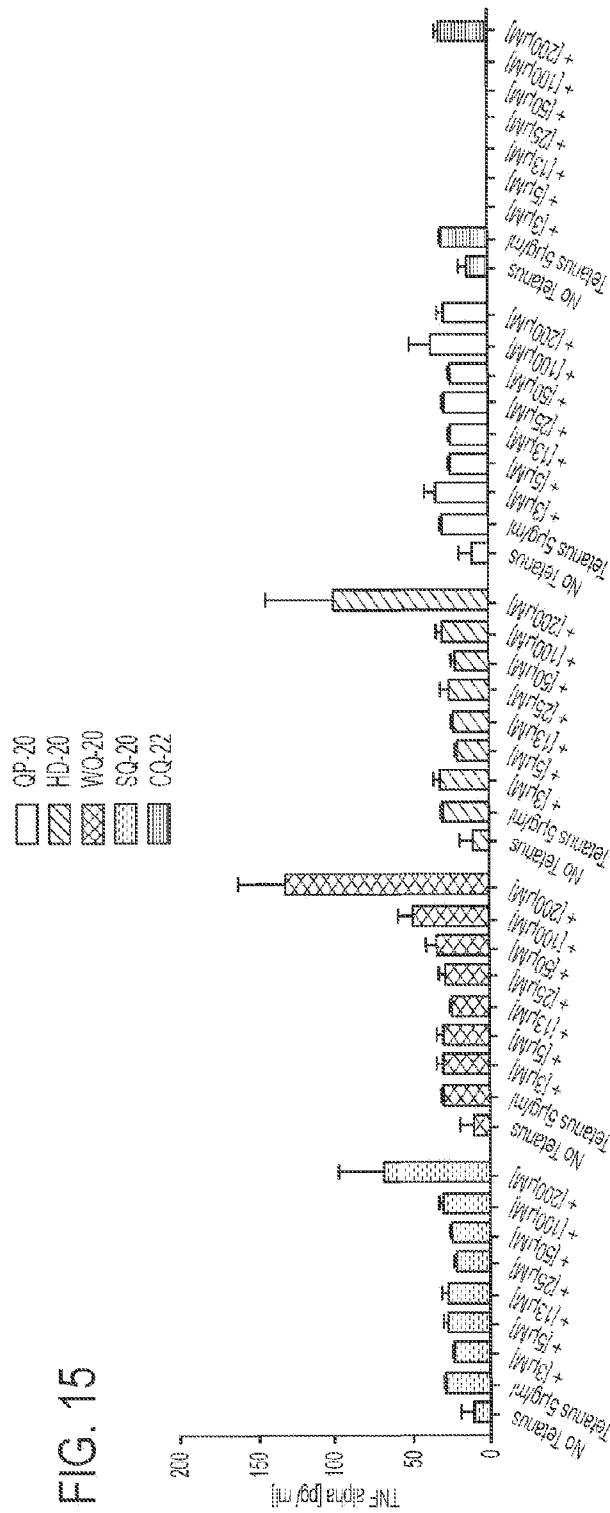


FIG. 16

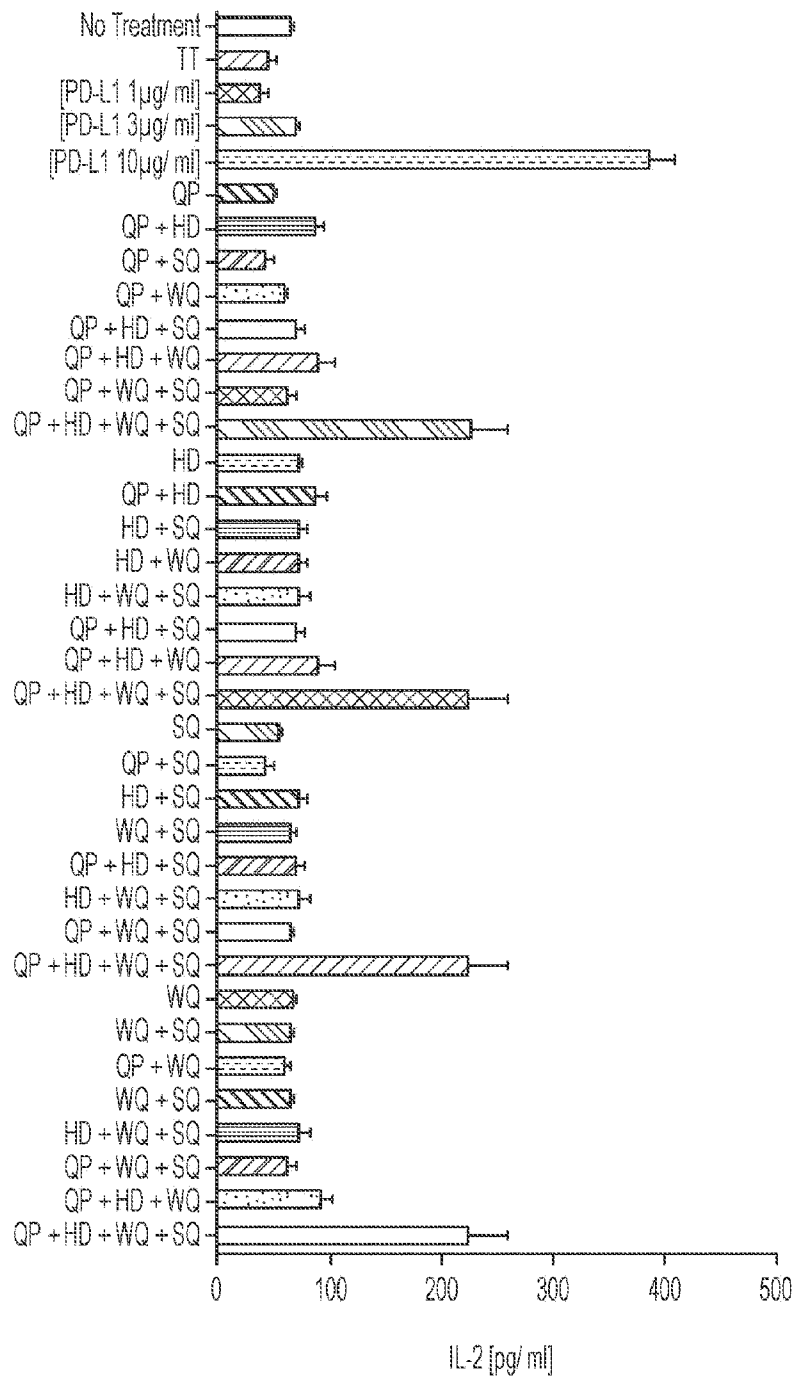


FIG. 17

