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(54) Title: TREATMENT OF LAG-3 POSITIVE TUMORS

(57) Abstract: The invention provides a method of treating a tumor in a human patient comprising (i) identifying a patient as having a LAG-3 positive tumor and (ii) administering to the patient a PD-1 pathway inhibitor, a combination of a PD1 pathway inhibitor and an immune checkpoint inhibitor, a combination of a LAG-3 inhibitor and a PD-1 pathway inhibitor, or an anti-CTLA4 antibody. In some embodiments, the method further comprises identifying the patient as having a LAG-3 positive PD-L1 positive tumor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. The methods of the invention can improve response rates to treatment with a PD-1 pathway inhibitor, a combination of a PD1 pathway inhibitor and an immune checkpoint inhibitor, or a combination of a LAG-3 inhibitor and a PD-1 pathway inhibitor.

TREATMENT OF LAG-3 POSITIVE TUMORS

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to U.S. Provisional Application Nos. 62/512,648, filed May 30, 2017; 62/513,813, filed June 1, 2017; 62/555,176, filed September 7, 2017; and 62/582,178, filed November 6, 2017, which are incorporated herein by reference in their entireties.

FIELD OF THE INVENTION

[0002] The invention disclosed herein relates to methods of treating a LAG-3 positive malignant tumor in a human patient with a PD-1 pathway inhibitor, a combination of a PD1 pathway inhibitor and an immune checkpoint inhibitor, a combination of a LAG-3 inhibitor and a PD-1 pathway inhibitor, or an anti-CTLA-4 antibody.

BACKGROUND OF THE INVENTION

[0003] Lymphocyte activation gene-3 (LAG-3; CD223) is a type I transmembrane protein that is expressed on the cell surface of activated CD4+ and CD8+ T cells and subsets of NK and dendritic cells (Triebel F, et al., J. Exp. Med. 1990; 171:1393-1405; Workman C J, et al., J. Immunol. 2009; 182(4):1885-91). LAG-3 is closely related to CD4, which is a co-receptor for T helper cell activation. Both molecules have 4 extracellular Ig-like domains and require binding to their ligand, major histocompatibility complex (MHC) class II, for their functional activity. In contrast to CD4, LAG-3 is only expressed on the cell surface of activated T cells and its cleavage from the cell surface terminates LAG-3 signaling. LAG-3 can also be found as a soluble protein but it does not bind to MHC class II and the function of soluble LAG-3 is unknown.

[0004] It has been reported that LAG-3 plays an important role in promoting regulatory T cell (Treg) activity and in negatively regulating T cell activation and proliferation (Workman C J, et al., J. Immunol. 2005; 174:688-695). Both natural and induced Treg express increased LAG-3, which is required for their maximal suppressive function (Camisaschi C, et al., J. Immunol. 2010; 184:6545-6551 and Huang C T, et al., Immunity. 2004; 21:503-513). Furthermore, ectopic expression of LAG-3 on CD4+ effector T cells reduced their proliferative capacity and conferred on them regulatory potential against third party T cells (Huang C T, et al., Immunity. 2004;

21:503-513). Recent studies have also shown that high LAG-3 expression on exhausted lymphocytic choriomeningitis virus (LCMV)-specific CD8+ T cells contributes to their unresponsive state and limits CD8+ T cell antitumor responses (Blackburn S D, et al., Nat. Immunol. 2009; 10:29-37 and Grosso J F, et al., J. Clin. Invest. 2007; 117:3383-3392). In fact, LAG-3 maintained tolerance to self and tumor antigens via direct effects on CD8+ T cells in 2 murine models (Grosso J F, et al., J. Clin. Invest. 2007; 117:3383-3392).

[0005] Immune tolerance observed in the setting of tumor development and tumor recurrence, however, seems to be mediated by the co-expression of various T cell negative regulatory receptors, not solely from LAG-3. Data from chronic viral infection models (Blackburn S D, et al., Nat. Immunol. 2009; 10:29-37, Grosso J F, et al., J. Clin. Invest. 2007; 117:3383-3392, and Lyford-Pike S, et al., Cancer Res. 2013; 73(6):1733-41), knock-out mice (Woo S R, et al., Cancer Res. 2012; 72:917-927; Okazaki T, et al., J. Exp Med. 2011; 208:395-407, and Bettini M. et al., J. Immunol. 2011; 187:3493-3498), tumor recurrence models (Goding S R, et al., J. Immunol. 2013; 190(9):4899-4909) and, to a more limited extent, human cancer patients (Goding S R, et al., J. Immunol. 2013; 190(9):4899-4909, Matsuzaki J, et al., Proc. Natl. Acad. Sci., USA. 2010; 107:7875-7880, and Gandhi M K, et al., Blood. 2006; 108:2280-2289) support a model wherein T cells that are continuously exposed to antigen become progressively inactivated through a process termed "exhaustion." Exhausted T cells are characterized by the expression of T cell negative regulatory receptors, predominantly Cytotoxic T-Lymphocyte Antigen-4 (CTLA-4), Programmed Cell Death 1 (PD-1), and LAG-3, whose action is to limit the cell's ability to proliferate, produce cytokines, and kill target cells and/or to increase Treg activity. However, the timing and sequence of expression of these molecules in the development and recurrence of tumors have not been fully characterized.

[0006] PD-1 is a cell surface signaling receptor that plays a critical role in the regulation of T cell activation and tolerance (Keir M E, et al., Annu Rev Immunol 2008; 26:677-704). It is a type I transmembrane protein and together with BTLA, CTLA-4, ICOS and CD28, comprise the CD28 family of T cell co-stimulatory receptors. PD-1 is primarily expressed on activated T cells, B cells, and myeloid cells (Dong H, et al., Nat Med. 1999; 5:1365-1369). It is also expressed on natural killer (NK) cells (Terme M, et al., Cancer Res 2011; 71:5393-5399). Binding of PD-1 by its ligands, PD-L1 and PD-L2, results in phosphorylation of the tyrosine residue in the proximal intracellular immune receptor tyrosine inhibitory domain, followed by recruitment of the phosphatase SHP-2, eventually resulting in down-regulation of T cell activation. One important

role of PD-1 is to limit the activity of T cells in peripheral tissues at the time of an inflammatory response to infection, thus limiting the development of autoimmunity (Pardoll D M., Nat Rev Cancer 2012; 12:252-264). Evidence of this negative regulatory role comes from the finding that PD-1-deficient mice develop lupus-like autoimmune diseases including arthritis and nephritis, along with cardiomyopathy (Nishimura H, et al., Immunity, 1999; 11:141-151; and Nishimura H, et al., Science, 2001; 291:319-322). In the tumor setting, the consequence is the development of immune resistance within the tumor microenvironment. PD-1 is highly expressed on tumor-infiltrating lymphocytes, and its ligands are up-regulated on the cell surface of many different tumors (Dong H, et al., Nat Med 2002; 8:793-800). Multiple murine cancer models have demonstrated that binding of ligand to PD-1 results in immune evasion. In addition, blockade of this interaction results in anti-tumor activity (Topalian S L, et al. NEJM 2012; 366(26):2443-2454; Hamid O, et al., NEJM 2013; 369:134-144). Moreover, it has been shown that inhibition of the PD-1/PD-L1 interaction mediates potent antitumor activity in preclinical models (U.S. Pat. Nos. 8,008,449 and 7,943,743).

[0007] Recently, several immune checkpoint pathway inhibitors have begun to provide new immunotherapeutic approaches for treating cancer, including the development of an antibody (Ab), ipilimumab (YERVOY®), that binds to and inhibits Cytotoxic T-Lymphocyte Antigen-4 (CTLA-4) for the treatment of patients with advanced melanoma, the development of antibodies such as nivolumab and pembrolizumab (formerly lambrolizumab; USAN Council Statement, (2013) Pembrolizumab: Statement on a nonproprietary name adopted by the USAN Council (ZZ-165), November 27, 2013) that bind specifically to the Programmed Death-1 (PD-1) receptor and block the inhibitory PD-1/PD-1 ligand pathway, and the development of an antibody, BMS-986016 (as described in US Pat. No. 9,505,839) that specifically binds LAG-3 and is capable of stimulating immune responses.

[0008] The promise of the emerging field of personalized medicine is that advances in pharmacogenomics will increasingly be used to tailor therapeutics to defined subpopulations, and ultimately, individual patients in order to enhance efficacy and minimize adverse effects. Recent successes include, for example, the development of imatinib mesylate (GLEEVEC®), a protein tyrosine kinase inhibitor that inhibits the bcr-abl tyrosine kinase, to treat Philadelphia chromosome-positive chronic myelogenous leukemia (CML); crizotinib (XALKORI®) to treat the 5% of patients with late-stage non-small cell lung cancers who express a mutant anaplastic lymphoma kinase (ALK) gene; and vemurafenib (ZELBORAF®), an inhibitor of mutated B-

RAF protein (V600E- BRAF) which is expressed in around half of melanoma tumors. However, unlike the clinical development of small molecule agents that target discrete activating mutations found in select cancer populations, a particular challenge in cancer immunotherapy has been the identification of predictive biomarkers to enable patient selection and guide on-treatment management. Accordingly, it is an object of the present invention to provide improved methods for treating tumors.

SUMMARY OF THE INVENTION

[0009] One aspect of the invention disclosed herein relates to a method of selecting a malignant tumor in a human patient for treating with a PD-1 pathway inhibitor, a LAG-3 inhibitor, a combination of a PD1 pathway inhibitor and an immune checkpoint inhibitor, or a combination of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the method comprises detecting LAG-3 expression in the tumor. In some embodiments, the method comprises detecting LAG-3 expression and PD-L1 expression in the tumor. Also disclosed herein are methods of treating LAG-3 positive tumors in a human patient comprising administering a LAG-3 inhibitor and a PD-1 pathway inhibitor.

[0010] One aspect of the invention disclosed herein relates to a method of selecting a malignant tumor in a human patient for immunotherapy, comprising: determining the level of LAG-3 expression in a tumor sample; and selecting the tumor for immunotherapy if the tumor is a LAG-3 positive tumor. Another aspect of the invention disclosed herein relates to a method of identifying a malignant tumor in a human patient as eligible for immunotherapy, comprising: determining the level of LAG-3 expression in a tumor sample; and identifying the tumor as eligible for immunotherapy if the tumor is a LAG-3 positive tumor. Another aspect of the invention disclosed herein relates to a method of identifying a malignant tumor in a human patient that is likely to be responsive to an immunotherapy, the method comprising: determining the level of LAG-3 expression in a tumor sample; and identifying the tumor as likely to be responsive to treatment if the tumor is a LAG-3 positive tumor. Another aspect of the invention disclosed herein relates to a method of classifying a malignant tumor in a human patient as likely to be responsive to an immunotherapy, the method comprising: determining the level of LAG-3 expression in a tumor sample; and classifying the tumor as likely to be responsive to immunotherapy if the tumor is a LAG-3 positive tumor. In some embodiments, a method disclosed herein further comprises determining the level of PD-L1 expression in the tumor

sample. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, a method disclosed herein comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, a method disclosed herein comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, a method disclosed herein comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, a method disclosed herein comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, a method disclosed herein comprises administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, a method disclosed herein comprises administering to the patient a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, a method disclosed herein comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, a method disclosed herein comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor.

[0011] Another aspect of the invention disclosed herein relates to a method of identifying a patient with a malignant tumor who is likely to respond to an immunotherapy, the method comprising: determining the level of LAG-3 expression in a tumor sample; and identifying the patient who is likely to respond to treatment if the tumor is a LAG-3 positive tumor. Another aspect of the invention disclosed herein relates to a method of selecting a patient with a malignant tumor for immunotherapy, the method comprising: determining the level of LAG-3 expression in a tumor sample; and selecting the patient for immunotherapy if the tumor is a LAG-3 positive tumor. In some embodiments, a method disclosed herein further comprises determining the level of PD-L1 expression in the tumor sample. In some embodiments, a method disclosed herein comprises administering to the patient a therapeutically effective amount of a

LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, a method disclosed herein comprises administering to the patient a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, a method disclosed herein comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, a method disclosed herein comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor.

[0012] Another aspect of the invention disclosed herein relates to a method of treating a malignant tumor in a human patient, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the patient is predicted to respond to treatment with the LAG-3 inhibitor and PD-1 pathway inhibitor based upon LAG-3 expression in a sample of the patient's tumor. Another aspect of the invention disclosed herein relates to a method of treating a malignant tumor in a human patient, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor; wherein the patient is predicted to respond to treatment with the LAG-3 inhibitor based upon LAG-3 expression in a sample of the patient's tumor. Another aspect of the invention disclosed herein relates to a method of treating a malignant tumor in a human patient, comprising: administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor; wherein the patient is predicted to respond to treatment with the PD-1 pathway inhibitor based upon LAG-3 expression in a sample of the patient's tumor. Another aspect of the invention disclosed herein relates to a method of treating a malignant tumor in a human patient, comprising: administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor; wherein the patient is predicted to respond to treatment with the PD-1 pathway inhibitor and an immune checkpoint inhibitor based upon LAG-3 expression in a sample of the patient's tumor. In some embodiments, the patient is predicted to respond to the treatment based upon LAG-3 and PD-L1 expression in a sample of the patient's tumor.

[0013] Another aspect of the invention disclosed herein relates to a method of treating a malignant tumor in a human patient in need thereof, comprising: determining the level of LAG-3 expression in a tumor sample; and administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor if the tumor is a LAG-3 positive tumor. Another aspect of the invention disclosed herein relates to a method of treating a malignant tumor in a human patient in need thereof, comprising: determining the level of LAG-3 expression

in a tumor sample; and administering to the patient a therapeutically effective amount of a LAG-3 inhibitor if the tumor is a LAG-3 positive tumor. Another aspect of the invention disclosed herein relates to a method of treating a malignant tumor in a human patient in need thereof, comprising: determining the level of LAG-3 expression in a tumor sample; and administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor if the tumor is a LAG-3 positive tumor. Another aspect of the invention disclosed herein relates to a method of treating a malignant tumor in a human patient in need thereof, comprising: determining the level of LAG-3 expression in a tumor sample; and administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor if the tumor is a LAG-3 positive tumor. In some embodiments, a method disclosed herein further comprises determining the level of PD-L1 expression in the tumor sample.

[0014] Another aspect of the invention disclosed herein relates to a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration. Another aspect of the invention disclosed herein relates to a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration. Another aspect of the invention disclosed herein relates to a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration. Another aspect of the invention disclosed herein relates to a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration. In some embodiments, the patient is identified as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the patient is identified as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration.

[0015] Another aspect of the invention disclosed herein relates to a method for treating a malignant tumor in a human patient in need thereof, comprising: identifying the patient as having

a LAG-3 positive malignant tumor; and administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. Another aspect of the invention disclosed herein relates to a method for treating a malignant tumor in a human patient in need thereof, comprising: identifying the patient as having a LAG-3 positive malignant tumor; and administering to the patient a therapeutically effective amount of a LAG-3 inhibitor. Another aspect of the invention disclosed herein relates to a method for treating a malignant tumor in a human patient in need thereof, comprising: identifying the patient as having a LAG-3 positive malignant tumor; and administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor. Another aspect of the invention disclosed herein relates to a method for treating a malignant tumor in a human patient in need thereof, comprising: identifying the patient as having a LAG-3 positive malignant tumor; and administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, a method disclosed herein further comprises identifying the patient as having a LAG-3 positive PD-L1 positive malignant tumor. In some embodiments, a method disclosed herein further comprises identifying the patient as having a LAG-3 positive PD-L1 negative malignant tumor.

[0016] Another aspect of the invention disclosed herein relates to a method for extending a progression-free survival period for over 12 months in a human patient afflicted with a malignant tumor comprising administering to the patient a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months. Another aspect of the invention disclosed herein relates to a method for extending a progression-free survival period for over 12 months in a human patient afflicted with a malignant tumor comprising administering to the patient a LAG-3 inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months. Another aspect of the invention disclosed herein relates to a method for extending a progression-free survival period for over 12 months in a human patient afflicted with a malignant tumor comprising administering to the patient a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months. Another aspect of the invention disclosed herein relates to a method for extending a progression-free survival period for over 12 months in a human patient

afflicted with a malignant tumor comprising administering to the patient a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months. In some embodiments, the patient is identified as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the patient is identified as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration. In some embodiments, the progression-free survival of the patient is extended after the administration for over about 13 months, about 14 months, about 15 months, about 16 months, about 17 months, about 18 months, about 2 years, about 3 years, about 4 years, about 5 years, about 6 years, about 7 years, about 8 years, about 9 years, or about 10 years. In certain embodiments, the progression-free survival of the patient is extended for over 14 months.

[0017] Another aspect of the invention disclosed herein relates to a method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration. Another aspect of the invention disclosed herein relates to a method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration. Another aspect of the invention disclosed herein relates to a method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration. Another aspect of the invention disclosed

herein relates to a method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration. In some embodiments, the patient is identified as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the patient is identified as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration. In some embodiments, a method disclosed herein further comprises identifying the patient as having a LAG-3 positive malignant tumor prior to the administration. In some embodiments, a method disclosed herein further comprises identifying the patient as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, a method disclosed herein further comprises identifying the patient as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration. In some embodiments, the patient experiences (i) extended progression-free survival for over 12 months, (ii) tumor size reduction at least about 10%, about 20%, about 30%, about 40%, or about 50% compared to the tumor size prior to the administration, or (iii) both.

[0018] Another aspect of the invention disclosed herein relates to a method for increasing an objective response rate to a cancer treatment to be higher than 50% in a human patient population, each of whom is afflicted with a malignant tumor, to a cancer treatment comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 55%, 60%, 65%, 70%, or 75%. Another aspect of the invention disclosed herein relates to a method for increasing an objective response rate to a cancer treatment to be higher than 50% in a human patient population, each of whom is afflicted with a malignant tumor, to a cancer treatment comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 55%, 60%, 65%, 70%, or 75%. Another aspect of the invention disclosed herein relates to a method for increasing an objective response rate to a cancer treatment to be higher than 50% in a human patient population, each of whom is

afflicted with a malignant tumor, to a cancer treatment comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 55%, 60%, 65%, 70%, or 75%. Another aspect of the invention disclosed herein relates to a method for increasing an objective response rate to a cancer treatment to be higher than 50% in a human patient population, each of whom is afflicted with a malignant tumor, to a cancer treatment comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 55%, 60%, 65%, 70%, or 75%. In some embodiments, each patient is identified as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, each patient is identified as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration.

[0019] Another aspect of the invention disclosed herein relates to a method for increasing a disease control rate to be higher than 50% in a human patient population, each of whom is afflicted with a malignant tumor, to a cancer treatment comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 55%, 60%, 65%, 70%, or 75%. Another aspect of the invention disclosed herein relates to a method for increasing a disease control rate to be higher than 50% in a human patient population, each of whom is afflicted with a malignant tumor, to a cancer treatment comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 55%, 60%, 65%, 70%, or 75%. Another aspect of the invention disclosed herein relates to a method for increasing a disease control rate to be higher than 50% in a human patient population, each of whom is afflicted with a malignant tumor, to a cancer treatment comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 55%, 60%, 65%, 70%, or 75%. Another aspect of the invention disclosed herein relates to a method for increasing a disease

control rate to be higher than 50% in a human patient population, each of whom is afflicted with a malignant tumor, to a cancer treatment comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 55%, 60%, 65%, 70%, or 75%. In some embodiments, each patient is identified as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, each patient is identified as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration. In some embodiments, the median duration of response is $\geq$ 3 month, $\geq$ 6 month, $\geq$ 12 month, or $\geq$ 18 month.

[0020] In some embodiments, a method disclosed herein further comprises identifying each patient of the patient population as having a LAG-3 positive malignant tumor prior to the administration. In some embodiments, a method disclosed herein further comprises identifying each patient of the patient population as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, a method disclosed herein further comprises identifying each patient of the patient population as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration. In some embodiments, each patient of the patient population is further characterized by (i) extended progression-free survival for over 12 months, (ii) tumor size reduction at least about 10%, about 20%, about 30%, about 40%, or about 50% compared to the tumor size prior to the administration, or (iii) both. In some embodiments, the patient population comprises at least about 100, 200, 300, 400, 500, 600, 700, 800, 900, or 1000 patients having a LAG-3 positive malignant tumor.

[0021] Another aspect of the invention disclosed herein relates to a method for selecting a human patient suitable for a combination therapy comprising: identifying a patient as having a LAG-3 positive malignant tumor; and instructing a healthcare provider to administer to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. Another aspect of the invention disclosed herein relates to a method for selecting a human patient suitable for a combination therapy comprising: identifying a patient as having a LAG-3 positive malignant tumor; and instructing a healthcare provider to administer to the patient a therapeutically effective amount of a LAG-3 inhibitor. Another aspect of the invention disclosed herein relates to a method for selecting a human patient suitable for a combination therapy comprising: identifying a patient as having a LAG-3 positive malignant tumor; and instructing a

healthcare provider to administer to the patient a therapeutically effective amount of a PD-1 pathway inhibitor. Another aspect of the invention disclosed herein relates to a method for selecting a human patient suitable for a combination therapy comprising: identifying a patient as having a LAG-3 positive malignant tumor; and instructing a healthcare provider to administer to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, a method disclosed herein further comprises identifying the patient as having a LAG-3 positive PD-L1 positive malignant tumor. In some embodiments, a method disclosed herein further comprises identifying the patient as having a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the administration treats the malignant tumor.

[0022] In some embodiments, identifying the patient as having a LAG-3 positive malignant tumor comprises determining LAG-3 expression in the malignant tumor. In some embodiments, identifying the patient as having a LAG-3 positive PD-L1 positive malignant tumor comprises determining PD-L1 expression in the malignant tumor. In some embodiments, identifying the patient as having a LAG-3 positive PD-L1 negative malignant tumor comprises determining PD-L1 expression in the malignant tumor. In some embodiments, LAG-3 expression is determined by reviewing the results of an assay capable of determining LAG-3 expression. In some embodiments, LAG-3 expression is determined by reviewing the results of an immunohistochemistry assay capable of detecting LAG-3 expression. In some embodiments, PD-L1 expression is determined by reviewing the results of an assay capable of determining PD-L1 expression. In some embodiments, PD-L1 expression is determined by reviewing the results of an immunohistochemistry assay capable of detecting PD-L1 expression.

[0023] In certain embodiments, a LAG-3 positive tumor comprises at least about 1%, at least about 2%, at least about 3%, at least about 4%, at least about 5%, at least about 7%, at least about 10%, at least about 15%, at least about 20%, at least about 25%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, or 100% cells expressing LAG-3. In certain embodiments, a LAG-3 positive tumor comprises at least about 1% cells expressing LAG-3. In certain embodiments, a LAG-3 positive tumor comprises at least about 5% cells expressing LAG-3. In some embodiments, the cells expressing LAG-3 comprise tumor infiltrating lymphocytes. In certain embodiments, the cells expressing LAG-3 are the total number of cells. In other embodiments, the cells express LAG-3 on the cell surface.

[0024] In some embodiments, the malignant tumor is selected from the group consisting of a liver cancer, bone cancer, pancreatic cancer, skin cancer, oral cancer, cancer of the head or neck, breast cancer, lung cancer, including small cell and non-small cell lung cancer, cutaneous or intraocular malignant melanoma, renal cancer, uterine cancer, ovarian cancer, colorectal cancer, colon cancer, rectal cancer, cancer of the anal region, stomach cancer, testicular cancer, uterine cancer, carcinoma of the fallopian tubes, carcinoma of the endometrium, carcinoma of the cervix, carcinoma of the vagina, carcinoma of the vulva, non-Hodgkin's lymphoma, cancer of the esophagus, cancer of the small intestine, cancer of the endocrine system, cancer of the thyroid gland, cancer of the parathyroid gland, cancer of the adrenal gland, sarcoma of soft tissue, cancer of the urethra, cancer of the penis, cancers of the childhood, lymphocytic lymphoma, cancer of the bladder, cancer of the kidney or ureter, carcinoma of the renal pelvis, neoplasm of the central nervous system (CNS), primary CNS lymphoma, tumor angiogenesis, spinal axis tumor, brain stem glioma, pituitary adenoma, Kaposi's sarcoma, epidermoid cancer, squamous cell cancer, environmentally induced cancers including those induced by asbestos, hematologic malignancies including, for example, multiple myeloma, B-cell lymphoma, Hodgkin lymphoma/primary mediastinal B-cell lymphoma, non-Hodgkin's lymphomas, acute myeloid lymphoma, chronic myelogenous leukemia, chronic lymphoid leukemia, follicular lymphoma, diffuse large B-cell lymphoma, Burkitt's lymphoma, immunoblastic large cell lymphoma, precursor B-lymphoblastic lymphoma, mantle cell lymphoma, acute lymphoblastic leukemia, mycosis fungoides, anaplastic large cell lymphoma, T-cell lymphoma, and precursor T-lymphoblastic lymphoma, and any combination thereof.

[0025] In some embodiments, the malignant tumor is chosen from melanoma, non-small cell lung cancer (NSCLC), human papilloma virus (HPV)-related tumor, and gastric adenocarcinoma.

[0026] In some embodiments, the malignant tumor is NSCLC, a virally-related cancer related tumor, or gastric adenocarcinoma.

[0027] In some embodiments, the malignant tumor is melanoma, gastric cancer, gastroesophageal junction cancer, non-small cell lung cancer, bladder cancer, head and neck squamous cell carcinoma, or renal cell cancer.

[0028] In some embodiments, the malignant tumor is lung cancer, melanoma, squamous cell carcinoma of the head and neck, renal cancer, gastric cancer, or hepatocellular carcinoma.

[0029] In some embodiments, the LAG-3 positive malignant tumor is a melanoma tumor comprising about 1% or more cells expressing LAG-3.

[0030] In some embodiments, the LAG-3 positive malignant tumor is a gastric cancer tumor comprising about 1% or more cells expressing LAG-3.

[0031] In some embodiments, the malignant tumor is refractory to treatment with an immune checkpoint inhibitor. In some embodiments, the malignant tumor is refractory to treatment with an anti-PD-1 antibody. In some embodiments, the malignant tumor is refractory to treatment with an anti-PD-L1 antibody.