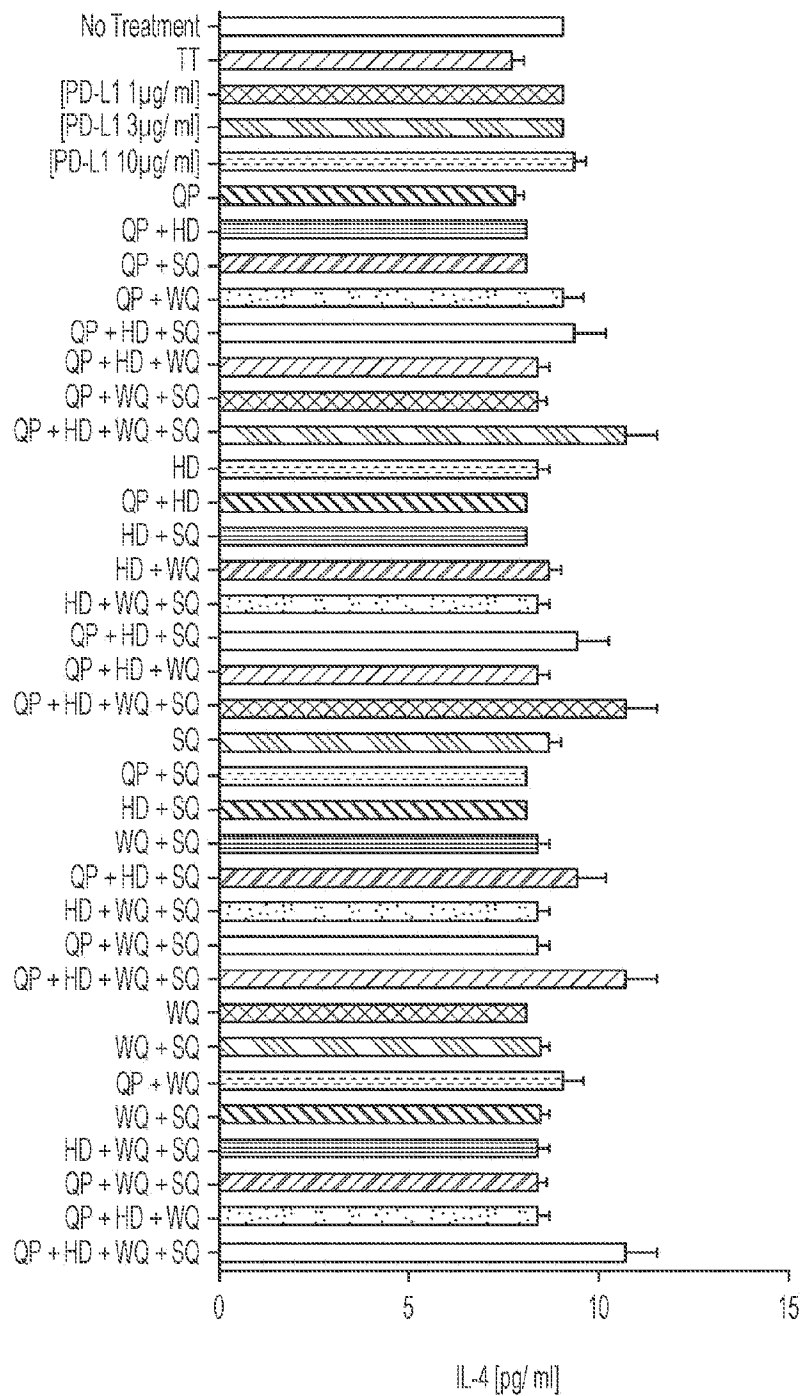


FIG. 18

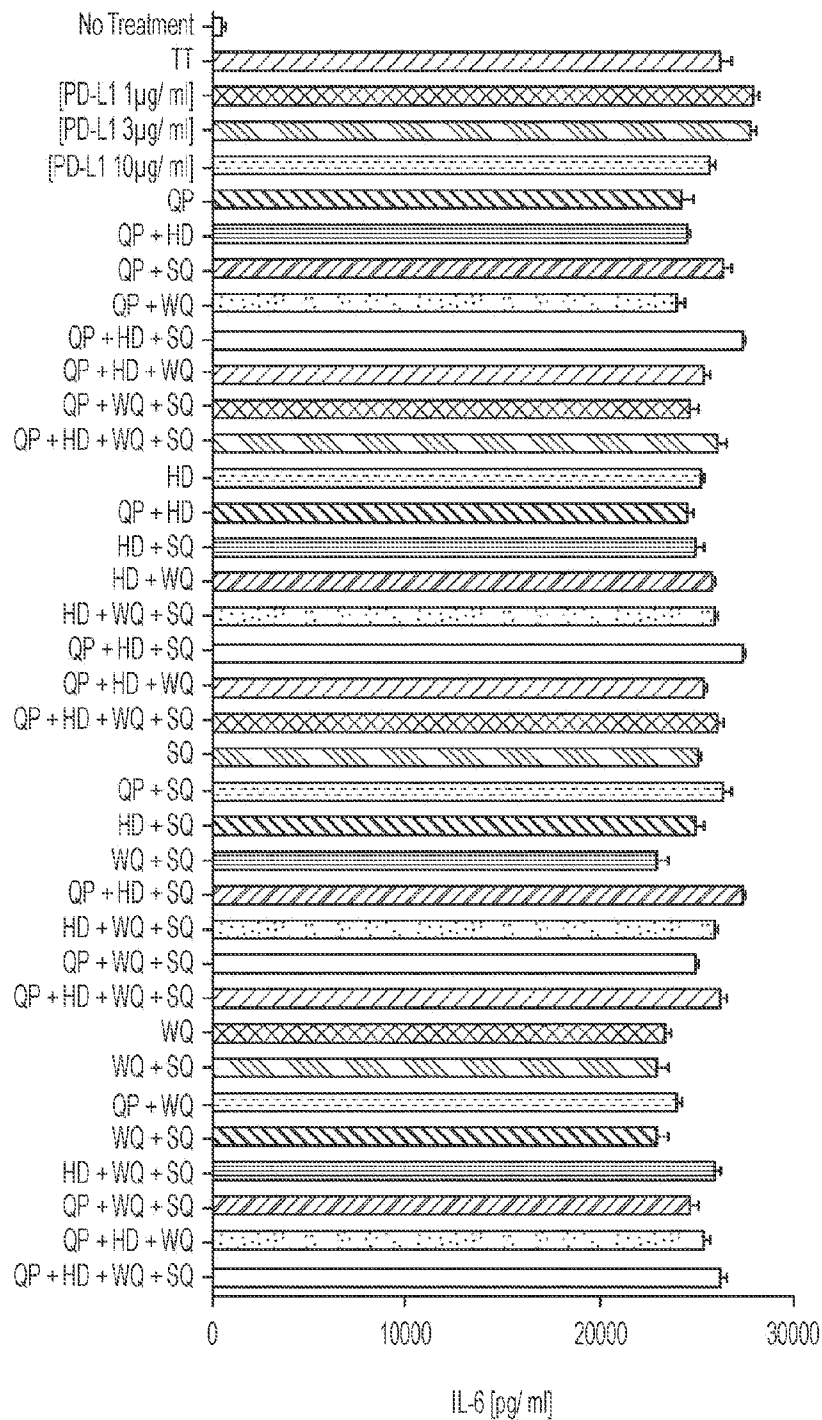


FIG. 19

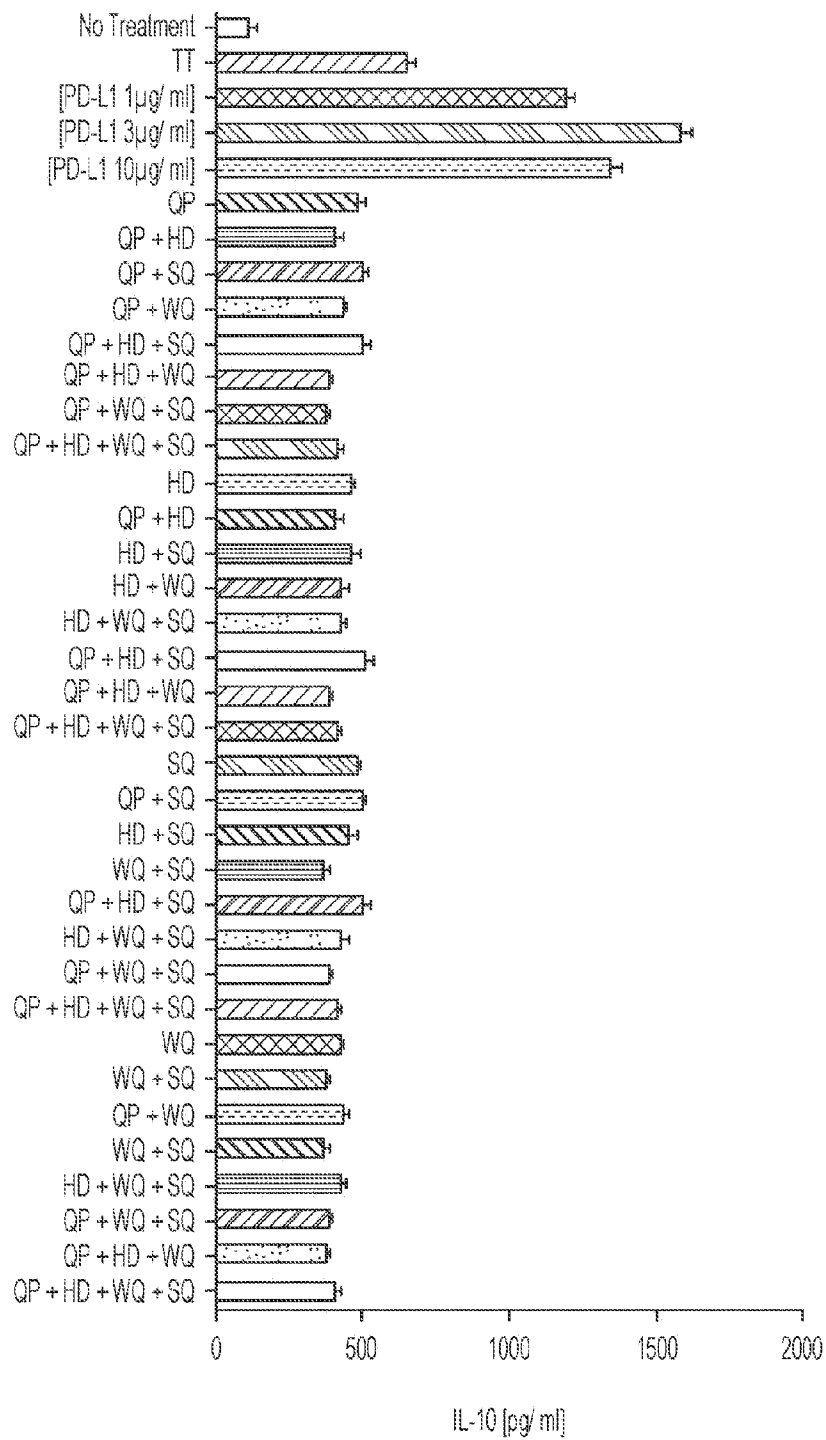


FIG. 20

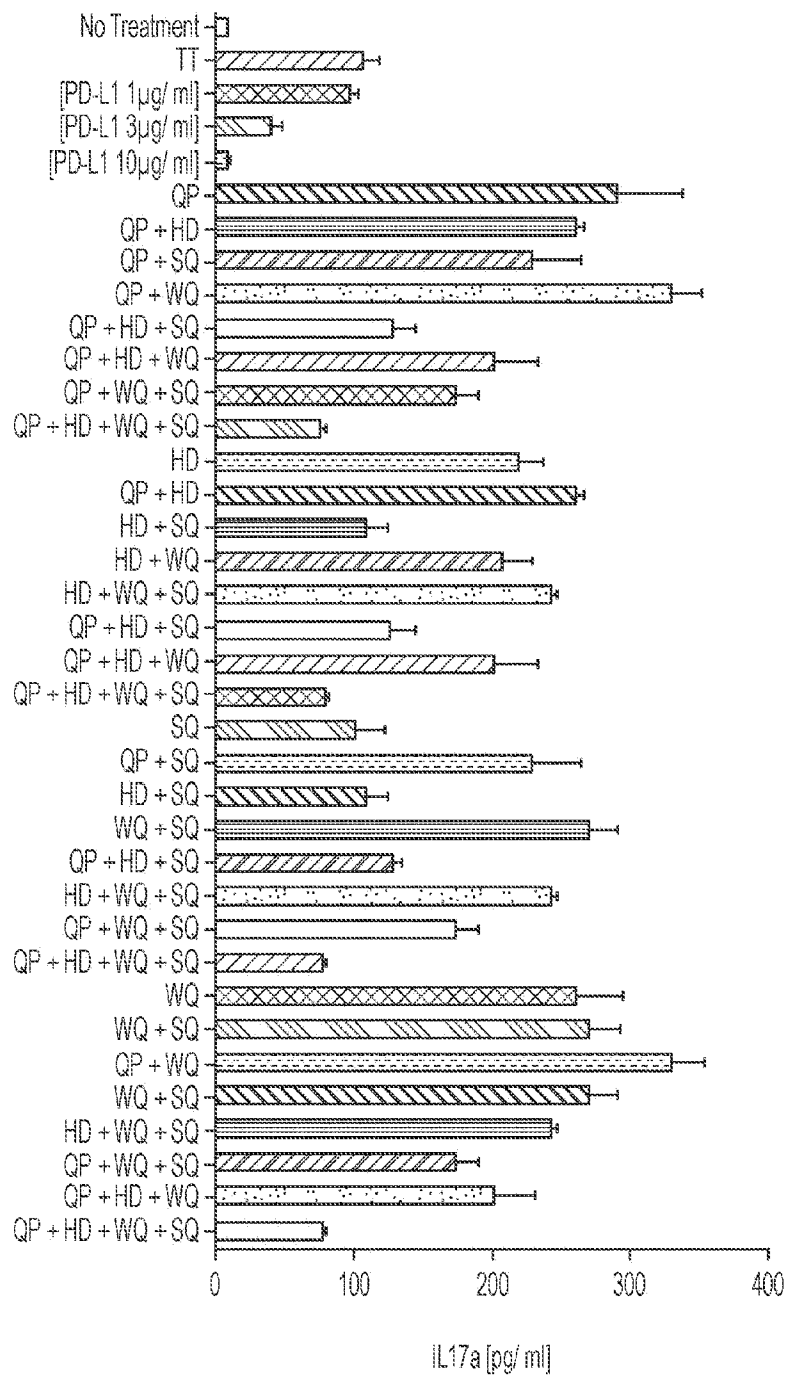


FIG. 21

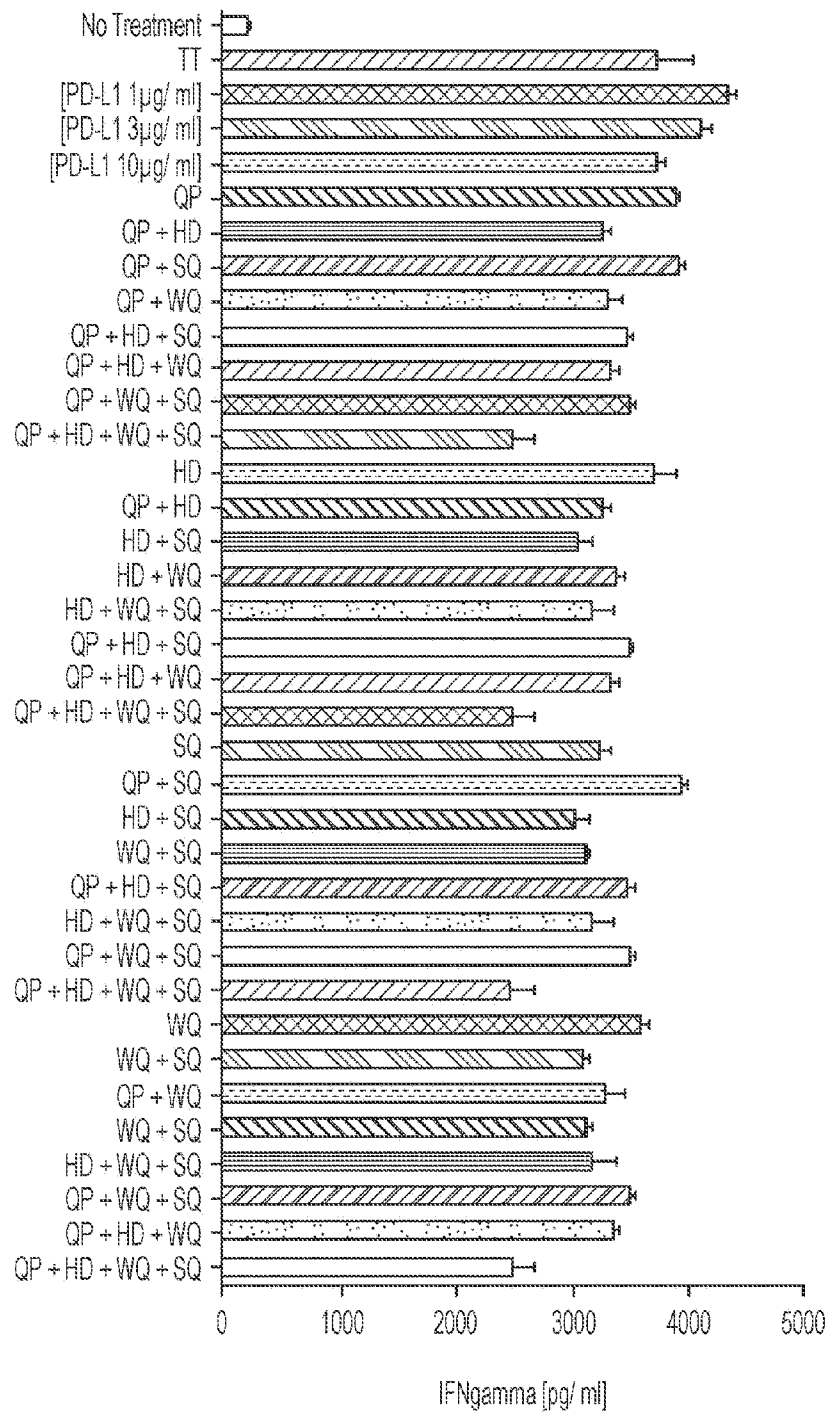