[0032] Another aspect of the invention disclosed herein relates to a method for treating melanoma in a human patient, comprising: identifying the patient as having a LAG-3 positive melanoma; and administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, identifying the patient as having a LAG-3 positive melanoma comprises determining LAG-3 expression in the melanoma tumor. In some embodiments, LAG-3 expression is determined by reviewing the results of an assay capable of determining LAG-3 expression. In some embodiments, LAG-3 expression is determined by an immunohistochemistry assay capable of detecting LAG-3 expression. In some embodiments, a method disclosed herein further comprises identifying the patient as having a LAG-3 positive PD-L1 positive malignant tumor. In some embodiments, a method disclosed herein further comprises identifying the patient as having a LAG-3 positive PD-L1 negative malignant tumor.

[0033] Another aspect of the invention disclosed herein relates to a method for treating a melanoma in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive melanoma prior to the administration. Another aspect of the invention disclosed herein relates to a method for extending a progression-free survival period for over 12 months in a human patient afflicted with a melanoma comprising administering to the patient a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive melanoma prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months. In some embodiments, the patient is identified as having a LAG-3 positive PD-L1 positive melanoma prior to the administration. In some embodiments, the patient is identified as having a LAG-3 positive PD-L1 negative melanoma prior to the administration.

[0034] Another aspect of the invention disclosed herein relates to a method for increasing an objective response rate to a cancer treatment to be higher than 15% in a human patient population, each of whom is afflicted with melanoma, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 15%. Another aspect of the invention disclosed herein relates to a method for increasing a disease control rate to a cancer treatment to be higher than 70% in a human patient population, each of whom is afflicted with melanoma, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive melanoma prior to the administration and wherein the objective response rate is higher than 70%. In some embodiments, a method disclosed herein further comprises identifying each patient of the patient population as having a LAG-3 positive melanoma prior to the administration. In some embodiments, the median duration of response is $\geq$ 3 month, $\geq$ 6 month, $\geq$ 12 month, or $\geq$ 18 month. In some embodiments, each patient is identified as having a LAG-3 positive PD-L1 positive melanoma prior to the administration. In some embodiments, each patient is identified as having a LAG-3 positive PD-L1 negative melanoma prior to the administration.

[0035] In some embodiments, the melanoma is refractory to treatment with an immune checkpoint inhibitor. In some embodiments, the melanoma is refractory to treatment with an anti-PD-1 antibody or an anti-PD-L1 antibody.

[0036] In some embodiments, determining the level of LAG-3 and/or PD-L1 expression comprises providing a test tissue sample obtained from the patient, the test tissue sample comprising tumor cells and/or tumor-infiltrating immune cells. In some embodiments, the test tissue sample is a tumor biopsy. In some embodiments, the test tissue sample is a formalin-fixed paraffin embedded (FFPE) sample.

[0037] In some embodiments, determining comprises detecting LAG-3 and/or PD-L1 protein or RNA expression in the test tissue sample.

[0038] In some embodiments, LAG-3 and/or PD-L1 expression is detected by an assay capable of detecting the level of LAG-3 and/or PD-L1 protein, respectively, in the test tissue sample.

[0039] In some embodiments, LAG-3 and/or PD-L1 expression is detected by an immunohistochemistry assay. In some embodiments, the immunohistochemistry assay is a

monoplex assay (assay designed to detect/measure the presence of a single analyte, e.g., antigen/antibody pair). In some embodiments, the immunohistochemistry assay is a multiplex assay (assay designed to detect/measure multiple analytes, either simultaneously or sequentially). In some embodiments, the immunohistochemistry assay comprises contacting the tumor sample with the 17B4, SP346, 11E3, 874501, or EPR4392(2) anti-human LAG-3 monoclonal antibody. In some embodiments, the immunohistochemistry assay comprises contacting the tumor sample with an anti-LAG-3 antibody comprising heavy and light chain variable regions comprising the sequences set forth in SEQ ID NOs:3 and 5, respectively.

[0040] In some embodiments, the immunohistochemistry assay uses a black or brown chromogen. In some embodiments, the immunohistochemistry assay uses a red chromogen. In some embodiments, the immunohistochemistry assay uses a blue chromogen. In some embodiments, the immunohistochemistry assay uses a green chromogen. In some embodiments, the immunohistochemistry assay uses a purple chromogen. In certain embodiments, the immunohistochemistry assay uses a yellow chromogen.

[0041] In some embodiments, the immunohistochemistry assay is scored at a low magnification (e.g., 4X or 10X). In some embodiments, low magnification is about 20X.

[0042] In some embodiments, the immunohistochemistry assay is scored at high magnification. In some embodiments, high magnification is about 40X, or greater (60X, 100X).

[0043] In some embodiments, the immunohistochemistry assay is scored by an image analysis software. In some embodiments, the immunohistochemistry assay is scored manually by a pathologist.

[0044] In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of cells in the test tissue sample that express LAG-3 and/or assessing the proportion of cells in the test tissue sample that express PD-L1. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of tumor cells in the test tissue sample that express LAG-3 and/or assessing the proportion of tumor cells in the test tissue sample that express PD-L1. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of immune cells in the test tissue sample that express LAG-3 and/or assessing the proportion of immune cells in the test tissue sample that express PD-L1. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of T cells in the test tissue sample that express LAG-3 and/or assessing the proportion of T cells in the test tissue sample that express PD-L1. In some embodiments, scoring the

immunohistochemistry assay comprises assessing the proportion of CD8+ T cells in the test tissue sample that express LAG-3 and/or assessing the proportion of CD8+ T cells in the test tissue sample that express PD-L1. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of CD4+ T cells in the test tissue sample that express LAG-3 and/or assessing the proportion of CD4+ T cells in the test tissue sample that express PD-L1. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of FOXP3+ T cells in the test tissue sample that express LAG-3 and/or assessing the proportion of FOXP3+ T cells in the test tissue sample that express PD-L1.

[0045] In some embodiments, cells with partial membrane/cytoplasmic LAG-3 localization are scored as LAG-3 expressing cells. In some embodiments, cells with dot-like LAG-3 localization are scored as LAG-3 expressing cells. In some embodiments, cells with complete membrane/cytoplasmic LAG-3 localization are scored as LAG-3 expressing cells. In some embodiments, cells with any LAG-3 localization pattern are scored as LAG-3 expressing cells.

[0046] In some embodiments, the immunohistochemistry assay is a multiplex assay that further comprises detecting the expression of MHC Class II by the tumor cells. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of cells in the test tissue sample that expresses MHC Class II. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of non-immune cells in the test tissue sample that expresses MHC II.

[0047] In some embodiments, LAG-3 and/or PD-L1 protein expression is detected by flow cytometry. In some embodiments, the test tissue sample obtained from the patient comprises tumor infiltrating immune cells. In some embodiments, the malignant tumor is a hematological malignancy and the tissue sample comprises circulating lymphocytes. In some embodiments, the flow cytometry is a multiplex assay. In some embodiments, the flow cytometry comprises detecting the expression of markers comprising LAG-3, PD-L1, CD4, CD8, FOXP3, MHC Class II and any combination thereof.

[0048] In some embodiments, scoring the flow cytometry comprises assessing the proportion of T cells in the test tissue sample that express LAG-3. In some embodiments, scoring the flow cytometry comprises assessing the proportion of CD8+ T cells in the test tissue sample that express LAG-3. In some embodiments, scoring the flow cytometry comprises assessing the proportion of CD4+ T cells in the test tissue sample that express LAG-3. In some embodiments,

scoring the flow cytometry comprises assessing the proportion of FOXP3+ T cells in the test tissue sample that express LAG-3.

[0049] In some embodiments, LAG-3 and/or PD-L1 expression is detected by an assay capable of detecting the level of LAG-3 and/or PD-L1, respectively, RNA in the tumor sample. In some embodiments, LAG-3 and/or PD-L1 expression is detected by an RT-PCR based assay. In some embodiments, scoring the RT-PCR based assay comprises assessing the level of LAG-3 and/or PD-L1 RNA expression in the test tissue sample relative to a predetermined level.

[0050] In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody or antigen-binding fragment thereof. In some embodiments, the anti-LAG-3 antibody is a bispecific antibody.

[0051] In some embodiments, the anti-LAG-3 antibody or antigen-binding fragment thereof comprises (a) a heavy chain variable region CDR1 comprising the sequence set forth in SEQ ID NO:7; (b) a heavy chain variable region CDR2 comprising the sequence set forth in SEQ ID NO:8; (c) a heavy chain variable region CDR3 comprising the sequence set forth in SEQ ID NO:9; (d) a light chain variable region CDR1 comprising the sequence set forth in SEQ ID NO:10; (e) a light chain variable region CDR2 comprising the sequence set forth in SEQ ID NO:11; and (f) a light chain variable region CDR3 comprising the sequence set forth in SEQ ID NO:12.

[0052] In some embodiments, the anti-LAG-3 antibody or antigen-binding fragment thereof comprises heavy and light chain variable regions comprising the sequences set forth in SEQ ID NOs:3 and 5, respectively.

[0053] In some embodiments, the anti-LAG-3 antibody is MK-4280 (28G-10), REGN3767, GSK2837781, IMP731 (H5L7BW), BAP050, IMP-701 (LAG-525), IMP321, FS-118, Sym022, TSR-033, MGD013, FS118, or GSK2831781.

[0054] In some embodiments, the PD-1 pathway inhibitor is an anti-PD-1 antibody or antigen-binding fragment thereof. In some embodiments, the anti-PD-1 antibody or antigen-binding fragment thereof comprises (a) a heavy chain variable region CDR1 comprising the sequence set forth in SEQ ID NO:23; (b) a heavy chain variable region CDR2 comprising the sequence set forth in SEQ ID NO:24; (c) a heavy chain variable region CDR3 comprising the sequence set forth in SEQ ID NO:25; (d) a light chain variable region CDR1 comprising the sequence set forth in SEQ ID NO:26; (e) a light chain variable region CDR2 comprising the

sequence set forth in SEQ ID NO:27; and (f) a light chain variable region CDR3 comprising the sequence set forth in SEQ ID NO:28.

[0055] In some embodiments, the anti-PD-1 antibody or antigen-binding fragment thereof comprises heavy and light chain variable regions comprising the sequences set forth in SEQ ID NOs:19 and 21, respectively.

[0056] In some embodiments, the anti-PD-1 antibody or antigen-binding fragment thereof comprises heavy and light chains comprising the sequences set forth in SEQ ID NOs:17 and 18, respectively.

[0057] In some embodiments, the anti-PD-1 antibody is pembrolizumab (KEYTRUDA; MK-3475), pidilizumab (CT-011), or nivolumab (OPDIVO; BMS-936558).

[0058] In some embodiments, the PD-1 pathway inhibitor is an anti-PD-L1 antibody or antigen-binding fragment thereof. In some embodiments, the anti-PD-L1 antibody is atezolizumab (Tecentriq or RG7446), durvalumab (Imfinzi or MEDI4736), avelumab (Bavencio) or BMS-936559.

[0059] In some embodiments, the PD-1 pathway inhibitor is an anti-PD-L2 antibody or antigen-binding fragment thereof.

[0060] In some embodiments, the immune checkpoint inhibitor is a CTLA-4 antagonist, a CD80 antagonist, a CD86 antagonist, a Tim-3 antagonist, a TIGIT antagonist, a CD20 antagonist, a CD96 antagonist, a IDO1 antagonist, a STING antagonist, a GARP antagonist, a CD40 antagonist, A2aR antagonist, a CEACAM1 (CD66a) antagonist, a CEA antagonist, a CD47 antagonist a PVRIG antagonist, a TDO antagonist, a VISTA antagonist, or a KIR antagonist.

[0061] In some embodiments, the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycles, four doses of the anti-LAG-3 antibody are administered at a dose of 3, 20, 80, 160, or 240 mg.

[0062] In some embodiments, the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycles, four doses of the anti-PD-1 antibody are administered at a dose of 80 or 240 mg.

[0063] In some embodiments, the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycles, four doses of the anti-PD-L1 antibody are administered at a dose of 3, 20, 80, 160, or 240 mg.

[0064] In some embodiments, the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycles, four

doses of the anti-LAG-3 antibody are administered at a dose of 3, 20, 80, 160, or 240 mg and four doses of the anti-PD-1 antibody are administered at a dose of 80 or 240 mg.

[0065] In some embodiments, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the following doses: (a) 3 mg of anti-LAG-3 antibody and 80 mg of anti-PD-1 antibody; (b) 3 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody; (c) 20 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody; (d) 80 mg of anti-LAG-3 antibody and 160 mg of anti-PD-1 antibody; (e) 80 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody; (f) 160 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody, or (g) 240 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody.

[0066] In some embodiments, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the dose of 80 mg of anti-LAG-3 antibody and 160 mg of anti-PD-1 antibody.

[0067] In some embodiments, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the dose of 80 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody.

[0068] In some embodiments, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the dose of 160 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody.

[0069] In some embodiments, the anti-PD-1 and anti-LAG-3 antibodies or antigen-binding fragments thereof are formulated for intravenous administration.

[0070] In some embodiments, the anti-PD-1 and anti-LAG-3 antibodies or antigen-binding fragments thereof are formulated together. In some embodiments, the anti-PD-1 and anti-LAG-3 antibodies or antigen-binding fragments thereof are formulated separately.

[0071] In some embodiments, the treatment consists of up to 12 cycles.

[0072] In some embodiments, anti-PD-1 antibody or antigen-binding fragment thereof is administered on Days 1, 15, 29, and 43 of each cycle.

[0073] In some embodiments, anti-LAG-3 antibody or antigen-binding fragment thereof is administered on Days 1, 15, 29, and 43 of each cycle.

[0074] In some embodiments, the anti-PD-1 antibody or antigen-binding fragment thereof is administered prior to administration of the anti-LAG-3 antibody or antigen-binding fragment thereof. In some embodiments, the anti-LAG-3 antibody or antigen-binding fragment thereof is administered within about 30 minutes prior to administration of the anti-PD-1 antibody or antigen-binding fragment thereof. In some embodiments, the anti-PD-1 antibody or antigen-binding fragment thereof is administered after administration of the anti-LAG-3 antibody or antigen-binding fragment thereof. In some embodiments, the anti-PD-1 antibody or antigen-

binding fragment thereof is administered before administration of the anti-LAG-3 antibody or antigen-binding fragment thereof. In some embodiments, the anti-PD-1 antibody or antigen-binding fragment thereof is administered concurrently with the anti-LAG-3 antibody or antigen-binding fragment thereof.

[0075] In some embodiments, the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor are administered as a first line of treatment. In some embodiments, the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor are administered as a second line of treatment.

[0076] In some embodiments, a method disclosed herein further comprises the administration of at least one additional therapeutic agent. In some embodiments, the at least one additional therapeutic agent is a chemotherapeutic agent. In some embodiments, the at least one additional therapeutic agent is an immune checkpoint inhibitor.

[0077] In some embodiments, the method produces at least one therapeutic effect chosen from a reduction in size of a tumor, reduction in number of metastatic lesions over time, complete response, partial response, and stable disease.

[0078] In some embodiments, administering the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor activates the patient's T cells. In some embodiments, administering the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor induces the expression activation markers by the patient's T cells.

[0079] In some embodiments, administering the anti-LAG-3 antibody or antigen-binding fragment thereof results in the occupancy of at least about 10%, at least about 20%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, at least about 95%, or about 100% of the LAG-3 receptors on the patient's T cells. In some embodiments, the T cells are CD8+ T cells. In some embodiments, the T cells are tumor infiltrating T cells.

[0080] In some embodiments, the PD-1 pathway inhibitor comprises an anti-PD-1 antibody or antigen-binding fragment thereof.

[0081] Another aspect of the invention disclosed herein relates to a kit for treating a patient afflicted with a malignant tumor, the kit comprising: a dosage ranging from about 0.1 to about 10 mg/kg body weight of an anti-LAG-3 antibody or an antigen-binding fragment thereof; a dosage ranging from about 0.1 to about 10 mg/kg body weight of an anti-PD-1 antibody or an

antigen-binding fragment thereof; and instructions for using the anti-LAG-3 antibody and anti-PD-1 antibody or the antigen-binding fragments thereof in any of the methods disclosed herein.

[0082] Another aspect of the invention disclosed herein relates to a kit for treating a patient afflicted with a malignant tumor, the kit comprising: a dosage ranging from about 0.1 to about 10 mg/kg body weight of an anti-PD1 antibody or an antigen-binding fragment thereof; a dosage an immune checkpoint inhibitor; and instructions for using the anti-PD-1 antibody or antigen-binding fragment thereof and immune checkpoint inhibitor in any of the methods disclosed herein.

[0083] Another aspect of the invention disclosed herein relates to a kit for treating a patient afflicted with a malignant tumor, the kit comprising: a dosage ranging from about 0.1 to about 10 mg/kg body weight of an anti-LAG-3 antibody or an antigen-binding fragment thereof; and instructions for using the anti-LAG-3 antibody or the antigen-binding fragment thereof in any of the methods disclosed herein.

[0084] Another aspect of the invention disclosed herein relates to a kit for treating a patient afflicted with a malignant tumor, the kit comprising: a dosage ranging from 0.1 to 10 mg/kg body weight of an anti-PD-1 antibody or an antigen-binding fragment; and instructions for using the anti-PD-1 antibody or the antigen-binding fragment thereof in any of the methods disclosed herein.

[0085] An aspect of the invention relates to a method of identifying a patient that is refractory to treatment with a PD-1 antagonist, the method comprising determining the level of LAG-3 expression, wherein an increased level of LAG-3 expression following treatment with the PD-1 antagonist, relative to the level of LAG-3 expression prior to treatment with the PD-1 antagonist, indicates that a patient is refractory to PD-1 antagonist therapy. Another aspect of the invention relates to a method of identifying a patient that is at risk of becoming refractory to treatment with a PD-1 antagonist, the method comprising determining the level of LAG-3 expression, wherein an increased level of LAG-3 expression following treatment with the PD-1 antagonist, relative to the level of LAG-3 expression prior to treatment with the PD-1 antagonist, indicates that a patient is at risk of becoming refractory to PD-1 antagonist therapy. Some aspects of the invention relate to a method of identifying a patient who is likely to respond to a LAG-3 therapy, the method comprising determining the level of LAG-3 expression in the patient, wherein an increased level of LAG-3 expression following treatment with a PD-1 antagonist, relative to the level of LAG-3 expression prior to treatment with the PD-1 antagonist, indicates

that a patient is likely to respond to a LAG-3 therapy. Certain aspects of the invention relate to a method of selecting a patient for treatment with a LAG-3 therapy, the method comprising determining the level of LAG-3 expression in the patient, wherein an increased level of LAG-3 expression following treatment with a PD-1 antagonist, relative to the level of LAG-3 expression prior to treatment with the PD-1 antagonist, indicates that a patient is likely to respond to a LAG-3 therapy. In one embodiment, the PD-1 antagonist is a PD-1 inhibitor. In certain embodiments, the PD-1 antagonist is a PD-1 antibody. In some embodiments, the LAG-3 therapy is a LAG-3 inhibitor. In particular embodiments, the LAG-3 therapy is an anti-LAG-3 antibody. In one embodiment, wherein the LAG-3 therapy is a combination therapy. In an embodiment, the LAG-3 combination therapy is a combination of an anti-LAG-3 antibody and an anti-PD-1 antibody.

BRIEF DESCRIPTION OF THE FIGURES

[0086] Figure 1. Staining patterns observed in monoplex LAG-3 immunohistochemistry (IHC) samples.

[0087] Figure 2. Frequency distribution of LAG-3+ cells as a ratio of total tumor cells in a sample analyzed with monoplex LAG-3 IHC.

[0088] Figures 3A-B. (Figure 3A) Study design and endpoints. (Figure 3B) Key eligibility criteria for patients in the melanoma prior IO expansion cohort.

[0089] Figure 4. Baseline demographics and disease characteristics.

[0090] Figure 5. Prior therapy.

[0091] Figure 6. LAG-3 expression status of first 40 IO experienced melanomas.

[0092] Figure 7. Response by investigator assessment of patients with melanoma who progressed on prior anti-PD1/PD-L1 therapy.

[0093] Figure 8. LAG-3 expression enriches for response.

[0094] Figure 9. Depth and duration of response by LAG-3 expression.

[0095] Figure 10. Duration of progression-free survival.

[0096] Figure 11. Response by baseline characteristics (investigator assessed).

[0097] Figure 12. LAG-3 expression status of gastric tumor samples. 48% (10/21) of the samples were scored as LAG-3 positive using a 1% cut-off in a monoplex IHC assay.

[0098] Figure 13. Change in target lesion size in gastric cancer patients in response to treatment with a combination of anti-LAG-3 and anti-PD-1 antibody. LAG-3 positive tumors were enriched among the patients that were responsive to the treatment. Tumor response was

determined according to RECIST. The patients in this study have not been previously exposed to anti-PD-1/PD-L1 treatment.

[0099] Figure 14. LAG-3 expression status of squamous cell cancer of the head and neck (SCCHN), renal carcinoma, hepatocellular carcinoma (HCC), and NSCLC tumor samples as determined by a monoplex IHC assay.

[0100] Figure 15A and B. Figure 15A. Pigmented melanoma sections. Nuclei were counterstained with hematoxylin with or without bleaching. Figure 15B. Pigmented melanoma LAG-3 IHC with or without prior bleaching. Nuclei were counterstained with hematoxylin.

[0101] Figure 16. Updated study design and endpoints.

[0102] Figure 17. Updated baseline demographics and disease characteristics.

[0103] Figure 18. Updated prior therapies.

[0104] Figure 19. Updated antitumor activity of BMS-986016 and Nivolumab combination therapy.

[0105] Figure 20. Updated response by baseline characteristics and LAG-3 expression.

[0106] Figure 21. Updated best change in target lesion size by LAG-3 and PD-L1 expression.

[0107] Figure 22. Updated depth and duration of response by LAG-3 and PD-L1 expression.

[0108] Figure 23. Updated ongoing clinical follow-up.

[0109] Figure 24. Role of LAG-3 and PD-1 in T-cell exhaustion and proposed clinical utility of combined with nivolumab.

[0110] Figure 25. LAG-3 patterns of expression by IHC staining of total nucleated cells in a melanoma tumor specimen.

[0111] Figures 26A-F. Association of LAG-3 with immune and inflammatory biomarkers: (A) LAG-3 vs CD8, (B) LAG-3 vs FOXP3, (C) LAG-3 vs CD163, (D) LAG-3 vs CD68, (E) LAG-3 vs PD-L1, (F) LAG-3 vs MHC II.

[0112] Figure 27. Ratio of LAG-3 positive tumor infiltrating lymphocytes (TILs) in tumors comprising <1% or ≤1% MHC II positive tumor cells.

[0113] Figures 28A-C. Relationship between inflammation clusters and biomarker expression in (A) urothelial cancer, (B) NSCLC, and (C) all tumor types.

[0114] Figures 29A-C. Heterogeneous MHC II tumor cell expression and LAG-3 + TILs. (A) LAG-3 + TIL numbers in MHC II high and MHC II low tumor cell regions in urothelial

carcinoma. (B-C) Ratio of LAG-3 + TIL cells in MHC II high and MHC II low tumor cell regions in urothelial and gastric carcinoma samples.

[0115] Figures 30A and B. LAG-3 mRNA levels at screening and at week 2–4 of nivolumab monotherapy.

DETAILED DESCRIPTION OF THE INVENTION

[0116] In one aspect, the present invention relates to an improved method of treatment for malignant tumors in a human patient. In particular, the present invention shows that the administration of an anti-LAG-3 antibody in combination with an anti-PD-1 antibody achieves surprisingly improved treatment outcomes in a patient population having a LAG-3 positive malignant tumor compared to a population comprising patients having both LAG-3 positive and LAG-3 negative tumors. Accordingly, in one aspect, the invention described herein relates to a method for identifying patents having a LAG-3 positive tumor, e.g., melanoma. In another aspect, the invention described herein relates to a method of treating a LAG-3 positive malignant tumor by administering a combination of a LAG-3 inhibitor (e.g., anti-LAG-3 antibody) and a PD-1 pathway inhibitor (e.g., an anti-PD-1 antibody).

[0117] In another aspect, the invention described herein relates to a method of treating a LAG-3 positive malignant tumor by administering a PD-1 pathway inhibitor (e.g., an anti-PD-1 antibody) or a combination of a PD-1 pathway inhibitor and an immune checkpoint inhibitor.

[0118] In another aspect, the invention described herein relates to a method of treating a LAG-3 positive malignant tumor by administering an anti-CTLA4 antibody.

1. Definitions

[0119] In order that the present disclosure may be more readily understood, certain terms are first defined. As used in this application, except as otherwise expressly provided herein, each of the following terms shall have the meaning set forth below. Additional definitions are set forth throughout the application.

[0120] An "antibody" (Ab) shall include, without limitation, a glycoprotein immunoglobulin which binds specifically to an antigen and comprises at least two heavy (H) chains and two light (L) chains interconnected by disulfide bonds, or an antigen-binding portion thereof. Each H chain comprises a heavy chain variable region (abbreviated herein as V_H) and a

heavy chain constant region. The heavy chain constant region comprises three constant domains, C_{H1}, C_{H2} and C_{H3}. Each light chain comprises a light chain variable region (abbreviated herein as V_L) and a light chain constant region. The light chain constant region is comprises one constant domain, C_L. The V_H and V_L regions can be further subdivided into regions of hypervariability, termed complementarity determining regions (CDRs), interspersed with regions that are more conserved, termed framework regions (FR). Each V_H and V_L comprises three CDRs and four FRs, arranged from amino-terminus to carboxy-terminus in the following order: FR1, CDR1, FR2, CDR2, FR3, CDR3, FR4. The variable regions of the heavy and light chains contain a binding domain that interacts with an antigen. The constant regions of the antibodies may mediate the binding of the immunoglobulin to host tissues or factors, including various cells of the immune system (*e.g.*, effector cells) and the first component (C1q) of the classical complement system. A heavy chain may have the C-terminal lysine or not. Unless specified otherwise herein, the amino acids in the variable regions are numbered using the Kabat numbering system and those in the constant regions are numbered using the EU system.

[0121] An immunoglobulin may derive from any of the commonly known isotypes, including but not limited to IgA, secretory IgA, IgG and IgM. IgG subclasses are also well known to those in the art and include but are not limited to human IgG1, IgG2, IgG3 and IgG4. "Isotype" refers to the antibody class or subclass (*e.g.*, IgM or IgG1) that is encoded by the heavy chain constant region genes. The term "antibody" includes, by way of example, monoclonal and polyclonal antibodies; chimeric and humanized antibodies; human or nonhuman antibodies; wholly synthetic antibodies; and single chain antibodies. A nonhuman antibody may be humanized by recombinant methods to reduce its immunogenicity in man. Where not expressly stated, and unless the context indicates otherwise, the term "antibody" includes monospecific, bispecific, or multi-specific antibodies, as well as a single chain antibody. In embodiments, the antibody is a bispecific antibody. In other embodiments, the antibody is a monospecific antibody.

[0122] As used herein, an “IgG antibody” has the structure of a naturally occurring IgG antibody, *i.e.*, it has the same number of heavy and light chains and disulfide bonds as a naturally occurring IgG antibody of the same subclass. For example, an anti-ICOS IgG1, IgG2, IgG3 or IgG4 antibody consists of two heavy chains (HCs) and two light chains (LCs), wherein the two heavy chains and light chains are linked by the same number and location of disulfide bridges that occur

in naturally occurring IgG1, IgG2, IgG3 and IgG4 antibodies, respectively (unless the antibody has been mutated to modify the disulfide bonds)

[0123] An "isolated antibody" refers to an antibody that is substantially free of other antibodies having different antigenic specificities (*e.g.*, an isolated antibody that binds specifically to PD-1 is substantially free of antibodies that bind specifically to antigens other than PD-1). An isolated antibody that binds specifically to PD-1 may, however, have cross-reactivity to other antigens, such as PD-1 molecules from different species. Moreover, an isolated antibody may be substantially free of other cellular material and/or chemicals.

[0124] The antibody may be an antibody that has been altered (e.g., by mutation, deletion, substitution, conjugation to a non-antibody moiety). For example, an antibody may include one or more variant amino acids (compared to a naturally occurring antibody) which change a property (e.g., a functional property) of the antibody. For example, numerous such alterations are known in the art which affect, e.g., half-life, effector function, and/or immune responses to the antibody in a patient. The term antibody also includes artificial polypeptide constructs which comprise at least one antibody-derived antigen binding site.

[0125] The term "monoclonal antibody" ("mAb") refers to a non-naturally occurring preparation of antibody molecules of single molecular composition, *i.e.*, antibody molecules whose primary sequences are essentially identical, and which exhibits a single binding specificity and affinity for a particular epitope. A mAb is an example of an isolated antibody. MAbs may be produced by hybridoma, recombinant, transgenic or other techniques known to those skilled in the art.

[0126] A "human" antibody (HuMAb) refers to an antibody having variable regions in which both the framework and CDR regions are derived from human germline immunoglobulin sequences. Furthermore, if the antibody contains a constant region, the constant region is also derived from human germline immunoglobulin sequences. The human antibodies of the invention may include amino acid residues not encoded by human germline immunoglobulin sequences (*e.g.*, mutations introduced by random or site-specific mutagenesis *in vitro* or by somatic mutation *in vivo*). However, the term "human antibody," as used herein, is not intended to include antibodies in which CDR sequences derived from the germline of another mammalian species, such as a mouse, have been grafted onto human framework sequences. The terms "human" antibodies and "fully human" antibodies and are used synonymously.

[0127] A "humanized antibody" refers to an antibody in which some, most or all of the amino acids outside the CDR domains of a non-human antibody are replaced with corresponding amino acids derived from human immunoglobulins. In one embodiment of a humanized form of an antibody, some, most or all of the amino acids outside the CDR domains have been replaced with amino acids from human immunoglobulins, whereas some, most or all amino acids within one or more CDR regions are unchanged. Small additions, deletions, insertions, substitutions or modifications of amino acids are permissible as long as they do not abrogate the ability of the antibody to bind to a particular antigen. A "humanized" antibody retains an antigenic specificity similar to that of the original antibody.

[0128] A "chimeric antibody" refers to an antibody in which the variable regions are derived from one species and the constant regions are derived from another species, such as an antibody in which the variable regions are derived from a mouse antibody and the constant regions are derived from a human antibody.

[0129] An "anti-antigen" antibody refers to an antibody that binds specifically to the antigen. For example, an anti-PD-1 antibody binds specifically to PD-1 and an anti-LAG-3 antibody binds specifically to LAG-3.

[0130] An "antigen-binding portion" of an antibody (also called an "antigen-binding fragment") refers to one or more fragments of an antibody that retain the ability to bind specifically to the antigen bound by the whole antibody. It has been shown that the antigen-binding function of an antibody can be performed by fragments or portions of a full-length antibody. Examples of binding fragments encompassed within the term "antigen-binding portion" or "antigen-binding fragment" of an antibody, e.g., an anti-LAG-3 antibody described herein, include:

(1) a Fab fragment (fragment from papain cleavage) or a similar monovalent fragment consisting of the VL, VH, LC and CH1 domains;

(2) a F(ab')2 fragment (fragment from pepsin cleavage) or a similar bivalent fragment comprising two Fab fragments linked by a disulfide bridge at the hinge region;

(3) a Fd fragment consisting of the VH and CH1 domains;

(4) a Fv fragment consisting of the VL and VH domains of a single arm of an antibody,

(5) a single domain antibody (dAb) fragment (Ward et al., (1989) Nature 341:544-46), which consists of a VH domain;

(6) a bi-single domain antibody which consists of two VH domains linked by a hinge (dual-affinity re-targeting antibodies (DARTs));

(7) a dual variable domain immunoglobulin;

(8) an isolated complementarity determining region (CDR); and

(9) a combination of two or more isolated CDRs, which can optionally be joined by a synthetic linker. Furthermore, although the two domains of the Fv fragment, VL and VH, are coded for by separate genes, they can be joined, using recombinant methods, by a synthetic linker that enables them to be made as a single protein chain in which the VL and VH regions pair to form monovalent molecules (known as single chain Fv (scFv); see e.g., Bird et al. (1988) Science 242:423-426; and Huston et al. (1988) Proc. Natl. Acad. Sci. USA 85:5879-5883). Such single chain antibodies are also intended to be encompassed within the term "antigen-binding portion" or "antigen-binding fragment" of an antibody. These antibody fragments are obtained using conventional techniques known to those with skill in the art, and the fragments are screened for utility in the same manner as are intact antibodies. Antigen-binding portions can be produced by recombinant DNA techniques, or by enzymatic or chemical cleavage of intact immunoglobulins.

[0131] The term "LAG-3", "LAG3", or "Lymphocyte Activation Gene-3" refers to Lymphocyte Activation Gene-3. The term LAG-3 as used herein includes human LAG-3 (hLAG-3), variants, isoforms, and species homologs of hLAG-3, and analogs having at least one common epitope with hLAG-3. The term LAG-3 as used herein includes variants, isoforms, homologs, orthologs and paralogs. For example, antibodies specific for a human LAG-3 protein may, in certain cases, cross-react with a LAG-3 protein from a species other than human. In other embodiments, the antibodies specific for a human LAG-3 protein may be completely specific for the human LAG-3 protein and may not exhibit species or other types of cross-reactivity, or may cross-react with LAG-3 from certain other species, but not all other species (e.g., cross-react with monkey LAG-3 but not mouse LAG-3). The term "human LAG-3" refers to human sequence LAG-3, such as the complete amino acid sequence of human LAG-3 having GenBank Accession No. NP_002277 (SEQ ID NO:13). The term "mouse LAG-3" refers to mouse sequence LAG-3, such as the complete amino acid sequence of mouse LAG-3 having GenBank Accession No. NP_032505. LAG-3 is also known in the art as, for example, CD223. The human LAG-3 sequence may differ from human LAG-3 of GenBank Accession No. NP_002277 by having, e.g.,

conserved mutations or mutations in non-conserved regions and the LAG-3 has substantially the same biological function as the human LAG-3 of GenBank Accession No. NP_002277. For example, a biological function of human LAG-3 is having an epitope in the extracellular domain of LAG-3 that is specifically bound by an antibody of the instant disclosure or a biological function of human LAG-3 is binding to MHC Class II molecules.

[0132] A particular human LAG-3 sequence will generally be at least 90% identical in amino acid sequence to human LAG-3 of GenBank Accession No. NP_002277 and contains amino acid residues that identify the amino acid sequence as being human when compared to LAG-3 amino acid sequences of other species (e.g., murine). In certain cases, a human LAG-3 can be at least 95%, or even at least 96%, 97%, 98%, or 99% identical in amino acid sequence to LAG-3 of GenBank Accession No. NP_002277. In certain embodiments, a human LAG-3 sequence will display no more than 10 amino acid differences from the LAG-3 sequence of GenBank Accession No. NP_002277. In certain embodiments, the human LAG-3 can display no more than 5, or even no more than 4, 3, 2, or 1 amino acid difference from the LAG-3 sequence of GenBank Accession No. NP_002277. Percent identity can be determined as described herein.

[0133] As used herein, the terms "Programmed Death 1," "Programmed Cell Death 1," "Protein PD-1," "PD-1," "PD1," "PDCD1," "hPD-1" and "hPD-I" are used interchangeably, and include variants, isoforms, species homologs of human PD-1, and analogs having at least one common epitope with PD-1. The complete PD-1 sequence can be found under GenBank Accession No. U64863 (SEQ ID NO:29).

[0134] The protein Programmed Death 1 (PD-1) is an inhibitory member of the CD28 family of receptors, that also includes CD28, CTLA-4, ICOS and BTLA. PD-1 is expressed on activated B cells, T cells, and myeloid cells (Agata et al., supra; Okazaki et al. (2002) Curr. Opin. Immunol. 14: 391779-82; Bennett et al. (2003) J Immunol 170:711-8). The initial members of the family, CD28 and ICOS, were discovered by functional effects on augmenting T cell proliferation following the addition of monoclonal antibodies (Hutloff et al. Nature (1999); 397:263-266; Hansen et al. Immunogenics (1980); 10:247-260). PD-1 was discovered through screening for differential expression in apoptotic cells (Ishida et al. EMBO J (1992); 11:3887-95). The other members of the family, CTLA-4 and BTLA, were discovered through screening for differential expression in cytotoxic T lymphocytes and TH1 cells, respectively. CD28, ICOS and CTLA-4 all have an unpaired cysteine residue allowing for homodimerization. In contrast,

PD-1 is suggested to exist as a monomer, lacking the unpaired cysteine residue characteristic in other CD28 family members.

[0135] The PD-1 gene is a 55 kDa type I transmembrane protein that is part of the Ig gene superfamily (Agata et al. (1996) Int Immunol 8:765-72). PD-1 contains a membrane proximal immunoreceptor tyrosine inhibitory motif (ITIM) and a membrane distal tyrosine-based switch motif (ITSM) (Thomas, M. L. (1995) J Exp Med 181:1953-6; Vivier, E and Daeron, M (1997) Immunol Today 18:286-91). Although structurally similar to CTLA-4, PD-1 lacks the MYPPPY motif (SEQ ID NO: 32) that is critical for B7-1 and B7-2 binding. Two ligands for PD-1 have been identified, PD-L1 and PD-L2, that have been shown to downregulate T cell activation upon binding to PD-1 (Freeman et al. (2000) J Exp Med 192:1027-34; Latchman et al. (2001) Nat Immunol 2:261-8; Carter et al. (2002) Eur J Immunol 32:634-43). Both PD-L1 and PD-L2 are B7 homologs that bind to PD-1, but do not bind to other CD28 family members. PD-L1 is abundant in a variety of human cancers (Dong et al. (2002) Nat. Med. 8:787-9). The interaction between PD-1 and PD-L1 results in a decrease in tumor infiltrating lymphocytes, a decrease in T-cell receptor mediated proliferation, and immune evasion by the cancerous cells (Dong et al. (2003) J. Mol. Med. 81:281-7; Blank et al. (2005) Cancer Immunol. Immunother. 54:307-314; Konishi et al. (2004) Clin. Cancer Res. 10:5094-100). Immune suppression can be reversed by inhibiting the local interaction of PD-1 with PD-L1, and the effect is additive when the interaction of PD-1 with PD-L2 is blocked as well (Iwai et al. (2002) Proc. Nat'l. Acad. Sci. USA 99:12293-7; Brown et al. (2003) J. Immunol. 170:1257-66).

[0136] Consistent with PD-1 being an inhibitory member of the CD28 family, PD-1 deficient animals develop various autoimmune phenotypes, including autoimmune cardiomyopathy and a lupus-like syndrome with arthritis and nephritis (Nishimura et al. (1999) Immunity 11:141-51; Nishimura et al. (2001) Science 291:319-22). Additionally, PD-1 has been found to play a role in autoimmune encephalomyelitis, systemic lupus erythematosus, graft-versus-host disease (GVHD), type I diabetes, and rheumatoid arthritis (Salama et al. (2003) J Exp Med 198:71-78; Prokunina and Alarcon-Riquelme (2004) Hum Mol Genet 13:R143; Nielsen et al. (2004) Lupus 13:510). In a murine B cell tumor line, the ITSM of PD-1 was shown to be essential to block BCR-mediated Ca.sup.2+-flux and tyrosine phosphorylation of downstream effector molecules (Okazaki et al. (2001) PNAS 98:13866-71).

[0137] "Programmed Death Ligand-1 (PD-L1)" is one of two cell surface glycoprotein ligands for PD-1 (the other being PD-L2) that downregulate T cell activation and cytokine

secretion upon binding to PD-1. The term "PD-L1" as used herein includes human PD-L1 (hPD-L1), variants, isoforms, and species homologs of hPD-L1, and 5 analogs having at least one common epitope with hPD-L1. The complete hPD-L1 sequence can be found under GenBank Accession No. Q9NZQ7.

[0138] The terms "Programmed Death Ligand-2" and "PD-L2" as used herein include human PD-L2 (hPD-L2), variants, isoforms, and species homologs of hPD-L2, and analogs having at least one common epitope with hPD-L2. The complete hPD-L2 sequence can be found under GenBank Accession No. Q9BQ51.

[0139] A "patient" as used herein includes any patient who is afflicted with a cancer (e.g., melanoma). The terms "subject" and "patient" are used interchangeably herein.

[0140] "Administering" refers to the physical introduction of a composition comprising a therapeutic agent to a subject, using any of the various methods and delivery systems known to those skilled in the art. Routes of administration for the formulations disclosed herein include intravenous, intramuscular, subcutaneous, intraperitoneal, spinal or other parenteral routes of administration, for example by injection or infusion. The phrase "parenteral administration" as used herein means modes of administration other than enteral and topical administration, usually by injection, and includes, without limitation, intravenous, intramuscular, intraarterial, intrathecal, intralymphatic, intralesional, intracapsular, intraorbital, intracardiac, intradermal, intraperitoneal, transtracheal, subcutaneous, subcuticular, intraarticular, subcapsular, subarachnoid, intraspinal, epidural and intrasternal injection and infusion, as well as in vivo electroporation. In some embodiments, the formulation is administered via a non-parenteral route, in some embodiments, orally. Other non-parenteral routes include a topical, epidermal or mucosal route of administration, for example, intranasally, vaginally, rectally, sublingually or topically. Administering can also be performed, for example, once, a plurality of times, and/or over one or more extended periods.

[0141] "Treatment" or "therapy" of a subject refers to any type of intervention or process performed on, or the administration of an active agent to, the subject with the objective of reversing, alleviating, ameliorating, inhibiting, slowing down or preventing the onset, progression, development, severity or recurrence of a symptom, complication or condition, or biochemical indicia associated with a disease.

[0142] As used herein, "effective treatment" refers to treatment producing a beneficial effect, e.g., amelioration of at least one symptom of a disease or disorder. A beneficial effect can

take the form of an improvement over baseline, i.e., an improvement over a measurement or observation made prior to initiation of therapy according to the method. A beneficial effect can also take the form of arresting, slowing, retarding, or stabilizing of a deleterious progression of a marker of solid tumor. Effective treatment may refer to alleviation of at least one symptom of a solid tumor. Such effective treatment may, e.g., reduce patient pain, reduce the size and/or number of lesions, may reduce or prevent metastasis of a tumor, and/or may slow tumor growth.

[0143] The term "effective amount" refers to an amount of an agent that provides the desired biological, therapeutic, and/or prophylactic result. That result can be reduction, amelioration, palliation, lessening, delaying, and/or alleviation of one or more of the signs, symptoms, or causes of a disease, or any other desired alteration of a biological system. In reference to solid tumors, an effective amount comprises an amount sufficient to cause a tumor to shrink and/or to decrease the growth rate of the tumor (such as to suppress tumor growth) or to prevent or delay other unwanted cell proliferation. In some embodiments, an effective amount is an amount sufficient to delay tumor development. In some embodiments, an effective amount is an amount sufficient to prevent or delay tumor recurrence. An effective amount can be administered in one or more administrations. The effective amount of the drug or composition may: (i) reduce the number of cancer cells; (ii) reduce tumor size; (iii) inhibit, retard, slow to some extent and may stop cancer cell infiltration into peripheral organs; (iv) inhibit (i.e., slow to some extent and may stop tumor metastasis; (v) inhibit tumor growth; (vi) prevent or delay occurrence and/or recurrence of tumor; and/or (vii) relieve to some extent one or more of the symptoms associated with the cancer. In one example, an "effective amount" is the amount of anti-LAG-3 antibody and the amount of anti-PD-1 antibody, in combination, clinically proven to affect a significant decrease in cancer or slowing of progression of cancer, such as an advanced solid tumor. As used herein, the terms "fixed dose", "flat dose" and "flat-fixed dose" are used interchangeably and refer to a dose that is administered to a patient without regard for the weight or body surface area (BSA) of the patient. The fixed or flat dose is therefore not provided as a mg/kg dose, but rather as an absolute amount of the agent (e.g., the anti-LAG-3 antibody and/or anti-PD-1 antibody).

[0144] The term "progression-free survival," which can be abbreviated as PFS, as used herein refers to the length of time during and after the treatment of a solid tumor (*i.e.*, melanoma) that a patient lives with the disease but it does not get worse.

[0145] "Dosing interval," as used herein, means the amount of time that elapses between multiple doses of a formulation disclosed herein being administered to a subject. Dosing interval can thus be indicated as ranges.

[0146] The term "dosing frequency" as used herein refers to the frequency of administering doses of a formulation disclosed herein in a given time. Dosing frequency can be indicated as the number of doses per a given time, *e.g.*, once a week or once in two weeks.

[0147] The use of the term "fixed dose" with regard to a composition of the invention means that two or more different antibodies in a single composition are present in the composition in particular (fixed) ratios with each other. In some embodiments, the fixed dose is based on the weight (*e.g.*, mg) of the antibodies. In certain embodiments, the fixed dose is based on the concentration (*e.g.*, mg/ml) of the antibodies. In some embodiments, the ratio is at least about 1:1, about 1:2, about 1:3, about 1:4, about 1:5, about 1:6, about 1:7, about 1:8, about 1:9, about 1:10, about 1:15, about 1:20, about 1:30, about 1:40, about 1:50, about 1:60, about 1:70, about 1:80, about 1:90, about 1:100, about 1:120, about 1:140, about 1:160, about 1:180, about 1:200, about 200:1, about 180:1, about 160:1, about 140:1, about 120:1, about 100:1, about 90:1, about 80:1, about 70:1, about 60:1, about 50:1, about 40:1, about 30:1, about 20:1, about 15:1, about 10:1, about 9:1, about 8:1, about 7:1, about 6:1, about 5:1, about 4:1, about 3:1, or about 2:1 mg first antibody to mg second antibody. For example, the 3:1 ratio of a first antibody and a second antibody can mean that a vial can contain about 240 mg of the first antibody and 80 mg of the second antibody or about 3 mg/ml of the first antibody and 1 mg/ml of the second antibody.

[0148] The use of the term "flat dose" with regard to the composition of the invention means a dose that is administered to a patient without regard for the weight or body surface area (BSA) of the patient. The flat dose is therefore not provided as a mg/kg dose, but rather as an absolute amount of the agent (*e.g.*, the anti-LAG-3 antibody and/or anti-PD-1 antibody). For example, a 60 kg person and a 100 kg person would receive the same dose of the composition (*e.g.*, 240 mg of an anti-PD-1 antibody and 80 mg of an anti- LAG-3 antibody in a single fixed dosing formulation vial containing both 240 mg of an anti-PD-1 antibody and 80 mg of an anti-LAG-3 antibody (or two fixed dosing formulation vials containing 120 mg of an anti-PD-1 antibody and 40 mg of an anti- LAG-3 antibody, etc.)).

[0149] The term "weight based dose" as referred to herein means that a dose that is administered to a patient is calculated based on the weight of the patient. For example, when a patient with 60 kg body weight requires 3 mg/kg of an anti-LAG-3 antibody in combination with

3 mg/kg of an anti-PD-1 antibody, one can draw the appropriate amounts of the anti-LAG-3 antibody (*i.e.*, 180 mg) and the anti-PD-1 antibody (*i.e.*, 180 mg) at once from a 1:1 ratio fixed dosing formulation of an anti-LAG3 antibody and an anti-PD-1 antibody.

[0150] The terms "about once a week," "once about every week," "once about every two weeks," or any other similar dosing interval terms as used herein means approximate number, and "about once a week" or "once about every week" can include every seven days ± two days, *i.e.,* every five days to every nine days. The dosing frequency of "once a week" thus can be every five days, every six days, every seven days, every eight days, or every nine days. "Once about every two weeks" can include every fourteen days ± three days, *i.e.*, every eleven days to every seventeen days. Similar approximations apply, for example, to once about every three weeks, once about every four weeks, once about every five weeks, once about every six weeks and once about every twelve weeks. In some embodiments, a dosing interval of once about every six weeks or once about every twelve weeks means that the first dose can be administered any day in the first week, and then the next dose can be administered any day in the sixth or twelfth week, respectively. In other embodiments, a dosing interval of once about every six weeks or once about every twelve weeks means that the first dose is administered on a particular day of the first week (e.g., Monday) and then the next dose is administered on the same day of the sixth or twelfth weeks (i.e., Monday), respectively.

[0151] A "cancer" refers a broad group of various diseases characterized by the uncontrolled growth of abnormal cells in the body. Unregulated cell division and growth results in the formation of malignant tumors that invade neighboring tissues and may also metastasize to distant parts of the body through the lymphatic system or bloodstream. A "cancer" or "cancer tissue" can include a tumor.

[0152] The term "tumor" as used herein refers to any mass of tissue that results from excessive cell growth or proliferation, either benign (non-cancerous) or malignant (cancerous), including pre-cancerous lesions.

[0153] The term "LAG-3 positive" or "LAG-3 expression positive," relating to LAG-3 expression, refers to the proportion of cells in a test tissue sample comprising tumor cells and tumor-infiltrating inflammatory cells above which the tissue sample is scored as expressing LAG-3. In some embodiments, for LAG-3 expression assayed by immunohistochemistry (IHC), the LAG-3 positive tumor or LAG-3 expression positive tumor means that at least about 0.01%, at least about 0.5%, at least about 1%, at least about 2%, at least about 3%, at least about 4%, at

least about 5%, at least about 6%, at least about 7%, at least about 8%, at least about 9%, at least about 10%, at least about 15%, at least about 20%, at least about 25%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, or 100% of the total number of cells express LAG-3. In other embodiments, for LAG-3 expression assayed by immunohistochemistry (IHC) or flow cytometry, the LAG-3 positive tumor or LAG-3 expression positive tumor means that at least about 0.01%, at least about 0.5%, at least about 1%, at least about 2%, at least about 3%, at least about 4%, at least about 5%, at least about 6%, at least about 7%, at least about 8%, at least about 9%, at least about 10%, at least about 15%, at least about 20%, at least about 25%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, or 100% of the total number of tumor-infiltrating inflammatory cells (e.g., T cells, CD8+ T cells, CD4+ T cells, FOXP3+ cells) express LAG-3. LAG-3 positive tumor or LAG-3 expression positive tumor can also be expressed herein as tumor expressing LAG-3. In some embodiments, the LAG-3 positive tumor or LAG-3 expression positive tumor means that at least about 0.1% to at least about 20% of the total number of cells express LAG-3. In some embodiments, a LAG-3 positive tumor or LAG-3 expression positive tumor means that at least about 0.1% to at least about 20% of the total number of tumor-infiltrating inflammatory cells (e.g., T cells, CD8+ T cells, CD4+ T cells, FOXP3+ cells) express LAG-3. In certain embodiments, a LAG-3 positive tumor or LAG-3 expression positive tumor means that at least about 0.1% to at least about 10% of the total number of cells express LAG-3. In certain embodiments, a LAG-3 positive tumor or LAG-3 expression positive tumor means that at least about 0.1% to at least about 10% of the total number of tumor-infiltrating inflammatory cells (e.g., T cells, CD8+ T cells, CD4+ T cells, FOXP3+ cells) express LAG-3. In some embodiments, a LAG-3 positive or LAG-3 expression positive tumor means that at least about 1% of the total number of cells express LAG-3 on the cell surface. In some embodiments, a LAG-3 positive or LAG-3 expression positive tumor means that at least about 1% of the total number of tumor-infiltrating inflammatory cells (e.g., T cells, CD8+ T cells, CD4+ T cells, FOXP3+ cells) express LAG-3 on the cell surface. In other embodiments, a LAG-3 positive or LAG-3 expression positive tumor means that at least about 5% of the total number of cells express LAG-3 on the cell surface. In other embodiments, a LAG-3 positive or LAG-3 expression positive tumor means that at least about 5% of the total number of tumor-infiltrating inflammatory cells (e.g., T cells, CD8+ T cells, CD4+ T cells, FOXP3+ cells) express LAG-3 on the cell surface. In one particular embodiment, LAG-3

positive or LAG-3 expression positive tumor means that at least about 1%, or in the range of 1-5% of the total number of cells express LAG-3 on the cell surface. In one particular embodiment, LAG-3 positive or LAG-3 expression positive tumor means that at least about 1%, or in the range of 1- 5% of the total number of tumor-infiltrating inflammatory cells (e.g., T cells, CD8+ T cells, CD4+ T cells, FOXP3+ cells) express LAG-3 on the cell surface.