FIG. 22

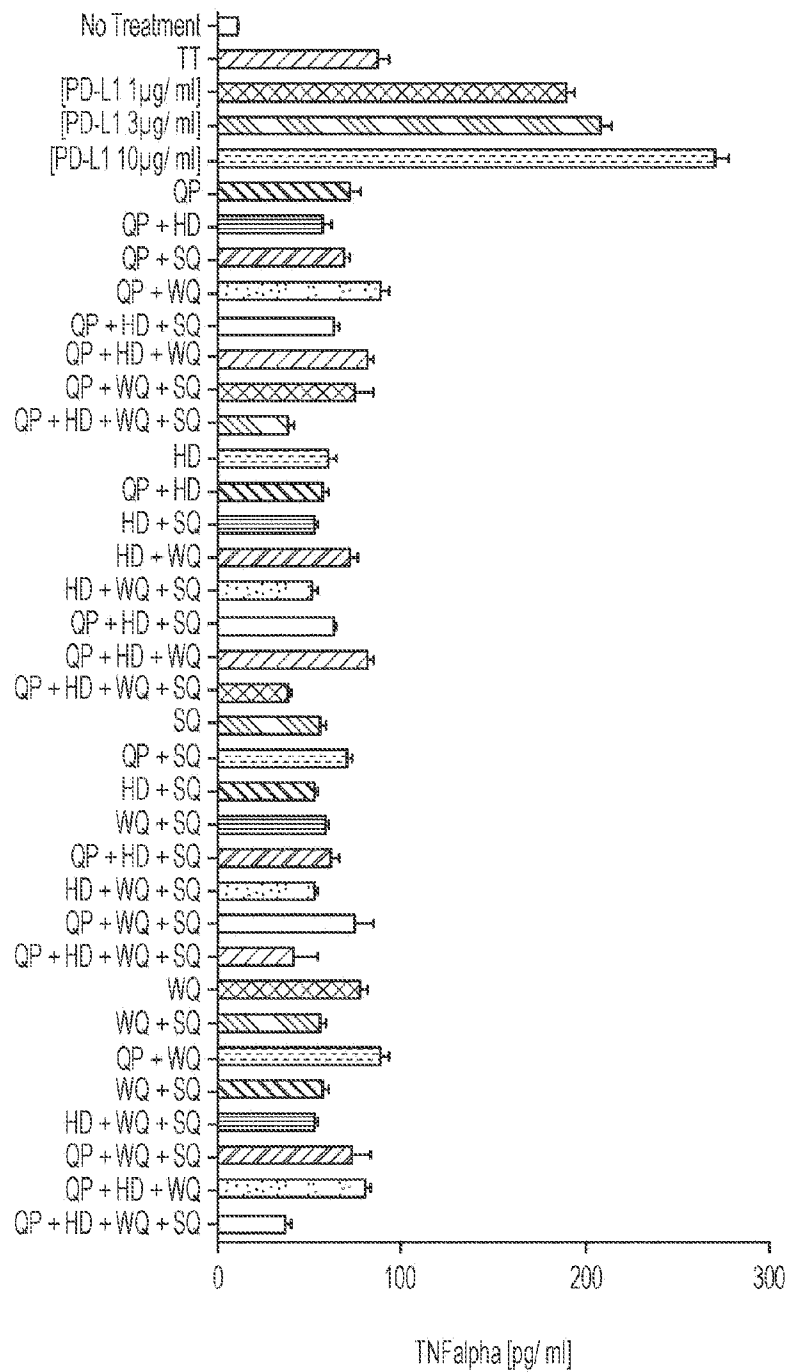


FIG. 23A

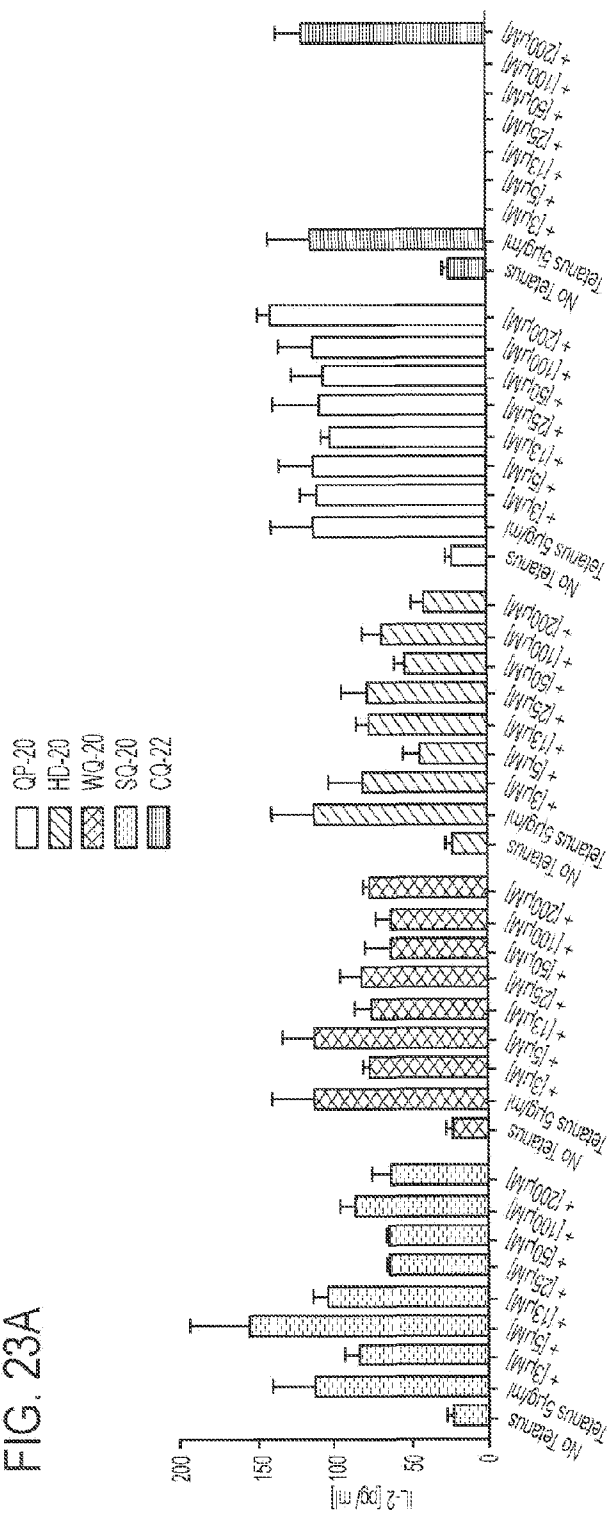