[0154] "LAG-3 negative" or "LAG-3 expression negative," refers to the lack of a detectable amount of LAG-3 expression. In some embodiments, for LAG-3 expression assayed by IHC, a LAG-3 negative tumor or LAG-3 expression negative tumor means that less than 0.01% of the total number of cells express a detectable level of LAG-3. In some embodiments, for LAG-3 expression assayed by IHC or flow cytometry, a LAG-3 negative tumor or LAG-3 expression negative tumor means that less than 0.01% of the total number of tumor-infiltrating inflammatory cells (e.g., T cells, CD8+ T cells, CD4+ T cells, FOXP3+ cells) express a detectable level of LAG-3. In some embodiments, for LAG-3 expression assayed by IHC, a LAG-3 negative tumor or LAG-3 expression negative tumor means that less than 1% of the total number of cells express a detectable level of LAG-3. In some embodiments, for LAG-3 expression assayed by IHC or flow cytometry, a LAG-3 negative tumor or LAG-3 expression negative tumor means that less than 1% of the total number of tumor-infiltrating inflammatory cells (e.g., T cells, CD8+ T cells, CD4+ T cells, FOXP3+ cells) express a detectable level of LAG-3. In some embodiments, a LAG-3 negative tumor or LAG-3 expression negative tumor means that zero (0) cells express a detectable level of LAG-3. In some embodiments, a LAG-3 negative or a LAG-3 expression negative tumor is any tumor other than a LAG-3 positive or a LAG-3 expression positive tumor.

[0155] The term "PD-L1 positive" or "PD-L1 expression positive," relating to cell surface PD-L1 expression, refers to the proportion of cells in a test tissue sample comprising tumor cells and tumor- infiltrating inflammatory cells above which the sample is scored as expressing cell surface PD-L1. For cell surface expression assayed by immunohistochemistry (IHC), e.g., with the mAb 28- 8, the PD-L1 positive tumor or PD-L1 expression positive tumor means that at least about 0.01%, at least about 0.5%, at least about 1%, at least about 2%, at least about 3%, at least about 4%, at least about 5%, at least about 6%, at least about 7%, at least about 8%, at least about 9%, at least about 10%, at least about 15%, at least about 20%, at least about 25%, or at least about 30% of the total number of cells express PD-L1. PD-L1 positive tumor or PD-L1 expression positive tumor can also be expressed herein as tumor expressing PD-L1. In other

embodiments, the PD-L1 positive tumor or PD-L1 expression positive tumor means that at least about 0.1% to at least about 20% of the total number of cells express PD-L1. In certain embodiments, the PD-L1 positive tumor or PD-L1 expression positive tumor means that at least about 0.1% to at least about 10% of the total number of cells express PD-L1. In some embodiments, the PD-L1 positive or PD-L1 expression positive tumor means that at least about 1% of the total number of cells express PD-L1 on the cell surface. In other embodiments, the PD-L1 positive or PD-L1 expression positive tumor means that at least about 5% of the total number of cells express PD-L1 on the cell surface. In one particular embodiment, PD-L1 positive or PD-L1 expression positive tumor means that at least about 1%, or in the range of 1- 5% of the total number of cells express PD-L1 on the cell surface.

[0156] The term "PD-L1 negative" or "PD-L1 expression negative," relating to cell surface PD-L1 expression, refers to the proportion of cells in a test tissue sample comprising tumor cells and tumor- infiltrating inflammatory cells that are not PD-L1 positive or PD-L1 expression positive.

[0157] An "immune response" refers to the action of a cell of the immune system (for example, T lymphocytes, B lymphocytes, natural killer (NK) cells, macrophages, eosinophils, mast cells, dendritic cells and neutrophils) and soluble macromolecules produced by any of these cells or the liver (including antibodies, cytokines, and complement) that results in selective targeting, binding to, damage to, destruction of, and/or elimination from a vertebrate's body of invading pathogens, cells or tissues infected with pathogens, cancerous or other abnormal cells, or, in cases of autoimmunity or pathological inflammation, normal human cells or tissues.

[0158] A "tumor-infiltrating inflammatory cell" is any type of cell that typically participates in an inflammatory response in a subject and which infiltrates tumor tissue. Such cells include tumor-infiltrating lymphocytes (TILs), macrophages, monocytes, eosinophils, histiocytes and dendritic cells.

[0159] The term "and/or" where used herein is to be taken as specific disclosure of each of the two specified features or components with or without the other. Thus, the term "and/or" as used in a phrase such as "A and/or B" herein is intended to include "A and B," "A or B," "A" (alone), and "B" (alone). Likewise, the term "and/or" as used in a phrase such as "A, B, and/or C" is intended to encompass each of the following aspects: A, B, and C; A, B, or C; A or C; A or B; B or C; A and C; A and B; B and C; A (alone); B (alone); and C (alone).

[0160] It is understood that wherever aspects are described herein with the language "comprising," otherwise analogous aspects described in terms of "consisting of" and/or "consisting essentially of" are also provided.

[0161] The terms "about" or "comprising essentially of" refer to a value or composition that is within an acceptable error range for the particular value or composition as determined by one of ordinary skill in the art, which will depend in part on how the value or composition is measured or determined, *i.e.*, the limitations of the measurement system. For example, "about" or "comprising essentially of" can mean within 1 or more than 1 standard deviation per the practice in the art. Alternatively, "about" or "comprising essentially of" can mean a range of up to 10% or 20% (*i.e.*, ±10% or ±20%). For example, about 3mg can include any number between 2.7 mg and 3.3 mg (for 10%) or between 2.4 mg and 3.6 mg (for 20%). Furthermore, particularly with respect to biological systems or processes, the terms can mean up to an order of magnitude or up to 5-fold of a value. When particular values or compositions are provided in the application and claims, unless otherwise stated, the meaning of "about" or "comprising essentially of" should be assumed to be within an acceptable error range for that particular value or composition.

[0162] As described herein, any concentration range, percentage range, ratio range or integer range is to be understood to include the value of any integer within the recited range and, when appropriate, fractions thereof (such as one-tenth and one-hundredth of an integer), unless otherwise indicated.

[0163] Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure is related. For example, the Concise Dictionary of Biomedicine and Molecular Biology, Juo, Pei-Show, 2nd ed., 2002, CRC Press; The Dictionary of Cell and Molecular Biology, 5th ed., 2013, Academic Press; and the Oxford Dictionary Of Biochemistry And Molecular Biology, 2006, Oxford University Press, provide one of skill with a general dictionary of many of the terms used in this disclosure.

[0164] Units, prefixes, and symbols are denoted in their Système International de Unites (SI) accepted form. Numeric ranges are inclusive of the numbers defining the range. The headings provided herein are not limitations of the various aspects of the disclosure, which can be had by reference to the specification as a whole. Accordingly, the terms defined immediately below are more fully defined by reference to the specification in its entirety.

[0165] Various aspects of the invention are described in further detail in the following subsections.

2. Methods of the Invention

[0166] In one aspect, the present invention is directed to a method for treating a LAG-3-positive malignant tumor (e.g., melanoma) in a subject in need thereof. A combination therapy of a LAG-3 inhibitor (e.g., anti-LAG-3 antibody) and a PD-1 pathway inhibitor (e.g., anti-PD-1 antibody) results in better therapeutic outcomes (e.g., objective response rate and disease control rate) in a patient population with LAG-3 positive malignant tumors (e.g., melanoma) than in a general patient population having a mix of LAG-3-negative malignant tumors and LAG-3-positive malignant tumors. In order to improve the treatment of malignant tumors, in one aspect, the present invention provides identifying a patient as having a LAG-3-positive tumor and providing an immunotherapy of a LAG-3 inhibitor (e.g., anti-LAG-3 antibody) and a PD-1 pathway inhibitor (e.g., anti-PD-1 antibody).

[0167] In another aspect, the present invention is directed to identifying a patient as having a LAG-3-positive tumor and treating the LAG-3 positive tumor by administering a PD-1 pathway inhibitor (e.g., an anti-PD-1 antibody) or a combination of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In one embodiment, the invention includes a method of identifying a patient as having a LAG-3-positive tumor and treating the LAG-3 positive tumor by administering an anti-PD-1 antibody. In one embodiment, the invention includes a method of identifying a patient as having a LAG-3-positive tumor and treating the LAG-3 positive tumor by administering an anti-PD-L1 antibody.

[0168] In another aspect, the present invention is directed to identifying a patient as having a LAG-3-positive tumor and treating the LAG-3 positive tumor by administering an anti-CTLA-4 antibody.

[0169] In one embodiment, the invention includes a method of selecting a malignant tumor in a human patient for immunotherapy, comprising: (a) determining the level of LAG-3 expression in a tumor sample; and (b) selecting the tumor for immunotherapy if the tumor is a LAG-3 positive tumor. In one embodiment, the invention includes a method of identifying a malignant tumor in a human patient as eligible for immunotherapy, comprising: (a) determining the level of LAG-3 expression in a tumor sample; and (b) identifying the tumor as eligible for

immunotherapy if the tumor is a LAG-3 positive tumor. In one embodiment, the invention includes a method of identifying a malignant tumor in a human patient that is likely to be responsive to a immunotherapy, the method comprising: (a) determining the level of LAG-3 expression in a tumor sample; and (b) identifying the tumor as likely to be responsive to treatment if the tumor is a LAG-3 positive tumor. In one embodiment, the invention includes a method of identifying a malignant tumor in a human patient that is likely to be responsive to a immunotherapy, the method comprising: (a) determining the level of LAG-3 expression in a tumor sample; and (b) identifying the tumor as likely to be responsive to treatment if the tumor is a LAG-3 positive tumor. In one embodiment, the invention includes a method of classifying a malignant tumor in a human patient as likely to be responsive to a immunotherapy, the method comprising: (a) determining the level of LAG-3 expression in a tumor sample; and (b) classifying the tumor as likely to be responsive to immunotherapy if the tumor is a LAG-3 positive tumor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of an anti-PD-1 antibody. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of an anti-PD-L1 antibody. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of an anti-PD-1 antibody. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of an anti-PD-L1 antibody. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of an anti-CTLA-4 antibody. In

some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. In certain embodiments, any of the present methods further comprise determining PD-L1 expression in the tumor sample.

[0170] In one embodiment, the invention includes a method of identifying a patient with a malignant tumor who is likely to respond to a immunotherapy, the method comprising: (a) determining the level of LAG-3 expression in a tumor sample; and (b) identifying the patient who is likely to respond to treatment if the tumor is a LAG-3 positive tumor. In one embodiment, the invention includes a method of selecting a patient with a malignant tumor for immunotherapy, the method comprising: (a) determining the level of LAG-3 expression in a tumor sample; and (b) selecting the patient for immunotherapy if the tumor is a LAG-3 positive tumor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of an anti-PD-1 antibody. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of an anti-PD-L1 antibody. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the method comprises

contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of an anti-PD-1 antibody. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of an anti-PD-L1 antibody. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the method comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the method comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. In certain embodiments, any of the present methods further comprise determining PD-L1 expression in the tumor sample.

[0171] In one embodiment, the invention includes a method of treating a malignant tumor in a human patient, comprising: administering to the patient an immunotherapy disclosed herein; wherein the patient is predicted to respond to treatment with the LAG-3 inhibitor and PD-1 pathway inhibitor based upon LAG-3 expression or based upon LAG-3 and PD-L1 expression in a sample of the patient's tumor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody.

[0172] In one embodiment, the invention includes a method of treating a malignant tumor in a human patient in need thereof, comprising: (a) determining the level of LAG-3 expression or the level of LAG-3 and PD-L1 expression in a tumor sample; and (b) administering to the patient a therapeutically effective amount of a LAG-3 inhibitor if the tumor is a LAG-3 positive tumor or

a LAG-3 positive PD-L1 positive tumor. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof, comprising: (a) identifying the patient as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor; and (b) administering to the patient a therapeutically effective amount of a LAG-3 inhibitor. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the LAG-3 positive tumor is a LAG-3 positive PD-L1 negative tumor. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody.

[0173] In one embodiment, the invention includes a method of treating a malignant tumor in a human patient in need thereof, comprising: (a) determining the level of LAG-3 expression or the level of LAG-3 and PD-L1 expression in a tumor sample; and (b) administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor if the tumor is a LAG-3 positive tumor or a LAG-3 positive PD-L1 positive tumor. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof, comprising: (a) identifying the patient as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor; and (b) administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the PD-1 pathway inhibitor is an anti-PD-1 antibody. In some embodiments, the PD-1 pathway inhibitor is an anti-PD-L1 antibody. In some embodiments, the LAG-3 positive tumor is a LAG-3 positive PD-L1 negative tumor. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor.

[0174] In one embodiment, the invention includes a method of treating a malignant tumor in a human patient in need thereof, comprising: (a) determining the level of LAG-3 expression or the level of LAG-3 and PD-L1 expression in a tumor sample; and (b) administering to the patient

a therapeutically effective amount of an anti-CTLA-4 antibody if the tumor is a LAG-3 positive tumor or a LAG-3 positive PD-L1 positive tumor. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof, comprising: (a) identifying the patient as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor; and (b) administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody, wherein the patient is identified as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the LAG-3 positive tumor is a LAG-3 positive PD-L1 negative tumor. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor.

[0175] In one embodiment, the invention includes a method of treating a malignant tumor in a human patient in need thereof, comprising: (a) determining the level of LAG-3 expression or the level of LAG-3 and PD-L1 expression in a tumor sample; and (b) administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and PD-1 pathway inhibitor if the tumor is a LAG-3 positive tumor or a LAG-3 positive PD-L1 positive tumor. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof, comprising: (a) identifying the patient as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor; and (b) administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the LAG-3 positive tumor is a LAG-3 positive PD-L1 negative tumor. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-L1 antibody. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody.

[0176] In one embodiment, the invention includes a method of treating a malignant tumor in a human patient in need thereof, comprising: (a) determining the level of LAG-3 expression or the level of LAG-3 and PD-L1 expression in a tumor sample; and (b) administering to the patient a therapeutically effective amount of a PD1 pathway inhibitor and an immune checkpoint inhibitor if the tumor is a LAG-3 positive tumor or a LAG-3 positive PD-L1 positive tumor. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof, comprising: (a) identifying the patient as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor; and (b) administering to the patient a therapeutically effective amount of a PD1 pathway inhibitor and an immune checkpoint inhibitor. In one embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a PD1 pathway inhibitor and an immune checkpoint inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the LAG-3 positive tumor is a LAG-3 positive PD-L1 negative tumor. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the PD-1 pathway inhibitor is an anti-PD-L1 antibody. In some embodiments, the PD-1 pathway inhibitor is an anti-PD-1 antibody.

[0177] In another embodiment, the invention includes a method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient an immunotherapy disclosed herein, wherein the patient is identified as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1

pathway inhibitor is an anti-PD-L1 antibody. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody.

[0178] In certain embodiments, the invention includes method for extending a progression-free survival period for over 12 months in a human patient afflicted with a malignant tumor comprising administering to the patient an immunotherapy disclosed herein, wherein the patient is identified as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the progression-free survival of the patient can be extended, after the administration, for over about 13 months, about 14 months, about 15 months, about 16 months, about 17 months, about 18 months, about 2 years, about 3 years, about 4 years, about 5 years, about 6 years, about 7 years, about 8 years, about 9 years, or about 10 years. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-L1 antibody.

[0179] In still other embodiments, the invention is includes a method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient an immunotherapy disclosed herein, wherein the patient is identified as having a LAG-3 positive malignant tumor (e.g., melanoma) or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration. In some embodiments, the method comprises identifying the patient as having a LAG-3 positive

malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-PD-1 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-PD-L1 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-L1 antibody.

[0180] The invention can also include a method of preventing a relapse and/or inducing a remission in a patient comprising administering to the patient an immunotherapy disclosed herein, wherein the patient is identified as having a LAG-3-positive malignant tumor (e.g., melanoma) or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the method of the invention comprises (i) identifying a patient as having a LAG-3-positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor; (ii) administering to the patient an immunotherapy disclosed herein. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-PD-1 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-PD-L1 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically

effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-L1 antibody.

[0181] In certain embodiments, the invention includes a method for increasing an objective response rate to be higher than 55% in a patient population, wherein each patient of the patient population is afflicted with a malignant tumor, in a cancer treatment comprising administering to the patient an immunotherapy disclosed herein, wherein each patient is identified as having a LAG-3 positive malignant tumor (e.g., melanoma) or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 55%, 60%, 65%, 70%, or 75%. In some embodiments, the method comprises identifying the patient as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and an anti-PD-1 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and an anti-PD-L1 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-L1 antibody.

[0182] In certain embodiments, the invention includes a method for increasing a disease control rate to be higher than 55% in a patient population, wherein each patient of the patient

population is afflicted with a malignant tumor, in a cancer treatment comprising administering to the patient an immunotherapy disclosed herein, wherein each patient is identified as having a LAG-3 positive malignant tumor (e.g., melanoma) or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration and wherein the disease control rate is higher than 55%, 60%, 65%, 70%, or 75%. In some embodiments, the method comprises identifying the patient as having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor prior to the administration. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-PD-1 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-PD-L1 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-L1 antibody.

[0183] In other embodiments, each patient in the methods experiences (i) extended progression-free survival for over 12 months, (ii) tumor size reduction at least about 10%, about 20%, about 30%, about 40%, or about 50% compared to the tumor size prior to the administration, or (iii) both. In some embodiments, the patient population can be at least 100 patients having a LAG-3 positive malignant tumor (e.g., melanoma) or a LAG-3 positive PD-L1 positive malignant tumor. In some embodiments, the patient population can be at least 200, 300, 400, 500, 600, 700, 800, 900, or 1000 patients having a LAG-3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor.

[0184] In further embodiments, the invention provides a method for selecting a human patient suitable for a combination therapy comprising: (a) identifying a patient as having a LAG-

3 positive malignant tumor or a LAG-3 positive PD-L1 positive malignant tumor; and (b) instructing a healthcare provider to administer to the patient an immunotherapy disclosed herein. In some embodiments, the LAG-3 positive malignant tumor is a LAG-3 positive PD-L1 negative malignant tumor. The method can further comprise administering an immunotherapy disclosed herein. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering to the patient a therapeutically effective amount of an anti-PD-1 antibody. In some embodiments, the immunotherapy comprises administering to the patient a therapeutically effective amount of an anti-PD-L1 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-L1 antibody. In some embodiments, the administration treats the malignant tumor.

[0185] The methods of the invention, as a result of the administration of an immunotherapy disclosed herein, can treat the malignant tumor, reduce the tumor size, prevent growth of the tumor, eliminate the tumor from the patient, prevent a relapse of a tumor, induce a remission in a patient, or any combination thereof. In certain embodiments, the administration of an immunotherapy disclosed herein induces a complete response. In other embodiments, the administration of the immunotherapy disclosed herein induces a partial response. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a LAG-3 inhibitor. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor. In some embodiments, the immunotherapy comprises administering to the patient a therapeutically effective amount of an anti-PD-1 antibody. In some embodiments, the immunotherapy comprises administering to the patient a therapeutically effective amount of

an anti-PD-L1 antibody. In some embodiments, the immunotherapy comprises administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody. In some embodiments, the immunotherapy comprises administering a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-1 antibody. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody and the PD-1 pathway inhibitor is an anti-PD-L1 antibody.

[0186] In some embodiments, the LAG-3 positive tumor comprises at least about 1%, at least about 2%, at least about 3%, at least about 4%, at least about 5%, at least about 7%, at least about 10%, at least about 15%, at least about 20%, at least about 25%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, or 100% cells expressing LAG-3. In some embodiments, the cells expressing LAG-3 comprise tumor infiltrating lymphocytes.

[0187] In some embodiments, the identifying comprises determining LAG-3 expression in a malignant tumor.

[0188] In some embodiments, LAG-3 expression is determined by receiving the results of an assay capable of determining LAG-3 expression.

[0189] In certain embodiments, any of the present methods further comprise determining PD-L1 expression in the tumor sample.

[0190] In certain embodiments, any of the present methods further comprise identifying the patient as having a PD-L1 positive malignant tumor prior to the administration. In certain embodiments, any of the present methods further comprise identifying the patient as having a PD-L1 negative malignant tumor prior to the administration.

[0191] In certain embodiments, any of the present methods further comprise determining PD-L1 expression in the malignant tumor.

[0192] In certain embodiments of any of the present methods, the patient is identified as having a PD-L1 positive malignant tumor prior to the administration. In certain embodiments of any of the present methods, the patient is identified as having a PD-L1 negative malignant tumor prior to the administration.

[0193] Method for determining PD-L1 expression in a tumor sample, methods for identifying the patient as having a PD-L1 positive malignant tumor, and methods for determining PD-L1 expression in a malignant tumor have been disclosed in PCT/US2016/029878.

[0194] In certain embodiments, the methods of the invention include methods of treating a human patient with unresectable or metastatic melanoma in need thereof with a combination of a PD-1 pathway inhibitor and a LAG-3 inhibitor, wherein the patient was previously treated with an anti-PD-1 inhibitor and/or an anti-PD-L1 inhibitor. In certain embodiments, the PD-1 pathway inhibitor is an anti-PD-1 antibody. In particular embodiments, the anti-PD-1 antibody is nivolumab. In some embodiments, the LAG-3 inhibitor is an anti-LAG-3 antibody. In certain embodiments, the LAG-3 antibody is BMS-986016. In embodiments, the melanoma is a LAG-3 expressing tumor. In particular embodiments, the melanoma is a LAG-3 expression tumor, with LAG-3 expression $\geq$ 1%.

Measurement of LAG-3 expression

[0195] In certain embodiments, identifying a patient suitable for a LAG-3 inhibitor/ PD-1 pathway inhibitor combination therapy, a PD-1 pathway inhibitor (e.g., an anti-PD-1 antibody) therapy, or an anti-CTLA-4 antibody therapy for the present methods includes measuring or assessing a LAG-3 expression in a sample, for example, a malignant tumor test tissue sample comprising tumor cells and tumor infiltrating inflammatory cells. The phrases "tumors expressing LAG-3," "LAG-3 expressing tumor," "LAG-3 positive tumor," and "LAG-3 expression positive tumor" are used interchangeably herein and encompass tumors comprising LAG-3 expressing tumor-infiltrating lymphocytes. The meaning of the phrases is provided elsewhere herein. The methods of measuring or assessing the LAG-3 expression can be achieved by any methods applicable.

[0196] In order to assess the LAG-3 expression, in one embodiment, a test tissue sample is obtained from the patient who is in need of the therapy. In some embodiments, a test tissue sample includes, but is not limited to, any clinically relevant tissue sample, such as a tumor biopsy, a core biopsy tissue sample, a fine needle aspirate, or a sample of bodily fluid, such as blood, plasma, serum, lymph, ascites fluid, cystic fluid, or urine. In some embodiments, the test tissue sample is from a primary tumor. In some embodiments, the test tissue sample is from a metastasis. In some embodiments, test tissue samples are taken from a subject at multiple time points, for example, before treatment, during treatment, and/or after treatment. In some embodiments, test tissue samples are taken from different locations in the subject, for example, a sample from a primary tumor and a sample from a metastasis in a distant location.

[0197] In some embodiments, the test tissue sample is a paraffin-embedded fixed tissue sample. In some embodiments, the test tissue sample is a formalin-fixed paraffin embedded (FFPE) tissue sample. In some embodiments, the test tissue sample is a fresh tissue (e.g., tumor) sample. In some embodiments, the test tissue sample is a frozen tissue sample. In some embodiments, the test tissue sample is a fresh frozen (FF) tissue (e.g., tumor) sample. In some embodiments, the test tissue sample is a cell isolated from a fluid. In some embodiments, the test tissue sample comprises circulating tumor cells (CTCs). In some embodiments, the test tissue sample comprises tumor-infiltrating lymphocytes (TILs). In some embodiments, the test tissue sample comprises tumor cells and tumor-infiltrating lymphocytes (TILs). In some embodiments, the test tissue sample comprises circulating lymphocytes. In some embodiments, the test tissue sample is an archival tissue sample. In some embodiments, the test tissue sample is an archival tissue sample with known diagnosis, treatment, and/or outcome history. In some embodiments, the sample is a block of tissue. In some embodiments, the test tissue sample is dispersed cells. In some embodiments, the sample size is from about 1 cell to about 1 x 10^6 cells or more. In some embodiments, the sample size is about 1 cell to about 1 x 10^5 cells. In some embodiments, the sample size is about 1 cell to about 10,000 cells. In some embodiments, the sample size is about 1 cell to about 1,000 cells. In some embodiments, the sample size is about 1 cells to about 100 cells. In some embodiments, the sample size is about 1 cell to about 10 cells. In some embodiments, the sample size is a single cell.

[0198] In another embodiment, the assessment of LAG-3 expression can be achieved without obtaining a test tissue sample. In some embodiments, selecting a suitable patient includes (i) optionally providing a test tissue sample obtained from a patient with cancer of the tissue, the test tissue sample comprising tumor cells and/or tumor-infiltrating inflammatory cells; and (ii) assessing the proportion of cells in the test tissue sample that express LAG-3 on the surface of the cells based on an assessment that the proportion of cells in the test tissue sample that express LAG-3 on the cell surface is higher than a predetermined threshold level.

[0199] In any of the methods comprising the measurement of LAG-3 expression in a test tissue sample, however, it should be understood that the step comprising the provision of a test tissue sample obtained from a patient is an optional step. That is, in certain embodiments the method includes this step, and in other embodiments, this step is not included in the method. It should also be understood that in certain embodiments the "measuring" or "assessing" step to identify, or determine the number or proportion of, cells in the test tissue sample that express

LAG-3 is performed by a transformative method of assaying for LAG-3 expression, for example by performing a reverse transcriptase-polymerase chain reaction (RT-PCR) assay or an IHC assay. In certain other embodiments, no transformative step is involved and LAG-3 expression is assessed by, for example, reviewing a report of test results from a laboratory. In some embodiments, LAG-3 expression is assessed by reviewing the results of an immunohistochemistry assay from a laboratory. In certain embodiments, the steps of the methods up to, and including, assessing LAG-3 expression provides an intermediate result that may be provided to a physician or other healthcare provider for use in selecting a suitable candidate for the combination therapy of a LAG-3 inhibitor and a PD-1 pathway inhibitor. In certain embodiments, the steps of the methods up to, and including, assessing LAG-3 expression provides an intermediate result that may be provided to a physician or other healthcare provider for use in selecting a suitable candidate for PD-1 pathway inhibitor (e.g., anti-PD-1 antibody) therapy. In certain embodiments, the steps of the methods up to, and including, assessing LAG-3 expression provides an intermediate result that may be provided to a physician or other healthcare provider for use in selecting a suitable candidate for anti-CTLA-4 antibody therapy. In certain embodiments, the steps that provide the intermediate result is performed by a medical practitioner or someone acting under the direction of a medical practitioner. In other embodiments, these steps are performed by an independent laboratory or by an independent person such as a laboratory technician.

[0200] In certain embodiments of any of the present methods, the proportion of cells that express LAG-3 is assessed by performing an assay to detect the presence of LAG-3 RNA. In further embodiments, the presence of LAG-3 RNA is detected by RT-PCR, *in situ* hybridization or RNase protection. In some embodiments, the presence of LAG-3 RNA is detected by an RT-PCR based assay. In some embodiments, scoring the RT-PCR based assay comprises assessing the level of LAG-3 RNA expression in the test tissue sample relative to a predetermined level.

[0201] In other embodiments, the proportion of cells that express LAG-3 is assessed by performing an assay to detect the presence of LAG-3 polypeptide. In further embodiments, the presence of LAG-3 polypeptide is detected by IHC, enzyme-linked immunosorbent assay (ELISA), *in vivo* imaging, or flow cytometry. In some embodiments, LAG-3 expression is assayed by IHC. In other embodiments of all of these methods, cell surface expression of LAG-3 is assayed using, *e.g.*, IHC or *in vivo* imaging.

[0202] In embodiments, the biomarker measured is LAG-3, CD4, CD8, FOXP3, CD163 CD68, and any combination thereof. In embodiments, the biomarker is measured using any detection method disclosed herein. In other embodiments, the proportion of cells that express LAG-3 in the test tissue sample is assessed by flow cytometry. In some embodiments, the test tissue sample assayed by flow cytometry comprises tumor infiltrating immune cells. In some embodiments, the malignant tumor is a hematological malignancy and the tissue sample assayed by flow cytometry comprises peripheral blood cells. In some embodiments, the flow cytometry is a multiplex assay. In some embodiments, scoring the flow cytometry comprises detecting the expression of markers comprising LAG-3, CD4, CD8, FOXP3, and any combination thereof. In some embodiments, LAG-3, CD4, CD8, and FOXP3 are detected as single markers. In some embodiments, scoring the flow cytometry comprises assessing the proportion of T cells in the test tissue sample that express LAG-3. In some embodiments, scoring the flow cytometry comprises assessing the proportion of CD8+ T cells in the test tissue sample that express LAG-3. In some embodiments, scoring the flow cytometry comprises assessing the proportion of CD4+ T cells in the test tissue sample that express LAG-3. In some embodiments, scoring the flow cytometry comprises assessing the proportion of FOXP3+ T cells in the test tissue sample that express LAG-3. In some embodiments, scoring the flow cytometry comprises detecting the expression of markers comprising CD163 and/or CD68. In some embodiments, scoring the flow cytometry comprises assessing the proportion of cells in the test tissue sample that express CD163 and/or CD68.

[0203] In certain embodiments of any of the present methods, the proportion of cells that express LAG-3 in the test tissue sample is assessed by performing an assay to detect the presence of LAG-3 polypeptide. In some embodiments, the presence of LAG-3 polypeptide is detected by an immunohistochemistry assay. In some embodiments, the test tissue sample is a tumor biopsy. In some embodiments, the test tissue sample is a formalin-fixed paraffin embedded (FFPE) sample.

[0204] In some embodiments, the immunohistochemistry assay is a monoplex assay. In some embodiments, the immunohistochemistry assay is a multiplex assay. In some embodiments, the multiplex immunohistochemistry assay is capable of detecting the presence of CD4, CD8, FOXP3, CD163, CD68, or any combination thereof.

[0205] In some embodiments, the immunohistochemistry assay comprises contacting the tumor sample with the 17B4 mouse anti-human LAG-3 IgG1 monoclonal antibody. In some

embodiments, the immunohistochemistry assay comprises contacting the tumor sample with an anti-LAG-3 antibody comprising heavy and light chain variable regions comprising the sequences set forth in SEQ ID NOs: 3 and 5, respectively. In some embodiments, the immunohistochemistry assay comprises contacting the tumor sample with the SP346 rabbit anti-human LAG-3 IgG monoclonal antibody. In some embodiments, the immunohistochemistry assay comprises contacting the tumor sample with the 11E3 (Novusbio), 874501 (Novusbio), or EPR4392(2) (Abcam) anti-human LAG-3 monoclonal antibody.

[0206] Melanin, for example, in melanoma tumor samples, can interfere with histological analysis by obscuring histological features, and by interfering with and/or masking staining during immunohistochemistry (IHC). Melanin can be removed by bleaching the samples. See, e.g., Shen & Wu, Appl Immunohistochem Mol Morphol, 23(4): 303–307 (2015); Orchard & Calonje, Am J Dermatopathol, 20(4): 357-61 (1998). In some embodiments, the immunohistochemistry assay comprises melanin bleaching prior to contacting the sample with an anti-LAG-3 antibody. See, e.g., Figure 15. In some embodiments, the melanin bleaching comprises contacting the sample with dilute hydrogen peroxide (0.1 to 30% v/v), trichloroisocyanuric acid (TCCA), potassium permanganate/oxalic acid, or other traditional oxidation methods for depigmenting (i.e., removing melanin from) tissue samples.

[0207] In some embodiments, the immunohistochemistry assay uses a black or brown chromogen. In some embodiments, the immunohistochemistry assay uses a red chromogen. In some embodiments, the immunohistochemistry assay uses a blue chromogen. In some embodiments, the immunohistochemistry assay uses a green chromogen. In some embodiments, the immunohistochemistry assay uses a purple chromogen. In some embodiments, the immunohistochemistry assay uses a yellow chromogen.

[0208] In some embodiments, the immunohistochemistry assay is scored at a low magnification. In some embodiments, low magnification is about 20X. In some embodiments, the immunohistochemistry assay is scored at high magnification. In some embodiments, high magnification is about 40X.

[0209] In some embodiments, the immunohistochemistry assay is scored by an image analysis software. In some embodiments, the immunohistochemistry assay is scored by pathologist visual immune score. In some embodiments, the immunohistochemistry assay is scored manually.

[0210] In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of cells in the test tissue sample that express LAG-3. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of immune cells in the test tissue sample that express LAG-3. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of T cells in the test tissue sample that express LAG-3. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of CD8+ T cells in the test tissue sample that express LAG-3. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of CD4+ T cells in the test tissue sample that express LAG-3. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of FOXP3+ T cells in the test tissue sample that express LAG-3.

[0211] LAG-3 polypeptide localization includes partial membrane/cytoplasmic localization, dot like localization, perinuclear, and complete membrane/cytoplasmic localization. In some embodiments, cells with partial membrane/cytoplasmic LAG-3 localization are scored. In some embodiments, cells with dot-like LAG-3 localization are scored. In some embodiments, cells with complete membrane/cytoplasmic LAG-3 localization are scored. In some embodiments, cells with perinuclear LAG-3 localization are scored. In some embodiments, cells with any LAG-3 localization pattern are scored.

[0212] In some embodiments, the immunohistochemistry assay is a multiplex assay that further comprises detecting the expression of MHC Class II by the tumor cells. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of cells in the test tissue sample that expresses MHC Class II. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of non-immune cells in the test tissue sample that expresses MHC Class II. In some embodiments, the distribution of MHC II expressing cells is heterogenous in the tumor sample. In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of cells that expresses MHC Class II in regions of the tumor sample comprising a high density of MHC Class II expressing cells.

[0213] In some embodiments, the immunohistochemistry assay is a multiplex assay that further comprises detecting the expression of CD163 and/or CD68 by tumor infiltrating lymphocytes (TIL). In some embodiments, scoring the immunohistochemistry assay comprises assessing the proportion of TILs in the test tissue sample that expresses CD163 and/or CD68.

[0214] Imaging techniques have provided important tools in cancer research and treatment. Recent developments in molecular imaging systems, including positron emission tomography (PET), single-photon emission computed tomography (SPECT), fluorescence reflectance imaging (FRI), fluorescence-mediated tomography (FMT), bioluminescence imaging (BLI), laser-scanning confocal microscopy (LSCM) and multiphoton microscopy (MPM), will likely herald even greater use of these techniques in cancer research. Some of these molecular imaging systems allow clinicians to not only see where a tumor is located in the body, but also to visualize the expression and activity of specific molecules, cells, and biological processes that influence tumor behavior and/or responsiveness to therapeutic drugs (Condeelis and Weissleder, *Cold Spring Harb. Perspect. Biol. 2(12)*:a003848 (2010)). Antibody specificity, coupled with the sensitivity and resolution of PET, makes immunoPET imaging particularly attractive for monitoring and assaying expression of antigens in tissue samples (McCabe and Wu, *Cancer Biother. Radiopharm. 25(3)*:253-61 (2010); Olafsen *et al.*, *Protein Eng. Des. Sel. 23(4)*:243-9 (2010)). In certain embodiments of any of the present methods, LAG-3 expression is assayed by immunoPET imaging. In certain embodiments immunoPET is performed using a zirconium-89 radiolabeled anti-LAG-3 antibody. In certain embodiments of any of the present methods, the proportion of cells in a test tissue sample that express LAG-3 is assessed by performing an assay to determine the presence of LAG-3 polypeptide on the surface of cells in the test tissue sample. In certain embodiments, the test tissue sample is a FFPE tissue sample. In other embodiments, the presence of LAG-3 polypeptide is determined by IHC assay. In further embodiments, the IHC assay is performed using an automated process. In some embodiments, the IHC assay is performed using an anti-LAG-3 mAb to bind to the LAG-3 polypeptide.

Assaying LAG-3 Expression by Automated IHC

[0215] In one embodiment of the present methods, an automated IHC method is used to assay the expression of LAG-3 in FFPE tissue specimens. This disclosure provides methods for detecting the presence of human LAG-3 antigen in a test tissue sample, or quantifying the level of human LAG-3 antigen or the proportion of cells in the sample that express the antigen, which methods comprise contacting the test sample, and a negative control sample, with a mAb that specifically binds to human LAG-3, under conditions that allow for formation of a complex between the antibody or portion thereof and human LAG-3. In certain embodiments, the test and control tissue samples are FFPE samples. The formation of a complex is then detected, wherein a

difference in complex formation between the test sample and the negative control sample is indicative of the presence of human LAG-3 antigen in the sample. Various methods are used to quantify LAG-3 expression.

[0216] In a particular embodiment, the automated IHC method comprises: (a) deparaffinizing and rehydrating mounted tissue sections in an autostainer; (b) retrieving antigen in an autostainer; (c) setting up reagents on an autostainer; and (d) running the autostainer to include steps of neutralizing endogenous peroxidase in the tissue specimen; blocking non-specific protein-binding sites on the slides; incubating the slides with primary Ab; incubating with a postprimary blocking agent; incubating with a postprimary antibody detection agent, such as another antibody that may or may not be conjugated to a detection enzyme; incubating with a polymeric-enzyme detection reagent; adding a chromogen substrate and developing; and counterstaining with hematoxylin. In some embodiments, the retrieving antigen comprises using any heat based antigen retrieval device.

[0217] In some embodiments, for assessing LAG-3 expression in tumor tissue samples, a pathologist examines the number of LAG-3+ tumor cells in each field under a microscope and mentally estimates the percentage of cells that are positive, then averages them to come to the final percentage. The different staining intensities are defined as 0/negative, 1+/weak, 2+/moderate, and 3+/strong. Typically, percentage values are first assigned to the 0 and 3+ buckets, and then the intermediate 1+ and 2+ intensities are considered. For highly heterogeneous tissues, the specimen is divided into zones, and each zone is scored separately and then combined into a single set of percentage values. The percentages of negative and positive cells for the different staining intensities are determined from each area and a median value is given to each zone. A final percentage value is given to the tissue for each staining intensity category: negative, 1+, 2+, and 3+. The sum of all staining intensities needs to be 100%.

[0218] In some embodiments, staining is also assessed in tumor-infiltrating inflammatory cells such as macrophages and lymphocytes. Macrophages and lymphocytes are assessed for LAG-3 staining and only recorded for all samples as being positive or negative for each cell category. Staining is also characterized according to an outside/inside tumor immune cell designation. "Inside" means the immune cell is within the tumor tissue and/or on the boundaries of the tumor region without being physically intercalated among the tumor cells. "Outside" means that there is no physical association with the tumor, the immune cells being found in the periphery associated with connective or any associated adjacent tissue.

[0219] In certain embodiments of these scoring methods, the samples are scored by two or more pathologists operating independently, and the scores are subsequently consolidated. In certain other embodiments, the identification of positive and negative cells is scored using appropriate software.

[0220] A histoscore (H-score) is used as a more quantitative measure of the IHC data. The histoscore is calculated as follows:

Histoscore = [(% tumor x 1 (low intensity)) + (% tumor x 2 (medium intensity))
+ (% tumor x 3 (high intensity)]

[0221] To determine the histoscore, the pathologist estimates the percentage of stained cells in each intensity category within a specimen. Because expression of most biomarkers is heterogeneous the histoscore is a truer representation of the overall expression. The final histoscore range is 0 (minimum score, no expression) to 300 (maximum score, strong and inclusive expression).

3. LAG-3 inhibitors

[0222] In one aspect, the invention features methods of using a LAG-3 inhibitor in the treatment of malignant tumors. As used herein LAG-3 inhibitor includes, but is not limited to, LAG-3 binding agents and soluble LAG-3 polypeptides. LAG-3 binding agents include antibodies that specifically bind to LAG-3.

[0223] In some embodiments, a LAG-3 inhibitor is a LAG-3-binding agent, for example an anti-LAG-3 antibody. In some embodiments, the LAG-3 inhibitor is a soluble LAG-3 polypeptide, for example, a LAG-3-Fc fusion polypeptide capable of binding to MHC Class II.

[0224] Anti-human-LAG-3 antibodies (or VH/VL domains derived therefrom) suitable for use in the invention can be generated using methods well known in the art. Alternatively, art recognized anti-LAG-3 antibodies can be used. In certain embodiments, LAG-3 inhibitors include an anti-LAG-3 bispecific antibody. In some embodiments, the anti-LAG-3 antibody binds LAG-3 and PD-1.

[0225] In some embodiments, the anti-LAG-3 antibody is BMS-986016 comprising heavy and light chains comprising the sequences shown in SEQ ID NOs:1 and 2, respectively, or antigen binding fragments and variants thereof, as described in PCT/US13/48999.

[0226] In other embodiments, the antibody has the heavy and light chain CDRs or variable regions of BMS-986016. Accordingly, in one embodiment, the antibody comprises

CDR1, CDR2, and CDR3 domains of the VH region of BMS-986016 having the sequence set forth in SEQ ID NO:3, and CDR1, CDR2 and CDR3 domains of the VL region of BMS-986016 having the sequence set forth in SEQ ID NO:5. In another embodiment, the antibody comprises CDR1, CDR2 and CDR3 domains comprising the sequences set forth in SEQ ID NOs:7, 8, and 9, respectively, and CDR1, CDR2 and CDR3 domains comprising the sequences set forth in SEQ ID NOs:10, 11, and 12, respectively. In another embodiment, the antibody comprises VH and/or VL regions comprising the amino acid sequences set forth in SEQ ID NO:3 and/or SEQ ID NO: 5, respectively. In another embodiment, the antibody comprises heavy chain variable (VH) and/or light chain variable (VL) regions encoded by the nucleic acid sequences set forth in SEQ ID NO:4 and/or SEQ ID NO:6, respectively. In another embodiment, the antibody competes for binding with and/or binds to the same epitope on LAG-3 as the above-mentioned antibodies. In another embodiment, the antibody binds an epitope of human LAG-3 comprising the amino acid sequence PGHPLAPG (SEQ ID NO:14). In another embodiment, the antibody binds an epitope of human LAG-3 comprising the amino acid sequence HPAAPSSW (SEQ ID NO:15) or PAAPSSWG (SEQ ID NO:16).

[0227] In another embodiment, the antibody has at least about 90% variable region amino acid sequence identity with the above-mentioned antibodies (e.g., at least about 90%, 95% or 99% variable region identity with SEQ ID NO:3 or SEQ ID NO:5).

[0228] In some embodiments, art recognized anti-LAG-3 antibodies can be used in the therapeutic methods of the invention. For example, the anti-human LAG-3 antibody described in US2011/0150892 A1, and referred to as monoclonal antibody 25F7 (also known as "25F7" and "LAG-3.1) can be used. Other art recognized anti-LAG-3 antibodies that can be used include IMP731 (H5L7BW) described in US 2011/007023, MK-4280 (28G-10) described in WO2016028672, REGN3767 described in Journal for ImmunoTherapy of Cancer, (2016) Vol. 4, Supp. Supplement 1 Abstract Number: P195, BAP050 described in WO2017/019894, IMP-701 (LAG-525), Sym022, TSR-033, MGD013, BI754111, FS118, AVA-017 and GSK2831781. These and other anti-LAG-3 antibodies useful in the claimed invention can be found in, for example: WO2016/028672, WO2017/106129, WO2017/062888, WO2009/044273, WO2018/069500, WO2016/126858, WO2014/179664, WO2016/200782, WO2015/200119, WO2017/019846, WO2017/198741, WO2017/220555, WO2017/220569, WO2018/071500, WO2017/015560, WO2017/025498, WO2017/087589, WO2017/087901, WO2018/083087, WO2017/149143, WO2017/219995, US2017/0260271, WO2017/086367, WO2017/086419,

WO2018/034227, and WO2014/140180. In one embodiment, the LAG-3 inhibitor is IMP321 (eftilagimod alpha). The contents of each of these references are incorporated by reference herein in their entirety.

[0229] Antibodies that compete with any of the above-referenced art-recognized antibodies for binding to LAG-3 also can be used.

[0230] In certain embodiments, an anti-LAG-3 antibody is used to determine LAG-3 expression. In some embodiments, an anti-LAG-3 antibody is selected for its ability to bind to LAG-3 in formalin-fixed, paraffin-embedded (FFPE) tissue specimens. In other embodiments, an anti-LAG-3 antibody is capable of binding to LAG-3 in frozen tissues. In further embodiments, an anti-LAG-3 antibody is capable of distinguishing membrane bound, cytoplasmic, and/or soluble forms of LAG-3.

[0231] In some embodiments, an anti-LAG-3 antibody useful for assaying, detecting, and/or quantifying LAG-3 expression in accordance with the methods described herein is the 17B4 mouse IgG1 anti-human LAG-3 monoclonal antibody, or an antigen binding fragment thereof. See, e.g., J. Matsuzaki, et al.; PNAS 107, 7875 (2010).

4. PD-1 pathway inhibitors

[0232] In one aspect, the invention features methods of using a PD-1 inhibitor in the treatment of malignant tumors. As used herein "PD-1 pathway inhibitor" includes, but is not limited to, PD-1 binding agents, PD-L1 binding agent and PD-L2 binding agents. PD-1 binding agents include antibodies that specifically bind to PD-1. PD-L1 and PD-L2 binding agents include antibodies that specifically bind to PD-L1 and/or PD-L2, as well as soluble PD-1 polypeptides that bind to PD-L1 and/or PD-L2.

[0233] In some embodiments, PD-1 pathway inhibitor is a PD-1-binding agent, for example an anti-PD-1 antibody. In some embodiments, the PD-1 pathway inhibitor is a PD-L1-binding agent, for example, an anti-PD-L1 antibody. In some embodiments, the PD-1 pathway inhibitor is a PD-L2-binding agent, for example an anti-PD-L2 antibody. In further embodiments, the PD-L1-binding agent is a soluble PD-1 polypeptide, for example, a PD-1-Fc fusion polypeptide capable of binding to PD-L1. In further embodiments, the PD-L2-binding agent is a soluble PD-1 polypeptide, for example, a PD-1-Fc fusion polypeptide capable of binding to PD-L2.

[0234] Anti-human-PD-1 antibodies (or VH and/or VL domains derived therefrom) suitable for use in the invention can be generated using methods well known in the art. Alternatively, art recognized anti-PD-1 antibodies can be used. For example, monoclonal antibodies 5C4 (referred to herein as Nivolumab or BMS-936558), 17D8, 2D3, 4H1, 4A11, 7D3, and 5F4, described in WO 2006/121168 can be used. Other known PD-1 antibodies include lambrolizumab (MK-3475) described in WO 2008/156712, and AMP-514 described in WO 2012/145493. Further known PD-1 antibodies and other PD-1 inhibitors include those described in, for example, WO 2009/014708, WO 03/099196, WO 2009/114335 and WO 2011/161699, which are herein incorporated by reference.. In one embodiment, the anti-PD-1 antibody is REGN2810. In one embodiment, the anti-PD-1 antibody is PDR001. Another known anti-PD-1 antibody is pidilizumab (CT-011).

[0235] In one embodiment, the anti-PD-1 antibody is nivolumab. Nivolumab (also known as "OPDIVO®"; formerly designated 5C4, BMS-936558, MDX-1106, or ONO-4538) is a fully human IgG4 (S228P) PD-1 immune checkpoint inhibitor antibody that selectively prevents interaction with PD-1 ligands (PD-L1 and PD-L2), thereby blocking the down-regulation of antitumor T-cell functions (U.S. Patent No. 8,008,449; Wang *et al.*, *Cancer Immunol Res.* 2(9):846-56 (2014)). In another embodiment, the anti-PD-1 antibody or fragment thereof cross-competes with nivolumab. In other embodiments, the anti-PD-1 antibody or fragment thereof binds to the same epitope as nivolumab. In certain embodiments, the anti-PD-1 antibody has the same CDRs as nivolumab.

[0236] In some embodiments, the anti-PD-1 antibody comprises heavy and light chains comprising the sequences shown in SEQ ID NOs:17 and 18, respectively, or antigen binding fragments and variants thereof.

[0237] In other embodiments, the antibody has heavy and light chain CDRs or variable regions of nivolumab. Accordingly, in one embodiment, the antibody comprises CDR1, CDR2, and CDR3 domains of the VH of nivolumab having the sequence set forth in SEQ ID NO:19, and CDR1, CDR2 and CDR3 domains of the VL of nivolumab having the sequence set forth in SEQ ID NO:21. In another embodiment, the antibody comprises CDR1, CDR2 and CDR3 domains comprising the sequences set forth in SEQ ID NOs:23, 24, and 25, respectively, and CDR1, CDR2 and CDR3 domains comprising the sequences set forth in SEQ ID NOs:26, 27, and 28, respectively. In another embodiment, the antibody comprises VH and/or VL regions comprising the amino acid sequences set forth in SEQ ID NO: 19 and/or SEQ ID NO: 21, respectively. In

another embodiment, the antibody comprises heavy chain variable (VH) and/or light chain variable (VL) regions encoded by the nucleic acid sequences set forth in SEQ ID NO:20 and/or SEQ ID NO:22, respectively. In another embodiment, the antibody competes for binding with and/or binds to the same epitope on PD-1 as the above-mentioned antibodies. In another embodiment, the antibody has at least about 90% variable region amino acid sequence identity with the above-mentioned antibodies (e.g., at least about 90%, 95% or 99% variable region identity with SEQ ID NO:19 or SEQ ID NO:21).

[0238] Human monoclonal antibodies (HuMAbs) that bind specifically to PD-1 with high affinity have been disclosed in U.S. Patent Nos. 8,008,449 and 8,779,105. Other anti-PD-1 mAbs have been described in, for example, U.S. Patent Nos. 6,808,710, 7,488,802, 8,168,757 and 8,354,509, and PCT Publication No. WO 2012/145493, which are herein incorporated by reference. In some embodiments, the anti-PD-1 antibody has been demonstrated to exhibit one or more of the following characteristics: (a) binds to human PD-1 with a K_D of 1 x 10^{-7} M or less, as determined by surface plasmon resonance using a Biacore biosensor system; (b) does not substantially bind to human CD28, CTLA-4 or ICOS; (c) increases T-cell proliferation in a Mixed Lymphocyte Reaction (MLR) assay; (d) increases interferon-γ production in an MLR assay; (e) increases IL-2 secretion in an MLR assay; (f) binds to human PD-1 and cynomolgus monkey PD-1; (g) inhibits the binding of PD-L1 and/or PD-L2 to PD-1; (h) stimulates antigen-specific memory responses; (i) stimulates antibody responses; and (j) inhibits tumor cell growth *in vivo*. Anti-PD-1 antibodies useful for the present invention include mAbs that bind specifically to human PD-1 and exhibit at least one, at least two, at least three, at least four, or at least five of the preceding characteristics. Anti-PD-1 antibodies that exhibit one or more of these characteristics have been disclosed in U.S. Patent Nos. 8,008,449, 8,779,105, 6,808,710, 7,488,802, 8,168,757 and 8,354,509, and PCT Publication No. WO 2012/145493, which are herein incorporated by reference. In another embodiment, the anti-PD-1 antibody is pembrolizumab. Pembrolizumab is a humanized monoclonal IgG4 (S228P) antibody directed against human cell surface receptor PD-1 (programmed death-1 or programmed cell death-1). Pembrolizumab is described, for example, in U.S. Patent Nos. 8,354,509 and 8,900,587, which are herein incorporated by reference.