FIG. 23B

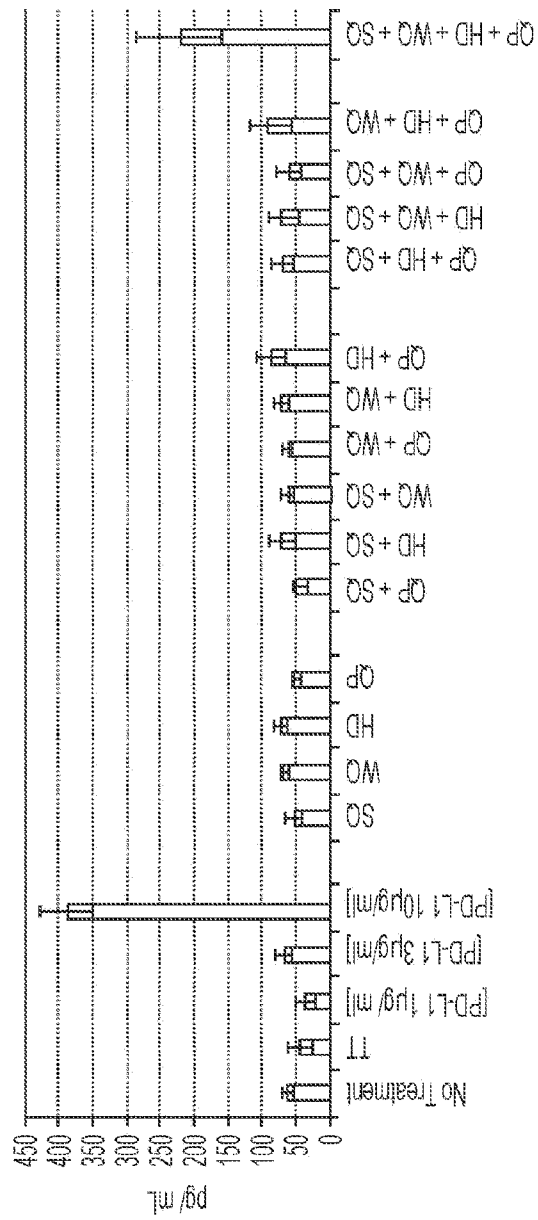


FIG. 24A

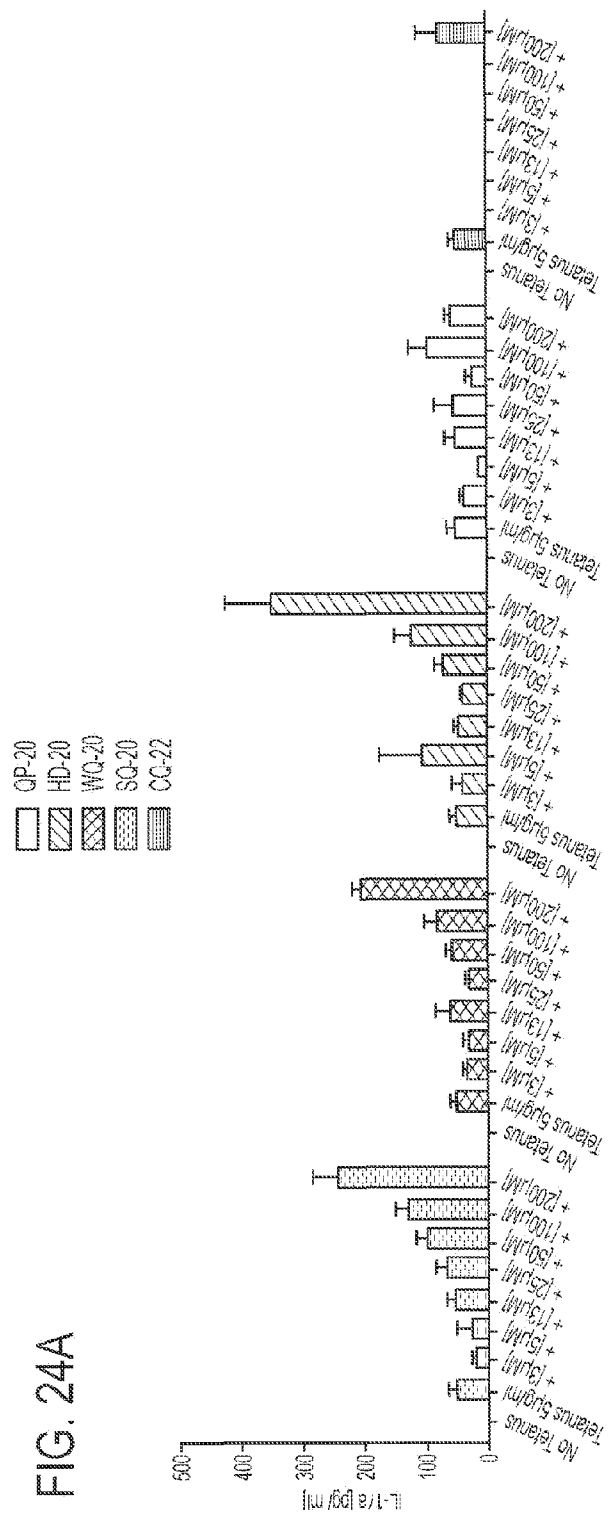


FIG. 24B