[0239] In some embodiments, the anti-PD-1 antibody or fragment thereof cross-competes with pembrolizumab. In some embodiments, the anti-PD-1 antibody or fragment thereof binds to the same epitope as pembrolizumab. In certain embodiments, the anti-PD-1 antibody has the

same CDRs as pembrolizumab. In another embodiment, the anti-PD-1 antibody is pembrolizumab. Pembrolizumab (also known as "KEYTRUDA®", lambrolizumab, and MK-3475) is a humanized monoclonal IgG4 antibody directed against human cell surface receptor PD-1 (programmed death-1 or programmed cell death-1). Pembrolizumab is described, for example, in U.S. Patent Nos. 8,354,509 and 8,900,587; *see also* http://www.cancer.gov/drugdictionary?cdrid=695789 (last accessed: December 14, 2014). Pembrolizumab has been approved by the FDA for the treatment of relapsed or refractory melanoma.

[0240] In other embodiments, the anti-PD-1 antibody or fragment thereof cross-competes with MEDI0608. In still other embodiments, the anti-PD-1 antibody or fragment thereof binds to the same epitope as MEDI0608. In certain embodiments, the anti-PD-1 antibody has the same CDRs as MEDI0608. In other embodiments, the anti-PD-1 antibody is MEDI0608 (formerly AMP-514), which is a monoclonal antibody. MEDI0608 is described, for example, in US Pat. No. 8,609,089B2 or in http://www.cancer.gov/drugdictionary?cdrid=756047 (last accessed December 14, 2014).

[0241] In certain embodiments, the first antibody is an anti-PD-1 antagonist. One example of the anti-PD-1 antagonist is AMP-224, which is a B7-DC Fc fusion protein. AMP-224 is discussed in U.S. Publ. No. 2013/0017199 or in http://www.cancer.gov/publications/dictionaries/cancer-drug?cdrid=700595 (last accessed July 8, 2015).

[0242] In other embodiments, the anti-PD-1 antibody or fragment thereof cross-competes with BGB-A317. In some embodiments, the anti-PD-1 antibody or fragment thereof binds the same epitope as BGB-A317. In certain embodiments, the anti-PD-1 antibody has the same CDRs as BGB-A317. In certain embodiments, the anti-PD-1 antibody is BGB-A317, which is a humanized monoclonal antibody. BGB-A317 is described in U.S. Publ. No. 2015/0079109.

[0243] In some embodiments, the antibody is pidilizumab (CT-011), which is an antibody previously reported to bind to PD-1 but which is believed to bind to a different target. pidilizumab is described in US Pat. No. 8,686,119 B2 or WO 2013/014668 A1.

[0244] In certain embodiments, the antibodies that cross-compete for binding to human PD-1 with, or bind to the same epitope region of human PD-1 as, nivolumab are mAbs. For administration to human subjects, these cross-competing antibodies can be chimeric antibodies,

or humanized or human antibodies. Such chimeric, humanized or human mAbs can be prepared and isolated by methods well known in the art.

[0245] Other anti-PD-1 monoclonal antibodies have been described in, for example, U.S. Patent Nos. 6,808,710, 7,488,802, 8,168,757 and 8,354,509, US Publication No. 2016/0272708, and PCT Publication Nos. WO 2012/145493, WO 2008/156712, WO 2015/112900, WO 2012/145493, WO 2015/112800, WO 2014/206107, WO 2015/35606, WO 2015/085847, WO 2014/179664, WO 2017/020291, WO 2017/020858, WO 2016/197367, WO 2017/024515, WO 2017/025051, WO 2017/123557, WO 2016/106159, WO 2014/194302, WO 2017/040790, WO 2017/133540, WO 2017/132827, WO 2017/024465, WO 2017/025016, WO 2017/106061, WO 2017/19846, WO 2017/024465, WO 2017/025016, WO 2017/132825, and WO 2017/133540 each of which is incorporated by reference in its entirety.

[0246] In some embodiments, the anti-PD-1 antibody is selected from the group consisting of nivolumab (also known as OPDIVO®, 5C4, BMS-936558, MDX-1106, and ONO-4538), pembrolizumab (Merck; also known as KEYTRUDA®, lambrolizumab, and MK-3475; *see* WO2008/156712), PDR001 (Novartis; *see* WO 2015/112900), MEDI-0680 (AstraZeneca; also known as AMP-514; *see* WO 2012/145493), cemiplimab (Regeneron; also known as REGN-2810; *see* WO 2015/112800), JS001 (TAIZHOU JUNSHI PHARMA; *see* Si-Yang Liu et al., *J. Hematol. Oncol. 10*:136 (2017)), BGB-A317 (Beigene; *see* WO 2015/35606 and US 2015/0079109), INCSHR1210 (Jiangsu Hengrui Medicine; also known as SHR-1210; *see* WO 2015/085847; Si-Yang Liu et al., *J. Hematol. Oncol. 10*:136 (2017)), TSR-042 (Tesaro Biopharmaceutical; also known as ANB011; *see* WO2014/179664), GLS-010 (Wuxi/Harbin Gloria Pharmaceuticals; also known as WBP3055; *see* Si-Yang Liu et al., *J. Hematol. Oncol. 10*:136 (2017)), AM-0001 (Armo), STI-1110 (Sorrento Therapeutics; *see* WO 2014/194302), AGEN2034 (Agenus; *see* WO 2017/040790), MGA012 (Macrogenics, *see* WO 2017/19846), and IBI308 (Innovent; *see* WO 2017/024465, WO 2017/025016, WO 2017/132825, and WO 2017/133540).

[0247] Anti-PD-1 antibodies useful for the compositions of the disclosed invention also include antigen-binding portions of the above antibodies. It has been amply demonstrated that the antigen-binding function of an antibody can be performed by fragments of a full-length antibody. Examples of binding fragments encompassed within the term "antigen-binding portion" of an antibody include (i) a Fab fragment, a monovalent fragment consisting of the V_L, V_H, C_L and C_{H1} domains; (ii) a $F(ab')_2$ fragment, a bivalent fragment comprising two Fab fragments linked by a

disulfide bridge at the hinge region; (iii) a Fd fragment consisting of the V_H and C_{H1} domains; and (iv) a Fv fragment consisting of the V_L and V_H domains of a single arm of an antibody.

[0248] Anti-PD-1 antibodies usable in the disclosed methods also include isolated antibodies that bind specifically to human PD-1 and cross-compete for binding to human PD-1 with any anti-PD-1 antibody disclosed herein, e.g., nivolumab (*see, e.g.*, U.S. Patent No. 8,008,449 and 8,779,105; WO 2013/173223). In some embodiments, the anti-PD-1 antibody binds the same epitope as any of the anti-PD-1 antibodies described herein, e.g., nivolumab. The ability of antibodies to cross-compete for binding to an antigen indicates that these monoclonal antibodies bind to the same epitope region of the antigen and sterically hinder the binding of other cross-competing antibodies to that particular epitope region. These cross-competing antibodies are expected to have functional properties very similar those of the reference antibody, *e.g.*, nivolumab, by virtue of their binding to the same epitope region of PD-1. Cross-competing antibodies can be readily identified based on their ability to cross-compete with nivolumab in standard PD-1 binding assays such as Biacore analysis, ELISA assays or flow cytometry (*see, e.g.*, WO 2013/173223).

[0249] Anti-PD-1 antibodies suitable for use in the disclosed methods are antibodies that bind to PD-1 with high specificity and affinity, block the binding of PD-L1 and or PD-L2, and inhibit the immunosuppressive effect of the PD-1 signaling pathway. In any of the compositions or methods disclosed herein, an anti-PD-1 "antibody" includes an antigen-binding portion or fragment that binds to the PD-1 receptor and exhibits the functional properties similar to those of whole antibodies in inhibiting ligand binding and upregulating the immune system. In certain embodiments, the anti-PD-1 antibody or antigen-binding portion thereof cross-competes with nivolumab for binding to human PD-1. In other embodiments, the anti-PD-1 antibody or antigen-binding portion thereof is a chimeric, humanized or human monoclonal antibody or a portion thereof. In certain embodiments, the antibody is a humanized antibody. In other embodiments, the antibody is a human antibody. Antibodies of an IgG1, IgG2, IgG3 or IgG4 isotype can be used.

[0250] In certain embodiments, the anti-PD-1 antibody or antigen-binding portion thereof comprises a heavy chain constant region which is of a human IgG1 or IgG4 isotype. In certain other embodiments, the sequence of the IgG4 heavy chain constant region of the anti-PD-1 antibody or antigen-binding portion thereof contains an S228P mutation which replaces a serine residue in the hinge region with the proline residue normally found at the corresponding position

in IgG1 isotype antibodies. This mutation, which is present in nivolumab, prevents Fab arm exchange with endogenous IgG4 antibodies, while retaining the low affinity for activating Fc receptors associated with wild-type IgG4 antibodies (Wang *et al.*, 2014 *Cancer Immunol Res.* 2(9):846-56). In yet other embodiments, the antibody comprises a light chain constant region which is a human kappa or lambda constant region. In other embodiments, the anti-PD-1 antibody or antigen-binding portion thereof is a mAb or an antigen-binding portion thereof. In certain embodiments of any of the therapeutic methods described herein comprising administration of an anti-PD-1 antibody, the anti-PD-1 antibody is nivolumab. In other embodiments, the anti-PD-1 antibody is pembrolizumab. In other embodiments, the anti-PD-1 antibody is chosen from the human antibodies 17D8, 2D3, 4H1, 4A11, 7D3 and 5F4 described in U.S. Patent No. 8,008,449. In still other embodiments, the anti-PD-1 antibody is MEDI0608 (formerly AMP-514), AMP-224, or BGB-A317.

[0251] In embodiments, the anti-PD-1 antibody is a bispecific antibody. In embodiments, the anti-PD-1 antibody is a bispecific antibody that binds both PD-1 and LAG-3.

5. Anti-PD-L1 Antibodies

[0252] In certain embodiments, the present application encompasses use of an anti-PD-L1 antibody as the PD-1 pathway inhibitor. In one embodiment, the anti-PD-L1 antibody inhibits the binding of PD-L1 receptor, i.e., PD-1 to its ligand PD-L1.

[0253] Anti-human-PD-L1 antibodies (or VH and/or VL domains derived therefrom) suitable for use in the invention can be generated using methods well known in the art. Alternatively, art recognized anti-PD-L1 antibodies can be used. For example, human anti-PD-L1 antibodies disclosed in U.S. Pat. No. 7,943,743 can be used. Such anti-PD-L1 antibodies include 3G10, 12A4 (also referred to as BMS-936559), 10A5, 5F8, 10H10, 1B12, 7H1, 11E6, 12B7, and 13G4. [0110] In some embodiments, the anti-PD-L1 antibody is atezolizumab (Tecentriq or RG7446) (see, e.g., Herbst et al. (2013) J Clin Oncol 31(suppl):3000. Abstract; U.S. Patent No. 8,217,149), durvalumab (Imfinzi or MEDI4736) (Khleif (2013) In: Proceedings from the European Cancer Congress 2013; September 27-October 1, 2013; Amsterdam, The Netherlands. Abstract 802), avelumab (Bavencio). Other art recognized anti-PD-L1 antibodies which can be used include those described in, for example, U.S. Pat. Nos. 7,635,757 and 8,217,149, U.S. Publication No. 2009/0317368, and PCT Publication Nos. WO 2011/066389 and WO

2012/145493, which are herein incorporated by reference. Antibodies that compete with any of these art-recognized antibodies or inhibitors for binding to PD-L1 also can be used. Examples of anti-PD-L1 antibodies useful in the methods of the present disclosure include the antibodies disclosed in US Patent No. 9,580,507. Anti-PD-L1 human monoclonal antibodies disclosed in U.S. Patent No. 9,580,507 have been demonstrated to exhibit one or more of the following characteristics: (a) bind to human PD-L1 with a KD of 1 x 10-7 M or less, as determined by surface plasmon resonance using a Biacore biosensor system; (b) increase T-cell proliferation in a Mixed Lymphocyte Reaction (MLR) assay; (c) increase interferon-γ production in an MLR assay; (d) increase IL-2 secretion in an MLR assay; (e) stimulate antibody responses; and (f) reverse the effect of T regulatory cells on T cell effector cells and/or dendritic cells. Anti-PD-L1 antibodies usable in the present invention include monoclonal antibodies that bind specifically to human PD-L1 and exhibit at least one, in some embodiments, at least five, of the preceding characteristics.

[0254] In certain embodiments, the anti-PD-L1 antibody is BMS-936559 (formerly 12A4 or MDX-1105) (see, e.g., U.S. Patent No. 7,943,743; WO 2013/173223). In other embodiments, the anti-PD-L1 antibody is MPDL3280A (also known as RG7446 and atezolizumab) (see, e.g., Herbst et al. 2013 J Clin Oncol 31(suppl):3000; U.S. Patent No. 8,217,149), MEDI4736 (Khleif, 2013, In: Proceedings from the European Cancer Congress 2013; September 27-October 1, 2013; Amsterdam, The Netherlands. Abstract 802), or MSB0010718C (also called Avelumab; see US 2014/0341917). In certain embodiments, antibodies that cross-compete for binding to human PD-L1 with, or bind to the same epitope region of human PD-L1 as the above-references PD-L1 antibodies are mAbs. For administration to human subjects, these cross-competing antibodies can be chimeric antibodies, or can be humanized or human antibodies. Such chimeric, humanized or human mAbs can be prepared and isolated by methods well known in the art. In certain embodiments, the anti-PD-L1 antibody is selected from the group consisting of BMS-936559 (also known as 12A4, MDX-1105; see, e.g., U.S. Patent No. 7,943,743 and WO 2013/173223), atezolizumab (Roche; also known as TECENTRIQ®; MPDL3280A, RG7446; see US 8,217,149; see, also, Herbst et al. (2013) J Clin Oncol 31(suppl):3000), durvalumab (AstraZeneca; also known as IMFINZI™, MEDI-4736; see WO 2011/066389), avelumab (Pfizer; also known as BAVENCIO®, MSB-0010718C; see WO 2013/079174), STI-1014 (Sorrento; see WO2013/181634), CX-072 (Cytomx; see WO2016/149201), KN035 (3D Med/Alphamab; see Zhang et al., Cell Discov. 7:3 (March 2017), LY3300054 (Eli Lilly Co.; see, e.g., WO

2017/034916), and CK-301 (Checkpoint Therapeutics; see Gorelik et al., AACR:Abstract 4606 (Apr 2016)).

[0255] In certain embodiments, the PD-L1 antibody is atezolizumab (TECENTRIQ®). Atezolizumab is a fully humanized IgG1 monoclonal anti-PD-L1 antibody.

[0256] In certain embodiments, the PD-L1 antibody is durvalumab (IMFINZI™). Durvalumab is a human IgG1 kappa monoclonal anti-PD-L1 antibody.

[0257] In certain embodiments, the PD-L1 antibody is avelumab (BAVENCIO®). Avelumab is a human IgG1 lambda monoclonal anti-PD-L1 antibody.

[0258] In other embodiments, the anti-PD-L1 monoclonal antibody is selected from the group consisting of 28-8, 28-1, 28-12, 29-8, 5H1, and any combination thereof.

[0259] Anti-PD-L1 antibodies usable in the disclosed methods also include isolated antibodies that bind specifically to human PD-L1 and cross-compete for binding to human PD-L1 with any anti-PD-L1 antibody disclosed herein, e.g., atezolizumab, durvalumab, and/or avelumab. In some embodiments, the anti-PD-L1 antibody binds the same epitope as any of the anti-PD-L1 antibodies described herein, e.g., atezolizumab, durvalumab, and/or avelumab. The ability of antibodies to cross-compete for binding to an antigen indicates that these antibodies bind to the same epitope region of the antigen and sterically hinder the binding of other cross-competing antibodies to that particular epitope region. These cross-competing antibodies are expected to have functional properties very similar those of the reference antibody, e.g., atezolizumab and/or avelumab, by virtue of their binding to the same epitope region of PD-L1. Cross-competing antibodies can be readily identified based on their ability to cross-compete with atezolizumab and/or avelumab in standard PD-L1 binding assays such as Biacore analysis, ELISA assays or flow cytometry (see, e.g., WO 2013/173223).

[0260] In certain embodiments, the antibodies that cross-compete for binding to human PD-L1 with, or bind to the same epitope region of human PD-L1 antibody as, atezolizumab, durvalumab, and/or avelumab, are monoclonal antibodies. For administration to human subjects, these cross-competing antibodies are chimeric antibodies, engineered antibodies, or humanized or human antibodies. Such chimeric, engineered, humanized or human monoclonal antibodies can be prepared and isolated by methods well known in the art.

[0261] Anti-PD-L1 antibodies usable in the methods of the disclosed invention also include antigen-binding portions of the above antibodies. It has been amply demonstrated that the antigen-binding function of an antibody can be performed by fragments of a full-length antibody.

[0262] Anti-PD-L1 antibodies suitable for use in the disclosed methods or compositions are antibodies that bind to PD-L1 with high specificity and affinity, block the binding of PD-1, and inhibit the immunosuppressive effect of the PD-1 signaling pathway. In any of the compositions or methods disclosed herein, an anti-PD-L1 "antibody" includes an antigen-binding portion or fragment that binds to PD-L1 and exhibits the functional properties similar to those of whole antibodies in inhibiting receptor binding and up-regulating the immune system. In certain embodiments, the anti-PD-L1 antibody or antigen-binding portion thereof cross-competes with atezolizumab, durvalumab, and/or avelumab for binding to human PD-L1.

[0263] Anti-PD-L1 antibodies useful for the invention include antibodies engineered starting from antibodies having one or more of the V_H and/or V_L sequences disclosed herein, which engineered antibodies can have altered properties from the starting antibodies. An anti-PD-L1 antibody can be engineered by a variety of modifications as described above for the engineering of modified anti-PD-1 antibodies of the invention.

6. Anti-CTLA-4 Antibodies

[0264] In certain embodiments, the present application encompasses use of an anti-CTLA-4 antibody. In one embodiment, the anti-CTLA-4 antibody binds to and inhibits CTLA-4. In some embodiments, the anti-CTLA-4 antibody is ipilimumab (YERVOY), tremelimumab (ticilimumab; CP-675,206), AGEN-1884, or ATOR-1015.

7. Immune Checkpoint Inhibitors

[0265] In one aspect, the invention features methods of using a PD-1 inhibitor in combination with an immune checkpoint inhibitor in the treatment of malignant tumors. Any art recognized immune checkpoint inhibitor can be used.

[0266] In certain embodiments, the immune checkpoint inhibitor is a CTLA-4 antagonist, a CD80 antagonist, a CD86 antagonist, a Tim-3 antagonist, a TIGIT antagonist, a CD20 antagonist, a CD96 antagonist, a IDO1 antagonist, a STING antagonist, a GARP antagonist, a CD40 antagonist, A2aR antagonist, a CEACAM1 (CD66a) antagonist, a CEA antagonist, a CD47 antagonist a PVRIG antagonist, a TDO antagonist, a VISTA antagonist, or a KIR antagonist.

[0267] In one embodiment, the immune checkpoint inhibitor is a CTLA-4 antagonist. In certain embodiments, the CTLA-4 antagonist is an anti-CTLA-4 antibody or antigen binding

fragment thereof. In some embodiments, the anti-CTLA-4 antibody is ipilimumab (YERVOY), tremelimumab (ticilimumab; CP-675,206), AGEN-1884, or ATOR-1015.

[0268] In one embodiment, the CTLA-4 antagonist is a soluble CTLA-4 polypeptide. In one embodiment, the soluble CTLA-4 polypeptide is abatacept (Orencia), belatacept (Nulojix), RG2077, or RG-1046. In another embodiment, the CTLA-4 antagonist is a cell based therapy. In some embodiments, the CTLA-4 antagonist is an anti-CTLA-4 mAb RNA/GITRL RNA-transfected autologous dendritic cell vaccine or an anti-CTLA-4 mAb RNA-transfected autologous dendritic cell vaccine.

[0269] In one embodiment, the immune checkpoint inhibitor is a KIR antagonist. In certain embodiments, the KIR antagonist is an anti-KIR antibody or antigen binding fragment thereof. In some embodiments, the anti-KIR antibody is lirilumab (1-7F9, BMS-986015, IPH 2101) or IPH4102.

[0270] In one embodiment, the immune checkpoint inhibitor is TIGIT antagonist. In one embodiment, the TIGIT antagonist is an anti-TIGIT antibody or antigen binding fragment thereof. In certain embodiments, the anti-TIGIT antibody is BMS-986207, AB 154, COM902 (CGEN-15137), or OMP-313M32.

[0271] In one embodiment, the immune checkpoint inhibitor is Tim-3 antagonist. In certain embodiments, the Tim-3 antagonist is an anti-Tim-3 antibody or antigen binding fragment thereof. In some embodiments, the anti-Tim-3 antibody is TSR-022 or LY3321367.

[0272] In one embodiment, the immune checkpoint inhibitor is a IDO1 antagonist. In another embodiment, the IDO1 antagonist is indoximod (NLG8189; 1-methyl-$_D$-TRP), epacadostat (INCB-024360, INCB-24360), KHK2455, PF-06840003, navoximod (RG6078, GDC-0919, NLG919), BMS-986205 (F001287), or pyrrolidine-2,5-dione derivatives.

[0273] In one embodiment, the immune checkpoint inhibitor is a STING antagonist. In certain embodiments, the STING antagonist is 2' or 3'-mono-fluoro substituted cyclic-di-nucleotides; 2'3'-di-fluoro substituted mixed linkage 2',5' – 3',5' cyclic-di-nucleotides; 2'-fluoro substituted, bis-3',5' cyclic-di-nucleotides; 2',2''-diF-Rp,Rp,bis-3',5' cyclic-di-nucleotides; or fluorinated cyclic-di-nucleotides.

[0274] In one embodiment, the immune checkpoint inhibitor is CD20 antagonist. In some embodiments, the CD20 antagonist is an anti-CD20 antibody or antigen binding fragment thereof. In one embodiment, the anti-CD20 antibody is rituximab (RITUXAN; IDEC-102; IDEC-C2B8), ABP 798, ofatumumab, or obinutuzumab.

[0275] In one embodiment, the immune checkpoint inhibitor is CD80 antagonist. In certain embodiments, the CD80 antagonist is an anti-CD80 antibody or antigen binding fragment thereof. In one embodiment, the anti-CD80 antibody is galiximab or AV 1142742.

[0276] In one embodiment, the immune checkpoint inhibitor is a GARP antagonist. In some embodiments, the GARP antagonist is an anti-GARP antibody or antigen binding fragment thereof. In certain embodiments, the anti-GARP antibody is ARGX-115.

[0277] In one embodiment, the immune checkpoint inhibitor is a CD40 antagonist. In certain embodiments, the CD40 antagonist is an anti-CD40 antibody for antigen binding fragment thereof. In some embodiments, the anti-CD40 antibody is BMS3h-56, lucatumumab (HCD122 and CHIR-12.12), CHIR-5.9, or dacetuzumab (huS2C6, PRO 64553, RG 3636, SGN 14, SGN-40). In another embodiment, the CD40 antagonist is a soluble CD40 ligand (CD40-L). In one embodiment, the soluble CD40 ligand is a fusion polypeptide. In one embodiment, the soluble CD40 ligand is a CD40-L/FC2 or a monomeric CD40-L.

[0278] In one embodiment, the immune checkpoint inhibitor is an A2aR antagonist. In some embodiments, the A2aR antagonist is a small molecule. In certain embodiments, the A2aR antagonist is CPI-444, PBF-509, istradefylline (KW-6002), preladenant (SCH420814), tozadenant (SYN115), vipadenant (BIIB014), HTL-1071, ST1535, SCH412348, SCH442416, SCH58261, ZM241385, or AZD4635.

[0279] In one embodiment, the immune checkpoint inhibitor is a CEACAM1 antagonist. In some embodiments, the CEACAM1 antagonist is an anti-CEACAM1 antibody or antigen binding fragment thereof. In one embodiment, the anti-CEACAM1 antibody is CM-24 (MK-6018).

[0280] In one embodiment, the immune checkpoint inhibitor is a CEA antagonist. In one embodiment, the CEA antagonist is an anti-CEA antibody or antigen binding fragment thereof. In certain embodiments, the anti-CEA antibody is cergutuzumab amunaleukin (RG7813, RO-6895882) or RG7802 (RO6958688).

[0281] In one embodiment, the immune checkpoint inhibitor is a CD47 antagonist. In some embodiments, the CD47 antagonist is an anti-CD47 antibody or antigen binding fragment thereof. In certain embodiments, the anti-CD47 antibody is HuF9-G4, CC-90002, TTI-621, ALX148, NI-1701, NI-1801, SRF231, or Effi-DEM.

[0282] In one embodiment, the immune checkpoint inhibitor is a PVRIG antagonist. In certain embodiments, the PVRIG antagonist is an anti-PVRIG antibody or antigen binding fragment thereof. In one embodiment, the anti-PVRIG antibody is COM701 (CGEN-15029).

[0283] In one embodiment, the immune checkpoint inhibitor is a TDO antagonist. In one embodiment, the TDO antagonist is a 4-(indol-3-yl)-pyrazole derivative, a 3-indol substituted derivative, or a 3-(indol-3-yl)-pyridine derivative. In another embodiment, the immune checkpoint inhibitor is a dual IDO and TDO antagonist. In one embodiment, the dual IDO and TDO antagonist is a small molecule.

[0284] In one embodiment, the immune checkpoint inhibitor is a VISTA antagonist. In some embodiments, the VISTA antagonist is CA-170 or JNJ-61610588.

8. Pharmaceutical Compositions

[0285] Pharmaceutical compositions suitable for administration to human patients are typically formulated for parenteral administration, e.g., in a liquid carrier, or suitable for reconstitution into liquid solution or suspension for intravenous administration.