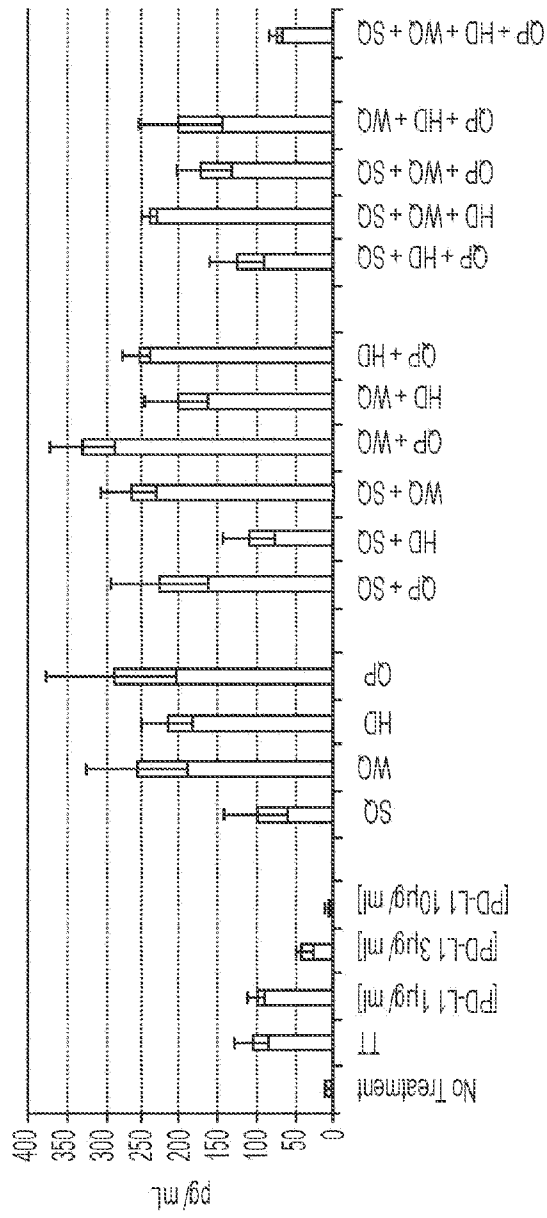


FIG. 25

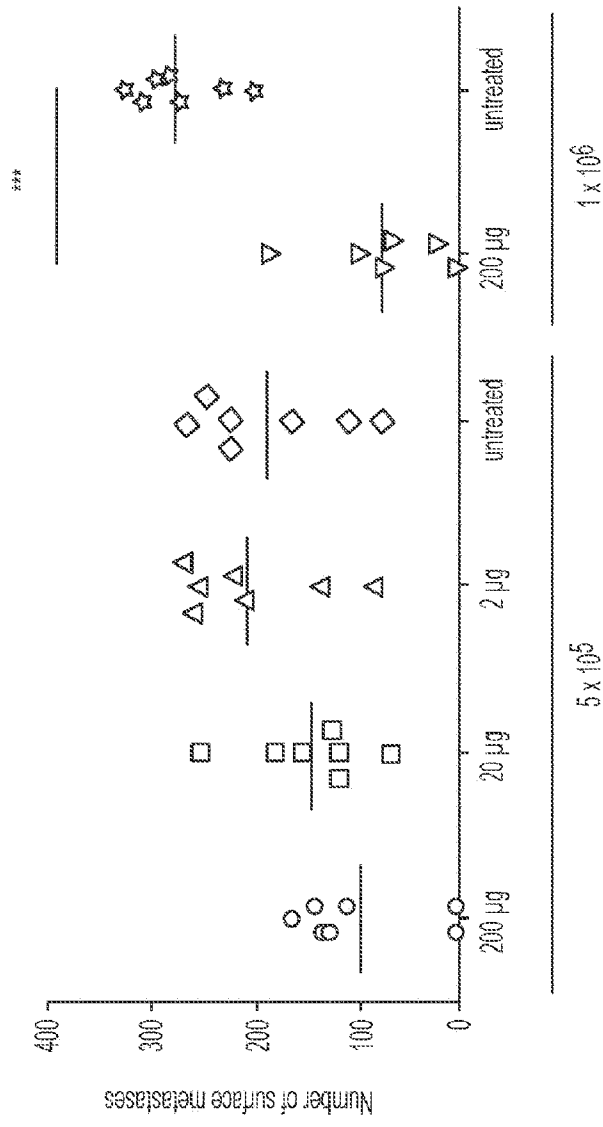


FIG. 26