[0286] In general, such compositions typically comprise a pharmaceutically acceptable carrier. As used herein, the term "pharmaceutically acceptable" means approved by a government regulatory agency or listed in the U.S. Pharmacopeia or another generally recognized pharmacopeia for use in animals, particularly in humans. The term "carrier" refers to a diluent, adjuvant, excipient, or vehicle with which the compound is administered. Such pharmaceutical carriers can be sterile liquids, such as water and oils, including those of petroleum, animal, vegetable or synthetic origin, such as peanut oil, soybean oil, mineral oil, sesame oil, glycerol polyethylene glycol ricinoleate, and the like. Water or aqueous solution saline and aqueous dextrose and glycerol solutions may be employed as carriers, particularly for injectable solutions (e.g., comprising an anti-LAG-3 and/or anti-PD-1 antibody). Liquid compositions for parenteral administration can be formulated for administration by injection or continuous infusion. Routes of administration by injection or infusion include intravenous, intraperitoneal, intramuscular, intrathecal and subcutaneous. In one embodiment, the anti-LAG-3 and/or anti-PD-1 antibodies are administered intravenously (e.g., in separate formulations or together (in the same formulation or in separate formulations)).

9. Patient Populations

[0287] Provided herein are clinical methods for treating malignant tumors (e.g., advanced refractory solid tumors and hematological malignancies) in human patients using an immunotherapy disclosed herein, for example, a LAG-3 inhibitor (e.g., an anti-LAG-3 antibody), a PD-1 pathway inhibitor (e.g., an anti-PD-1 antibody), an anti-CTLA-4 antibody, or a combination of a LAG-3 inhibitor (e.g., an anti-LAG-3 antibody) and a PD-1 pathway inhibitor (e.g., an anti-PD-1 antibody).

Examples of cancers and/or malignant tumors that may be treated using the methods of the invention, include liver cancer, hepatocellular carcinoma (HCC), bone cancer, pancreatic cancer, skin cancer, oral cancer, cancer of the head or neck, breast cancer, lung cancer, small cell lung cancer, NSCLC, cutaneous or intraocular malignant melanoma, renal cancer, uterine cancer, ovarian cancer, colorectal cancer, colon cancer, rectal cancer, cancer of the anal region, stomach cancer, testicular cancer, uterine cancer, carcinoma of the fallopian tubes, carcinoma of the endometrium, carcinoma of the cervix, carcinoma of the vagina, carcinoma of the vulva, squamous cell carcinoma of the head and neck (SCCHN), non-Hodgkin's lymphoma, cancer of the esophagus, cancer of the small intestine, cancer of the endocrine system, cancer of the thyroid gland, cancer of the parathyroid gland, cancer of the adrenal gland, sarcoma of soft tissue, cancer of the urethra, cancer of the penis, solid tumors of childhood, lymphocytic lymphoma, cancer of the bladder, cancer of the kidney or ureter, carcinoma of the renal pelvis, neoplasm of the central nervous system (CNS), primary CNS lymphoma, tumor angiogenesis, spinal axis tumor, brain stem glioma, pituitary adenoma, Kaposi's sarcoma, epidermoid cancer, squamous cell cancer, environmentally induced cancers including those induced by asbestos, hematologic malignancies including, for example, multiple myeloma, B-cell lymphoma, Hodgkin lymphoma/primary mediastinal B-cell lymphoma, non-Hodgkin's lymphomas, acute myeloid lymphoma, chronic myelogenous leukemia, chronic lymphoid leukemia, follicular lymphoma, diffuse large B-cell lymphoma, Burkitt's lymphoma, immunoblastic large cell lymphoma, precursor B-lymphoblastic lymphoma, mantle cell lymphoma, acute lymphoblastic leukemia, mycosis fungoides, anaplastic large cell lymphoma, T-cell lymphoma, and precursor T-lymphoblastic lymphoma, and any combinations of said cancers. The present invention is also applicable to treatment of metastatic cancers. In embodiments, the cancer is renal cell carcinoma (RCC), gastric/gastoesophogeal junction carcinoma, non-small cell lung carcinoma (NSCLC), melanoma, squamous cell carcinoma of the head and neck (SCCHN), hepatocellular carcinoma, or urothelial carcinoma.

[0288] In certain embodiments, the melanoma is unresectable or metastatic melanoma. In embodiments, the patient was previously treated with an anti-PD-1 or an anti-PD-L1 antibody. In certain embodiments, the tumor is a LAG-3 expressing tumor. In particular embodiments, the tumor is a LAG-3 expressing tumor with LAG-3 expression ≥ 1%.

[0289] In one embodiment, the human patient suffers from unresectable metastatic melanoma and was previously treated with an anti-PD-1 or anti-PD-L1 metastatic inhibitor. In a particular embodiment, the human patient suffers from unresectable metastatic melanoma and was previously treated with an anti-PD-1 or anti-PD-L1 metastatic inhibitor and the tumor is a LAG-3 expressing tumor. In one embodiment, the human patient suffers from unresectable metastatic melanoma and was previously treated with an anti-PD-1 or anti-PD-L1 metastatic inhibitor and the tumor is a LAG-3 expressing tumor. In a certain embodiment, , the human patient suffers from unresectable metastatic melanoma and was previously treated with an anti-PD-1 or anti-PD-L1 metastatic inhibitor and the tumor is a LAG-3 expressing tumor with LAG-3 expression ≥ 1%.

[0290] In one embodiment, the human patient suffers from a malignant tumor that is refractory to treatment with an immune checkpoint inhibitor. In another embodiment, the patient suffers from a malignant tumor that is refractory to treatment with a PD-1 inhibitor. In another embodiment, the patient suffers from a malignant tumor that is refractory to treatment with an anti-PD-1 antibody. In another embodiment, the patient suffers from a malignant tumor that is refractory to treatment with an anti-PD-L1 antibody. In some embodiments, the malignant tumor is gastric cancer, renal cancer, HCC, SCCHN, or NSCLC.

[0291] In one embodiment, the human patient suffers from melanoma. In another embodiment, the patient suffers from melanoma that is refractory to treatment with an immune checkpoint inhibitor. In another embodiment, the patient suffers from melanoma that is refractory to treatment with a PD-1 inhibitor. In another embodiment, the patient suffers from melanoma that is refractory to treatment with an anti-PD-1 antibody. In another embodiment, the patient suffers from melanoma that is refractory to treatment with an anti-PD-L1 antibody.

[0292] In one embodiment, the human patient suffers from melanoma, gastric cancer, renal cancer, HCC, SCCHN, or NSCLC. In one embodiment, the human patient suffers from melanoma.

[0293] In one embodiment, the human patient suffers from NSCLC or a virally-related cancer (e.g., a human papilloma virus (HPV)-related tumor) or gastric adenocarcinoma. In a particular embodiment, the HPV-related tumor is HPV+ head and neck cancer (HNC). In another particular embodiment, the gastric adenocarcinoma is associated with Epstein-Barr virus (EBV) infection.

[0294] Patients can be tested or selected for one or more of the above described clinical attributes prior to, during or after treatment.

[0295] In accordance with the methods described herein, the malignant tumors can be tested to determine LAG-3 expression. In some embodiments, the malignant tumors treated in accordance with the methods disclosed herein are LAG-3 positive tumors. In some embodiments, the malignant tumor is a LAG-3 positive melanoma. In another embodiment, the malignant tumor is a LAG-3 positive gastric cancer, renal cancer, HCC, SCCHN, or NSCLC.

[0296] In some embodiments, at least about 0.5%, at least about 0.75%, at least about 1%, at least about 1.25%, at least about 1.5%, at least about 1.75%, at least about 2%, at least about 3% cells of the total number of cells in a LAG-3 positive melanoma tumor express LAG-3.

[0297] In some embodiments, at least about 0.5%, at least about 1%, at least about 2%, at least about 3%, at least about 4%, at least about 5%, at least about 6%, at least about 7%, at least about 8%, at least about 9%, at least about 10%, at least about 15%, at least about 20%, at least about 25%, or at least about 30% of the total number of cells of a malignant tumor express LAG-3. In some embodiments, the malignant tumor is melanoma, gastric cancer, renal cancer, HCC, SCCHN, or NSCLC.

[0298] In accordance with the methods described herein, the malignant tumors can be tested to determine LAG-3 and PD-L1 expression. In some embodiments, the malignant tumors treated in accordance with the methods disclosed herein are LAG-3 positive PD-L1 positive tumors. In some embodiments, the malignant tumor is a LAG-3 positive PD-L1 positive melanoma. In another embodiment, the malignant tumor is a LAG-3 positive PD-L1 positive gastric cancer, renal cancer, HCC, SCCHN, or NSCLC.

[0299] In some embodiments, the malignant tumors treated in accordance with the methods disclosed herein are LAG-3 positive PD-L1 negative tumors. In some embodiments, the malignant tumor is a LAG-3 positive PD-L1 negative melanoma. In another embodiment, the malignant tumor is a LAG-3 positive PD-L1 negative gastric cancer, renal cancer, HCC, SCCHN, or NSCLC.

10. Immunotherapies

[0300] In one aspect, immunotherapies provided herein involve administration of a LAG-3 inhibitor (e.g., an anti-LAG-3 antibody) and another antibody that blocks an inhibitory immune receptor (e.g., a receptor, which upon binding to its natural ligand, inhibits/neutralizes activity, such as cytotoxic activity), particularly an anti-PD-1 antibody or an anti-PD-L1 antibody, to treat subjects having malignant tumors (e.g., advanced refractory solid tumors or hematological malignancies). In another aspect, immunotherapies provided herein involve administration of an anti-PD-1 antibody or an anti-PD-L1 antibody to treat subjects having malignant tumors (e.g., advanced refractory solid tumors or hematological malignancies). In another aspect, immunotherapies provided herein involve administration of an anti-CTLA-4 antibody to treat subjects having malignant tumors (e.g., advanced refractory solid tumors or hematological malignancies).

[0301] In one embodiment, the invention provides an anti-LAG-3 antibody and an anti-PD-1 antibody in combination according to a defined clinical dosage regimen, to treat subjects having a malignant tumor (e.g., an advanced refractory solid tumor). In a particular embodiment, the anti-LAG-3 antibody is BMS-986016. In another embodiment, the anti-PD-1 antibody is BMS-936558. In another embodiment, dosage regimens are adjusted to provide the optimum desired response (e.g., an effective response).

[0302] In another embodiment, the invention provides an anti-LAG-3 antibody and an anti-PD-L1 antibody in combination according to a defined clinical dosage regimen, to treat subjects having a malignant tumor (e.g., an advanced refractory solid tumor). In a particular embodiment, the anti-LAG-3 antibody is BMS-986016. In another embodiment, the anti-PD-L1 antibody is BMS-936559. In another embodiment, dosage regimens are adjusted to provide the optimum desired response (e.g., an effective response).

[0303] In another aspect, the invention provides an anti-LAG-3 antibody according to a defined clinical dosage regimen, to treat subjects having a malignant tumor (e.g., an advanced refractory solid tumor). In a particular embodiment, the anti-LAG-3 antibody is BMS-986016. In another embodiment, dosage regimens are adjusted to provide the optimum desired response (e.g., an effective response).

[0304] In another aspect, the invention provides an anti-PD-1 antibody according to a defined clinical dosage regimen, to treat subjects having a malignant tumor (e.g., an advanced refractory solid tumor). In a particular embodiment, the anti-PD-1 antibody is BMS-936558. In

another embodiment, dosage regimens are adjusted to provide the optimum desired response (e.g., an effective response).

[0305] In another aspect, the invention provides an anti-PD-L1 antibody according to a defined clinical dosage regimen, to treat subjects having a malignant tumor (e.g., an advanced refractory solid tumor). In a particular embodiment, the anti-PD-L1 antibody is BMS-936559. In another embodiment, dosage regimens are adjusted to provide the optimum desired response (e.g., an effective response).

[0306] In another aspect, the invention provides an anti-CTLA-4 antibody according to a defined clinical dosage regimen, to treat subjects having a malignant tumor (e.g., an advanced refractory solid tumor). In a particular embodiment, the anti- CTLA4 antibody is ipilimumab (YERVOY). In a particular embodiment, the anti- CTLA4 antibody is tremelimumab (ticilimumab; CP-675,206), AGEN-1884, or ATOR-1015. In another embodiment, dosage regimens are adjusted to provide the optimum desired response (e.g., an effective response).

[0307] In another aspect, immunotherapies provided herein involve administration of an anti-PD-1 antibody and an immune checkpoint inhibitor to treat subjects having malignant tumors (e.g., advanced refractory solid tumors or hematological malignancies). In one embodiment, the anti-PD-1 antibody is BMS-936558. In one embodiment, the immune checkpoint inhibitor is a CTLA-4 antagonist, a CD80 antagonist, a CD86 antagonist, a Tim-3 antagonist, a TIGIT antagonist, a CD20 antagonist, a CD96 antagonist, a IDO1 antagonist, a STING antagonist, a GARP antagonist, a CD40 antagonist, A2aR antagonist, a CEACAM1 (CD66a) antagonist, a CEA antagonist, a CD47 antagonist a PVRIG antagonist, a TDO antagonist, a VISTA antagonist, or a KIR antagonist.

[0308] In another aspect, immunotherapies provided herein involve administration of an anti-PD-L1 antibody and an immune checkpoint inhibitor to treat subjects having malignant tumors (e.g., advanced refractory solid tumors or hematological malignancies). In one embodiment, the anti-PD-L1 antibody is BMS-936559. In one embodiment, the immune checkpoint inhibitor is a CTLA-4 antagonist, a CD80 antagonist, a CD86 antagonist, a Tim-3 antagonist, a TIGIT antagonist, a CD20 antagonist, a CD96 antagonist, a IDO1 antagonist, a STING antagonist, a GARP antagonist, a CD40 antagonist, A2aR antagonist, a CEACAM1 (CD66a) antagonist, a CEA antagonist, a CD47 antagonist a PVRIG antagonist, a TDO antagonist, a VISTA antagonist, or a KIR antagonist.

[0309] As used herein, adjunctive or combined administration (coadministration) includes simultaneous administration of the compounds in the same or different dosage form, or separate administration of the compounds (e.g., sequential administration). Thus, for example, the anti-LAG-3 and anti-PD-1 antibodies can be simultaneously administered in a single formulation. Alternatively, the anti-LAG-3 and anti-PD-1 antibodies can be formulated for separate administration and are administered concurrently or sequentially (e.g., one antibody is administered within about 30 minutes prior to administration of the second antibody).

[0310] For example, the anti-PD-1 antibody can be administered first followed by (e.g., immediately followed by) the administration of the anti-LAG-3 antibody, or vice versa. In one embodiment, the anti-PD-1 antibody is administered prior to administration of the anti-LAG-3 antibody. In another embodiment, the anti-PD-1 antibody is administered after administration of the anti-LAG-3 antibody. In another embodiment, the anti-LAG-3 antibody and anti-PD-1 antibody are administered concurrently. Such concurrent or sequential administration preferably results in both antibodies being simultaneously present in treated patients.

11. Treatment Protocols

[0311] In one aspect, suitable treatment protocols for treating a malignant tumor in a human patient include administering to the patient an effective amount of a LAG3 inhibitor (e.g., an anti-LAG-3 antibody).

[0312] In some embodiments, a suitable treatment protocol for treating a malignant tumor in a human patient include, for example, administering to the patient an effective amount of an anti-LAG-3 antibody, such as one comprising CDR1, CDR2 and CDR3 domains of the heavy chain variable region having the sequence set forth in SEQ ID NO:3, and CDR1, CDR2 and CDR3 domains of the light chain variable region having the sequence set forth in SEQ ID NO:5, wherein the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycles, at least four doses of the anti-LAG-3 antibody are administered at a flat dose of about 1, 3, 10, 20, 50, 80, 100, 130, 150, 16, 180, 200, 240 or 280 mg. In another embodiment, four doses of the anti-LAG-3 antibody are administered at a dose of 0.01, 0.03, 0.25, 0.1, 0.3, 1 or 3, 5, 8 or 10 mg/kg body weight.

[0313] In one aspect, suitable treatment protocols for treating a malignant tumor in a human patient include administering to the patient an effective amount of a PD1 pathway inhibitor (e.g., an anti-PD1 antibody).

In some embodiments, a suitable treatment protocol for treating a malignant tumor in a human patient include, for example, administering to the patient an effective amount of an anti-PD-1 antibody, such as one comprising CDR1, CDR2 and CDR3 domains of the heavy chain variable region having the sequence set forth in SEQ ID NO:19, and CDR1, CDR2 and CDR3 domains of the light chain variable region having the sequence set forth in SEQ ID NO:21, wherein the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycles, at least four doses of the anti-PD-1 antibody are administered at flat dose of about 50, 80, 100, 130, 150, 180, 200, 240 or 280 mg. In another embodiment, four doses of the anti-PD-1 antibody are administered at a dose of 0.1, 0.3, 1, 3, 5, 8 or 10 mg/kg body weight.

[0314] In one aspect, suitable treatment protocols for treating a malignant tumor in a human patient include administering to the patient an effective amount of an anti-CTLA-4 antibody.

[0315] In some embodiments, a suitable treatment protocol for treating a malignant tumor in a human patient include, for example, administering to the patient an effective amount of an anti-CTLA-4 antibody, wherein the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycles, at least four doses of the anti-CTLA-4 antibody are administered at flat dose of about 50, 80, 100, 130, 150, 180, 200, 240 or 280 mg. In another embodiment, four doses of the anti-CTLA-4 antibody are administered at a dose of 0.1, 0.3, 1, 3, 5, 8 or 10 mg/kg body weight.

[0316] In one aspect, suitable treatment protocols for treating a malignant tumor in a human patient include administering to the patient an effective amount of each of a LAG3 inhibitor (e.g., an anti-LAG-3 antibody) and a PD-1 pathway inhibitor (e.g., an anti-PD-1 antibody).

[0317] In some embodiments, a suitable treatment protocol for treating a malignant tumor in a human patient include, for example, administering to the patient an effective amount of each of:

(a) an anti-LAG-3 antibody, such as one comprising CDR1, CDR2 and CDR3 domains of the heavy chain variable region having the sequence set forth in SEQ ID NO:3, and CDR1, CDR2 and CDR3 domains of the light chain variable region having the sequence set forth in SEQ ID NO:5,

(b) an anti-PD-1 antibody, such as one comprising CDR1, CDR2 and CDR3 domains of the heavy chain variable region having the sequence set forth in SEQ ID NO:19, and CDR1, CDR2 and CDR3 domains of the light chain variable region having the sequence set forth in SEQ ID NO:21,

wherein the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycles, at least four doses of the anti-LAG-3 antibody are administered at a flat dose of about 1, 3, 10, 20, 50, 80, 100, 130, 150, 16, 180, 200, 240 or 280 mg and at least four doses of the anti-PD-1 antibody are administered at flat dose of about 50, 80, 100, 130, 150, 180, 200, 240 or 280 mg. In another embodiment, four doses of the anti-LAG-3 antibody are administered at a dose of 0.01, 0.03, 0.25, 0.1, 0.3, 1 or 3, 5, 8 or 10 mg/kg body weight and four doses of the anti-PD-1 antibody are administered at a dose of 0.1, 0.3, 1, 3, 5, 8 or 10 mg/kg body weight.

[0318] In one embodiment, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the following doses:

(a) 3 mg of anti-LAG-3 antibody and 80 mg of anti-PD-1 antibody;

(b) 3 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody;

(c) 20 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody;

(d) 80 mg of anti-LAG-3 antibody and 160 mg of anti-PD-1 antibody;

(e) 80 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody;

(f) 160 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody; or

(g) 240 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody.

[0319] In one embodiment, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at a dose of 20 mg of anti-LAG-3 antibody and 80 mg of anti-PD-1 antibody. In one embodiment, the tumor is lung cancer.

[0320] In one embodiment, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at a dose of 20 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody.

[0321] In one embodiment, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at a dose of 80 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody. In one embodiment, the tumor is melanoma (e.g., anti-PD1/PD-L1 antibody experienced melanoma or first line melanoma treatment), RCC (e.g., IO naïve RCC), NSCLC (e.g., anti-PD1/PD-L1 antibody experienced NSCLC), gastric cancer (e.g., IO naïve gastric cancer), HCC (e.g., IO naïve HCC), NSCLC (e.g., first line treatment of NSCLC), or SCCHN (e.g., IO naïve SCCHN).

[0322] In one embodiment, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at a dose of 240 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody.

[0323] In one embodiment, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at a dose of 160 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody. In one embodiment, the tumor is melanoma (e.g., anti-PD1/PD-L1 antibody experienced melanoma or first line melanoma treatment), RCC (e.g., IO naïve RCC), NSCLC (e.g., anti-PD1/PD-L1 antibody experienced NSCLC), gastric cancer (e.g., IO naïve gastric cancer), HCC (e.g., IO naïve HCC), NSCLC (e.g., first line treatment of NSCLC), or SCCHN (e.g., IO naïve SCCHN). In another embodiment, the tumor is Hodgkin's lymphoma (e.g., prior IO treated Hodgkin's lymphoma); DLBCL, PD-1/PD-L1 naïve Hodgkin's lymphoma, or PD-1/PD-L1 progressed/refractory Hodgkin's lymphoma.

[0324] In another embodiment, the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the following doses:

(a) 0.3 mg/kg of anti-LAG-3 antibody and 1 mg/kg of anti-PD-1 antibody;

(b) 0.3 mg/kg of anti-LAG-3 antibody and 3 mg/kg of anti-PD-1 antibody;

(c) 0.25 mg/kg of anti-LAG-3 antibody and 3 mg/kg of anti-PD-1 antibody;

(d) 1 mg/kg of anti-LAG-3 antibody and 3 mg/kg of anti-PD-1 antibody; or

(e) 3 mg/kg of anti-LAG-3 antibody and 3 mg/kg of anti-PD-1 antibody.

[0325] In one embodiment, the dose of the anti-LAG-3 and/or anti-PD-1 antibody is calculated per body weight, e.g., mg/kg body weight. In another embodiment, the dose of the anti-LAG-3 and/or anti-PD-1 antibody is a flat-fixed dose. In another embodiment, the dose of the anti-LAG-3 and/or anti-PD-1 antibody is varied over time. For example, the anti-LAG-3 antibody and/or anti-PD-1 antibody may be initially administered at a high dose and may be lowered over time. In another embodiment, the anti-LAG-3 antibody and/or anti-PD-1 antibody is initially administered at a low dose and increased over time.

[0326] In another embodiment, the amount of the anti-LAG-3 and/or anti-PD-1 antibodies administered is constant for each dose. In another embodiment, the amount of antibody administered varies with each dose. For example, the maintenance (or follow-on) dose of the antibody can be higher or the same as the loading dose which is first administered. In another embodiment, the maintenance dose of the antibody can be lower or the same as the loading dose.

[0327] In another embodiment, the anti-LAG-3 and/or anti-PD-1 antibodies are formulated for intravenous administration. In one embodiment, the anti-PD-1 antibody is administered on Days 1, 15, 29, and 43 of each cycle. In another embodiment, the anti-LAG-3 antibody is administered on Days 1, 15, 29, and 43 of each cycle.

[0328] In other embodiments, the anti-LAG-3 and/or anti-PD-1 antibodies are administered about once per week, once about every or three two weeks, about once per month or as long as a clinical benefit is observed or until there is a complete response, confirmed progressive disease or unmanageable toxicity.

[0329] In another embodiment, a cycle of administration is eight weeks, which can be repeated, as necessary. In another embodiment, the treatment consists of up to 12 cycles.

[0330] In another embodiment, 4 doses of the anti-PD-1 antibody are administered per eight week cycle. In another embodiment, 4 doses of the anti-LAG-3 antibody are administered per eight week cycle.

[0331] In another embodiment, the anti-PD-1 antibody and anti-LAG-3 antibody are administered as a first line of treatment (e.g., the initial or first treatment). In another embodiment, the anti-PD-1 antibody and anti-LAG-3 antibody are administered as a second line of treatment (e.g., after the initial or first treatment, including after relapse and/or where the first treatment has failed).

[0332] In one embodiment, the invention provides a method of treating a human patient with unresectable or metastatic melanoma, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the patient has previously been treated with a PD-1 inhibitor. In some embodiments, the invention provides a method of treating a human patient with unresectable or metastatic melanoma, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the patient has previously been treated with a PD-L1 inhibitor. In certain embodiments, the invention is directed to a method of treating a human patient with unresectable or metastatic melanoma, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the patient has previously been treated with a PD-1 inhibitor, and wherein the melanoma expresses LAG-3. In one embodiment, the invention is directed to a method of treating a human patient with unresectable or metastatic melanoma, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the

patient has previously been treated with a PD-L1 inhibitor, and wherein the melanoma expresses LAG-3. In one embodiment, the invention provides a method of treating a human patient with melanoma that progressed while-on or after treatment with a PD-1 pathway inhibitor or a PD-L1 pathway inhibitor, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the patient has previously been treated with an anti-PD-1 inhibitor. In some embodiments, the invention provides a method of treating a human patient with melanoma that progressed while-on or after treatment with a PD-1 pathway inhibitor or a PD-L1 pathway inhibitor, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the patient has previously been treated with an anti-PD-L1 inhibitor. In certain embodiments, the invention provides a method of treating a human patient with melanoma that progressed while-on or after treatment with a PD-1 pathway inhibitor or a PD-L1 pathway inhibitor, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the patient has previously been treated with an anti-PD-1 inhibitor, and wherein the melanoma expresses LAG-3. In one embodiment, the invention provides a method of treating a human patient with melanoma that progressed while-on or after treatment with a PD-1 pathway inhibitor or a PD-L1 pathway inhibitor, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the patient has previously been treated with an anti-PD-L1 inhibitor, and wherein the melanoma expresses LAG-3. In some embodiments, the LAG-3 expression of the melanoma is $\geq$ 1%. In particular embodiments, the PD-1 pathway inhibitor administered is an anti-PD-1 antibody. In one embodiment, the PD-1 antibody is nivolumab. In certain embodiments, the LAG-3 inhibitor is a LAG-3 antibody. In one embodiment, the LAG-3 antibody is BMS-986016. In an embodiment, the PD-1 pathway inhibitor administered is an anti-PD-L1 antibody.