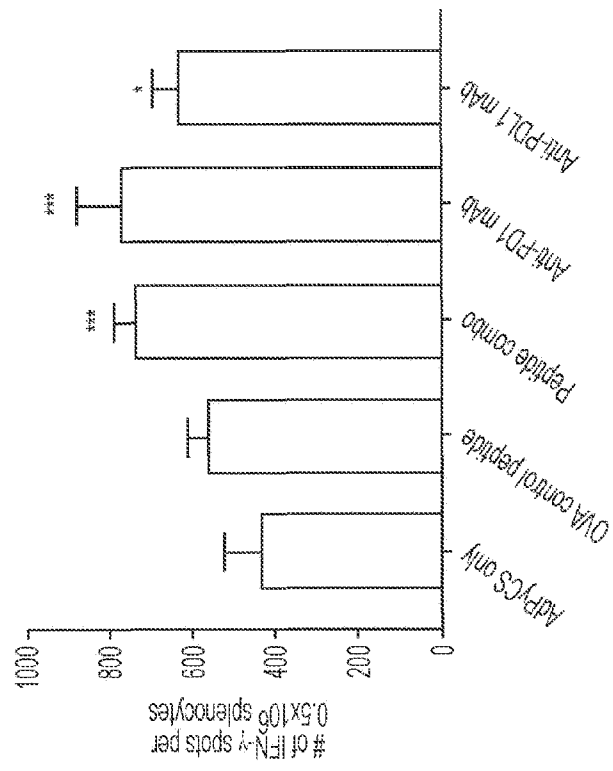
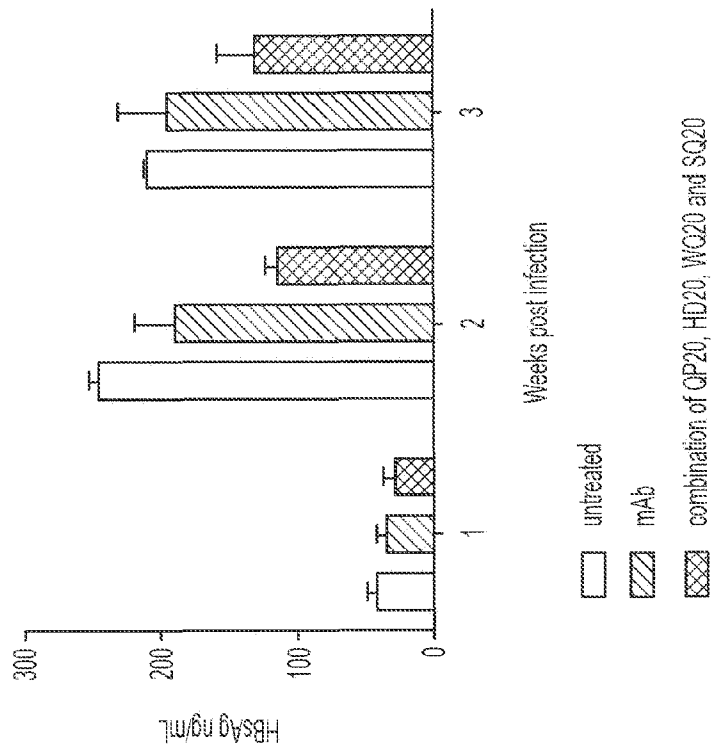


FIG. 27



SEKVENSLISTE

Sekvenslisten er udeladt af skriftet og kan hentes fra det Europæiske Patent Register.

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