[0333] In one embodiment, the anti-LAG-3 antibody is BMS-986016 and the anti-PD-1 antibody is nivolumab. In one embodiment, the anti-LAG-3 antibody is MK-4280 and the anti-PD-1 antibody is pembrolizumab. In one embodiment, the anti-LAG-3 antibody is REGN3767 and the anti-PD-1 antibody is REGN2810. In one embodiment, the anti-LAG-3 antibody is LAG525 (Int'l Publ. No. WO2015/138920) and the anti-PD-1 antibody is PDR001.

[0334] In another aspect, the invention features any of the aforementioned embodiments, wherein the anti-PD-1 antibody is replaced by, or combined with, an anti-PD-L1 or anti-PD-L2 antibody.

[0335] In another aspect, the invention features any of the aforementioned embodiments, wherein administering the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor (e.g., anti-PD-1 antibody) activates the patient's T cells. In some embodiments, administering the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor (e.g., anti-PD-1 antibody) induces the expression of activation markers by the patient's T cells. Expression of activation markers by the patient's T cells can be detected by analyzing a patient sample, for example, peripheral lymphocytes or tumor-infiltrating lymphocytes using flow cytometry.

[0336] In another aspect, the invention features any of the aforementioned embodiments, wherein administering the anti-LAG-3 antibody or antigen-binding fragment thereof results in the occupancy of at least about 10%, at least about 20%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, at least about 95%, or about 100% of the LAG-3 receptors on the patient's T cells. In some embodiments, the T cells are CD8+ T cells. In some embodiments, the T cells are tumor infiltrating T cells.

[0337] In another aspect, the invention features any of the aforementioned embodiments, wherein the treatment protocol further comprises the administration of at least one additional therapeutic agent. In some embodiments, the at least one additional therapeutic agent is a chemotherapeutic agent. In some embodiments, the at least one additional therapeutic agent is an immune checkpoint inhibitor.

12. Outcomes

[0338] With Respect to Target Lesions, Responses to Therapy May Include:

Complete Response (CR) (RECIST V1.1)	Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.
Partial Response (PR) (RECIST V1.1)	At least a 30% decrease in the sum of the diameters of target lesions, taking as reference the baseline sum diameters.
Progressive Disease (PD) (RECIST V1.1)	At least a 20% increase in the sum of the diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progression).
Stable Disease (SD) (RECIST V1.1)	Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum

	diameters while on study.
Immune-related Complete Response (irCR) (irRECIST)	Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.
Immune-related Partial Response (irPR) (irRECIST)	At least a 30% decrease in the sum of diameters of target lesions and all new measurable lesions (i.e., Percentage Change in Tumor Burden), taking as reference the baseline sum diameters. Note: the appearance of new measurable lesions is factored into the overall Tumor Burden, but does not automatically qualify as progressive disease until the sum of the diameters increases by ≥20% when compared to nadir.
Immune-related Progressive Disease (irPD) (irRECIST)	At least a 20% increase in Tumor Burden (ie the sum of diameters of target lesions, and any new measurable lesions) taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. Tumor assessments using immune-related criteria for progressive disease incorporates the contribution of new measurable lesions. Each net percentage change in tumor burden per assessment accounts for the size and growth kinetics of both old and new lesions as they appear.
Immune-related Stable Disease (irSD) (irRECIST)	Neither sufficient shrinkage to qualify for irPR nor sufficient increase to qualify for irPD, taking as reference the smallest sum diameters while on study.

[0339] With respect to non-target lesions, responses to therapy may include:

Complete Response (CR) (RECIST V1.1)	Disappearance of all non-target lesions. All lymph nodes must be non- pathological in size (<10 mm short axis).
Non-CR/Non-PD (RECIST V1.1)	Persistence of one or more non-target lesion(s).
Progressive Disease (PD) (RECIST V1.1)	*Unequivocal progression* of existing non- target lesions. The appearance of one or more new lesions is also considered progression.
Immune-related Complete Response (irCR) (irRECIST)	Disappearance of all non-target lesions. All lymph nodes must be non-pathological in size (<10 mm short axis).
Immune-related Progressive Disease (irPD) (irRECIST)	Increases in number or size of non-target lesion(s) does not constitute progressive disease unless/until Tumor Burden increases by 20% (ie the sum of the diameters at nadir of target lesions and any new measurable lesions increases by the required amount). Non-target lesions are not considered in the definition of Stable Disease and Partial Response.

[0340] Patients treated according to the methods disclosed herein preferably experience improvement in at least one sign of cancer. In one embodiment, improvement is measured by a reduction in the quantity and/or size of measurable tumor lesions. In another embodiment, lesions

can be measured on chest x-rays or CT or MRI films. In another embodiment, cytology or histology can be used to evaluate responsiveness to a therapy.

[0341] In one embodiment, the patient treated exhibits a complete response (CR), a partial response (PR), stable disease (SD), immune-related complete disease (irCR), immune-related partial response (irPR), or immune-related stable disease (irSD). In another embodiment, the patient treated experiences tumor shrinkage and/or decrease in growth rate, i.e., suppression of tumor growth. In another embodiment, unwanted cell proliferation is reduced or inhibited. In yet another embodiment, one or more of the following can occur: the number of cancer cells can be reduced; tumor size can be reduced; cancer cell infiltration into peripheral organs can be inhibited, retarded, slowed, or stopped; tumor metastasis can be slowed or inhibited; tumor growth can be inhibited; recurrence of tumor can be prevented or delayed; one or more of the symptoms associated with cancer can be relieved to some extent.

[0342] In other embodiments, administration of effective amounts of the anti-LAG-3 antibody, anti-PD-1 antibody, anti-PD-L1 antibody, anti-CTLA-4 antibody, a combination of the anti-LAG-3 antibody and anti-PD-1 antibody, or a combination of the anti-PD-1 antibody and an immune checkpoint inhibitor according to any of the methods provided herein produces at least one therapeutic effect selected from the group consisting of reduction in size of a tumor, reduction in number of metastatic lesions appearing over time, complete remission, partial remission, or stable disease.

[0343] In still other embodiments, the methods of treatment produce a clinical benefit rate (CBR=CR+PR+SD≥6 months) better than that achieved by a method of treatment that does not comprise a step of (i) determining the level of LAG-3 expression in a tumor sample prior to treatment, (ii) selecting a LAG-3 positive tumor for treatment, (iii) treating a tumor that has been identified as LAG-3 positive prior to treatment, or (iv) any combinations thereof. In other embodiments, the improvement of clinical benefit rate is about 20% 20%, 30%, 40%, 50%, 60%, 70%, 80% or more compared to a method of treatment that does not comprise a step of (i) determining the level of LAG-3 expression in a tumor sample prior to treatment, (ii) selecting a LAG-3 positive tumor for treatment, (iii) treating a tumor that has been identified as LAG-3 positive prior to treatment, or (iv) any combinations thereof.

[0344] In still other embodiments, the methods of treatment produce an objective response rate (ORR=CR+PR) of at least about 15%, at least about 20%, at least about 25%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at

least about 80%, at least about 90%, or about 100%. In one embodiment, the methods of treatment produce an objective response rate of at least about 15%, wherein the malignant tumor is a LAG-3 positive melanoma that is resistant to treatment with an anti-PD-1 or anti-PD-L1antibody. In some embodiments, the median duration of response is ≥ 3 month, ≥ 6 month, ≥ 12 month, or ≥ 18 month. In one embodiment, the median duration of response is ≥ 6 month. In some embodiments, the frequency of patients with duration of response ≥ 6 month is at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, or 100%.

[0345] In still other embodiments, the methods of treatment produce an objective response rate (ORR=CR+PR) better than that achieved by a method of treatment that does not comprise a step of (i) determining the level of LAG-3 expression in a tumor sample prior to treatment, (ii) selecting a LAG-3 positive tumor for treatment, (iii) treating a tumor that has been identified as LAG-3 positive prior to treatment, or (iv) any combinations thereof. In other embodiments, the improvement of objective response rate is about 20% 20%, 30%, 40%, 50%, 60%, 70%, 80% or more compared to a method of treatment that does not comprise a step of (i) determining the level of LAG-3 expression in a tumor sample prior to treatment, (ii) selecting a LAG-3 positive tumor for treatment, (iii) treating a tumor that has been identified as LAG-3 positive prior to treatment, or (iv) any combinations thereof. In some embodiments, the median duration of response is ≥ 3 month, ≥ 6 month, ≥ 12 month, or ≥ 18 month. In one embodiment, the median duration of response is ≥ 6 month.

[0346] In still other embodiments, the methods of treatment produce a disease control rate (DRR=CR+PR+SD) of at least about 20%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, or about 100%. In one embodiment, the methods of treatment produce a disease control rate of at least about 70%, wherein the malignant tumor is a LAG-3 positive melanoma that is resistant to treatment with an anti-PD-1 or anti-PD-L1antibody. In some embodiments, the median duration of response is ≥ 3 month, ≥ 6 month, ≥ 12 month, or ≥ 18 month. In one embodiment, the median duration of response is ≥ 6 month. In some embodiments, the frequency of patients with duration of response ≥ 6 month is at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, or 100%.

[0347] In still other embodiments, the methods of treatment produce a disease control rate (DRR=CR+PR+SD) better than that achieved by a method of treatment that does not

comprise a step of (i) determining the level of LAG-3 expression in a tumor sample prior to treatment, (ii) selecting a LAG-3 positive tumor for treatment, (iii) treating a tumor that has been identified as LAG-3 positive prior to treatment, or (iv) any combinations thereof. In other embodiments, the improvement of disease control rate is about 20% 20%, 30%, 40%, 50%, 60%, 70%, 80% or more compared to a method of treatment that does not comprise a step of (i) determining the level of LAG-3 expression in a tumor sample prior to treatment, (ii) selecting a LAG-3 positive tumor for treatment, (iii) treating a tumor that has been identified as LAG-3 positive prior to treatment, or (iv) any combinations thereof. In some embodiments, the median duration of response is $\geq$ 3 month, $\geq$ 6 month, $\geq$ 12 month, or $\geq$ 18 month. In one embodiment, the median duration of response is $\geq$ 6 month.

13. Kits and Unit Dosage Forms

[0348] Also within the scope of the present invention are diagnostic kits comprising an anti-LAG-3 antibody for assaying LAG-3 expression as a biomarker for screening patients for the immunotherapy or for predicting the efficacy of the immunotherapy. Kits typically include a label indicating the intended use of the contents of the kit and instructions for use. The term "label" includes any writing, or recorded material supplied on or with the kit, or which otherwise accompanies the kit. In certain embodiments of a diagnostic kit, a first anti-LAG-3 antibody for assaying, detecting, and/or quantifying LAG-3 expression is co-packaged with at least one therapeutic antibody (e.g., a second anti-LAG-3 antibody, an anti-PD-1 antibody, an anti-PD-L1 antibody, and/or an anti-CTLA-4 antibody) for the treatment of a LAG-3 positive tumor. In some embodiments, the kit further comprises an anti-PD-L1 antibody for assaying, detecting, and/or quantifying PD-L1 expression as a biomarker for predicting the efficacy of the immunotherapy. In one embodiment, the immunotherapy comprises administering to the patient a therapeutically effective amount of a LAG-3 inhibitor (e.g., anti-LAG-3 antibody) and a PD-1 pathway inhibitor (e.g., anti-PD1 antibody or anti-PD-L1 antibody). In one embodiment, the immunotherapy comprises administering to the patient a therapeutically effective amount of a LAG-3 inhibitor (e.g., anti-LAG-3 antibody). In one embodiment, the immunotherapy comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor (e.g., anti-PD1 antibody or anti-PD-L1 antibody). In one embodiment, the immunotherapy comprises administering to the patient a therapeutically effective amount of an anti-PD1 antibody. In one embodiment, the immunotherapy comprises administering to the patient a therapeutically

effective amount of an anti-CTLA-4 antibody. In one embodiment, the immunotherapy comprises administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor (e.g., anti-PD1 antibody or anti-PD-L1 antibody) and an immune checkpoint inhibitor.

[0349] In certain embodiments, the diagnostic kit comprises an anti-human LAG-3 monoclonal antibody for assaying, detecting, and/or quantifying LAG-3 expression. See, e.g., J. Matsuzaki, et al.; PNAS 107, 7875 (2010).

[0350] Also provided herein are therapeutic kits which include a pharmaceutical composition containing an anti-LAG-3 antibody, such as BMS-986016, and an anti-PD-1 antibody, such as nivolumab, and a pharmaceutically-acceptable carrier, in a therapeutically effective amount adapted for use in the preceding methods. In certain embodiments of a therapeutic kit, the anti-LAG-3 antibody is co-packaged with an anti-PD-1 antibody in unit dosage form. The kits optionally also can include instructions, e.g., comprising administration schedules, to allow a practitioner (e.g., a physician, nurse, or patient) to administer the composition contained therein to administer the composition to a patient having cancer (e.g., a solid tumor). The kit also can include a syringe.

[0351] Optionally, the diagnostic and/or therapeutic kits include multiple packages of the single-dose pharmaceutical compositions each containing an effective amount of the anti-LAG-3 or anti-PD-1 antibody for a single administration in accordance with the methods provided above. Instruments or devices necessary for administering the pharmaceutical composition(s) also may be included in the kits. For instance, a kit may provide one or more pre-filled syringes containing an amount of the anti-LAG-3 or anti-PD-1 antibody.

[0352] In one embodiment, the present invention provides a kit for treating a patient afflicted with a malignant tumor, the kit, for example, comprising:

(a) a dose of an anti-LAG-3 antibody, such as one comprising CDR1, CDR2 and CDR3 domains of the heavy chain variable region having the sequence set forth in SEQ ID NO:3, and CDR1, CDR2 and CDR3 domains of the light chain variable region having the sequence set forth in SEQ ID NO:5;

(b) a dose of an anti-PD-1 antibody, such as one comprising CDR1, CDR2 and CDR3 domains of the heavy chain variable region having the sequence set forth in SEQ ID NO:19, and CDR1, CDR2 and CDR3 domains of the light chain variable region having the sequence set forth in SEQ ID NO:21; and

(c) instructions for using the anti-LAG-3 antibody and anti-PD-1 antibody in the methods described herein.

[0353] In one embodiment, the present invention provides a kit for treating a patient afflicted with a malignant tumor, the kit, for example, comprising:

(a) a dose of an anti-LAG-3 antibody, such as one comprising CDR1, CDR2 and CDR3 domains of the heavy chain variable region having the sequence set forth in SEQ ID NO:3, and CDR1, CDR2 and CDR3 domains of the light chain variable region having the sequence set forth in SEQ ID NO:5; and

(b) instructions for using the anti-LAG-3 antibody in the methods described herein.

[0354] In one embodiment, the present invention provides a kit for treating a patient afflicted with a malignant tumor, the kit, for example, comprising:

(a) a dose of an anti-PD-1 antibody, such as one comprising CDR1, CDR2 and CDR3 domains of the heavy chain variable region having the sequence set forth in SEQ ID NO:19, and CDR1, CDR2 and CDR3 domains of the light chain variable region having the sequence set forth in SEQ ID NO:21; and

(b) instructions for using the anti-PD-1 antibody in the methods described herein.

[0355] In one embodiment, the present invention provides a kit for treating a patient afflicted with a malignant tumor, the kit, for example, comprising:

(a) a dose of an anti-PD-L1 antibody, such as BMS-936559; and

(b) instructions for using the anti-PD-L1 antibody in the methods described herein.

[0356] In one embodiment, the present invention provides a kit for treating a patient afflicted with a malignant tumor, the kit, for example, comprising:

(a) a dose of an anti-CTLA-4 antibody, such as ipilimumab (YERVOY); and

(b) instructions for using the anti-CTLA-4 antibody in the methods described herein.

[0357] In one embodiment, the present invention provides a kit for treating a patient afflicted with a malignant tumor, the kit, for example, comprising:

(a) a dose of an anti-LAG-3 antibody, such as one comprising CDR1, CDR2 and CDR3 domains of the heavy chain variable region having the sequence set forth in SEQ ID NO:3, and CDR1, CDR2 and CDR3 domains of the light chain variable region having the sequence set forth in SEQ ID NO:5;

(b) a dose of an immune checkpoint inhibitor; and

(c) instructions for using the anti-PD-1 antibody and the immune checkpoint inhibitor in the methods described herein.

[0358] In some embodiments, the malignant tumor is a LAG-3 positive tumor. In some embodiments, the malignant tumor is a LAG-3/PD-L1 positive tumor. In some embodiments, the malignant tumor is a LAG-3 positive/PD-L1 negative tumor.

[0359] In some embodiments, the malignant tumor is melanoma.

[0360] The present invention is further illustrated by the following examples which should not be construed as further limiting.

EXAMPLES

EXAMPLE 1

Optimization and Validation of an Assay for the Automated Detection of LAG3 (Mouse Clone 17B4) by Single Stain Immunohistochemistry with DAB Chromogen and Evaluation by Image Analysis in Formalin-Fixed, Paraffin Embedded Human Tissue

[0361] The purpose of this study was to validate an immunohistochemical assay for lymphocyte activation gene- 3 (LAG3) using a commercially available antibody (mouse clone 17B4) from LS Biosciences, for use in formalin fixed, paraffin embedded (FFPE) human tissue.

[0362] Immunohistochemistry (IHC) refers to the process of localizing proteins or other molecules in cells of a tissue section. Immunohistochemical staining is widely used in the diagnosis of cancer and has recently been used to help predict whether patients are likely to respond to a targeted chemotherapeutic agent. As opposed to many other analytical techniques such as the Western blot or ELISA, IHC retains the spatial localization of protein expression within a tissue specimen. This technique involves using an antibody (primary antibody) to specifically bind a target within the cellular context and then using the bound antibody to deposit a dye in the region of the target.

[0363] Test System. FFPE validation were performed on remnant, de-identified, or anonymized human samples. Tissues used for sensitivity testing and analysis included 40 bladder urothelial cancer, 41 gastric/GEJ cancer, 41 HNSCC, 41 melanoma, 41 NSCLC, and 43 RCC. The positive and negative control selected for LAG3 IHC was a tonsil tissue. Tonsil tissue contains cellular features that are positive and negative for LAG3.

[0364] Test Articles. The LAG3 mouse clone 17B4 antibody was purchased from LS Biosciences (Seattle, WA) and stored at -20°C. A mouse IgG isotype control antibody was purchased from BD Pharmingen (San Jose, CA) and stored at 2-8°C.

[0365] Immunohistochemistry. Immunohistochemistry was performed in accordance with standard laboratory techniques.

[0366] Pre-Processing. The procedure for IHC analysis of LAG3 (mouse clone 17B4) was performed using automated detection at room temperature (RT) on the Leica Bond Rx (Leica Biosystems, Buffalo Grove, IL) using commercially available reagents. Specimens were sectioned at 4-micron thickness, mounted onto positive-charged glass slides, dried, baked, deparaffinized, and rehydrated offline. Tissues were then placed onto the autostainer and underwent pretreatment using Epitope Retrieval Solution 1 (Catalog# AR9961, Leica) for 20 minutes at 100°C followed by a rinse with Bond Wash Buffer (Catalog # AR9590, Leica) at RT.

[0367] DAB Chromogen Assay Tissues were incubated with Peroxide Block (Catalog# DS9800, Leica) for 5 minutes followed by 3 rinses in Bond Wash Buffer. Tissues were incubated with Protein Block, Serum Free (Catalog# X0909, Dako, Carpinteria, CA) for 5 minutes followed by incubation with the primary antibody or isotype negative control reagent diluted in Bond Primary Antibody Diluent (Catalog# AR9352, Leica) for 30 minutes and 3 rinses in Bond Wash Buffer. Tissues were incubated with Post Primary (Bond Polymer Refine Detection Kit, Catalog# DS9800, Leica) for 8 minutes followed by 3 rinses in Bond Wash buffer for 2 minutes each. Tissues were incubated with Polymer (Bond Polymer Refine Detection Kit) for 8 minutes followed by 3 rinses in Bond Wash buffer for 2 minutes each and 2 rinses in distilled water. Tissues were incubated with DAB (Bond Polymer Refine Detection Kit) for 10 minutes followed by 4 rinses in distilled water.

[0368] Red Chromogen Assay Tissues were then incubated with 3% hydrogen peroxide for 5 minutes followed by 3 rinses in Bond Wash Buffer. Tissues were incubated with Protein Block, Serum Free for 5 minutes followed by incubation with the primary antibody or isotype negative control reagent diluted in Bond Primary Antibody Diluent for 30 minutes and 3 rinses in Bond Wash Buffer. Tissues were incubated with Post Primary AP (Catalog# DS9390, Bond Polymer Refine Red Detection Kit, Leica) for 20 minutes followed by 3 rinses in Bond Wash buffer for 2 minutes each. Tissues were incubated with Polymer AP (Bond Polymer Refine Red Detection Kit) for 30 minutes followed by 3 rinses in Bond Wash buffer for 2 minutes each and 2

rinses in distilled water. Tissues were incubated with Red Refine (Bond Polymer Refine Red Detection Kit) for 10 minutes followed by 4 rinses in distilled water.

[0369] Post-Processing Tissues were incubated with Hematoxylin (Bond Polymer Refine Detection Kit) for 5 minutes followed by a rinse in distilled water and a rinse in Bond Wash Buffer. Coverslip mounting occurred offline using an automated glass coverslipper (Leica) in accordance with standard procedures.

[0370] Slides were scanned using an Aperio Turbo AT system (Aperio, Vista, CA) to produce whole slide images. A 20X JPEG image of each stain is provided for this report.

[0371] Image Analysis Tissues stained with LAG3 (mouse clone 17B4) using DAB chromogen or red chromogen were evaluated by image analysis with a Nuclear v9 algorithm from Aperio The ROI includes the area of tumor tissue with intervening stroma. Areas excluded from analysis include normal tissue, larger stromal areas, necrotic tissue, tar (if possible), and staining artifact.

[0372] A nuclear algorithm was selected because heavy cytoplasmic stains in small cells, such as immune cells, often obscure the hematoxylin in the nucleus. The cytoplasmic and membrane algorithm require visualization of hematoxylin in the nucleus to quantify a cell. The nuclear algorithm has a featured called “fill holes” that will fill the central portion of a lymphocyte if there is hematoxylin present and record it as one cell.

[0373] Pathologist Visual Immune Score A subset of samples within the dynamic range were also scored by a pathologist during QC of image analysis. The purpose of the pathologist visual immune score is to provide a back-up result in the event of an image analysis score that does not produce an accurate result as deemed by a board-certified pathologist. Reasons for image analysis failure may include but not limited to: 1) light counterstain; 2) crushed tissue; 3) presence of tar in NSCLC tissues; 4) staining of hemosiderin; or 5) presence of melanin that precludes evaluation. The pathologist visual immune score is the percentage of positive immune cells within the annotated region (to mimic the algorithm).

[0374] LAG3 IHC Assay Validation - Sensitivity A sensitivity analysis was performed using the optimized LAG3 (mouse clone 17B4) IHC assay on 247 FFPE human tissues (40 bladder urothelial cancer, 41 gastric/GEJ cancer, 41 HNSCC, 41 melanoma, 41 NSCLC, 43 RCC) tissues to demonstrate the dynamic range of the assay within the 6 indications. All specimens were evaluated by image analysis of 1 ROI (tumor + intervening stroma) and a subset

of the tissues (10 each within the 6 indications) were also evaluated by pathologist visual immune score.

[0375] On average, LAG-3 (mouse clone 17B4) expression was highest in melanoma (3.54%), followed by bladder urothelial cancer (2.58%), NSCLC (1.68%), HNSCC (1.47%), Gastric/GEJ cancer (1.27%), and RCC (1.24%). Positivity ranged from 0.01% to 25.57% with an average of 1.95% and a median of 0.84%. Using a threshold of 2% demonstrated 192 negative and 55 positive tissues (12 bladder urothelial, 6 gastric/GEJ cancer, 7 HNSCC, 18 melanoma, 8 NSCLC, and 4 RCC).

[0376] Figure 1 shows anti-LAG-3 staining patterns observed in the tumor samples using monoplex IHC. The staining patterns observed included partial membrane/cytoplasmic localization, dot like localization, and complete membrane/cytoplasmic localization.

[0377] Figure 2 shows the frequency distribution of LAG-3 positive cell as a ratio of total tumor cells across various tumors as detected by monoplex LAG-3 IHC.

EXAMPLE 2

Initial Efficacy of Anti-Lymphocyte Activation Gene-3 (Anti-LAG-3; BMS-986016) in Combination With Nivolumab in Patients With melanoma Previously Treated With Anti-PD1/PD-L1 Therapy

[0378] Simultaneous blockade of the negative T-cell regulators LAG-3 and PD-1 may function synergistically to restore T-cell activation and enhance antitumor immunity. Data from a phase 1/2a study of BMS-986016 (fully human IgG4 mAb that targets LAG-3) ± nivolumab (fully human IgG4 mAb that targets PD-1) demonstrated that the combination was well tolerated and showed promising antitumor activity in patients with melanoma who were refractory to or relapsed during prior anti–PD-1/PD-L1 therapy (NCT01968109; Ascierto et al. *J Clin Oncol.* 2017;35(suppl) [abstract 9520]). Below is efficacy data in patients with advanced melanoma who progressed on prior anti–PD-1/PD-L1 therapy.

[0379] This was a phase I/IIa, open label, dose escalation and cohort expansion study evaluating the safety, tolerability, and efficacy of BMS-986016 administered alone or in combination with nivolumab in patients with advanced solid tumors. Patients received study therapy intravenously once every two weeks for up to twelve 8-week treatment cycles. Combination dose for expansion was BMS-986016 80 mg + nivolumab 240 mg.

[0380] Study design and endpoints are shown in Figures 3 and 17.

[0381] Key eligibility criteria for patients in the melanoma prior IO expansion cohort are shown in Figure 3.

[0382] Results. As of the April 7, 2017 data cut-off, 212 patients were treated, including 55 patients with melanoma who progressed on prior anti-PD1/PD-L1 therapy (mel prior IO). Of the 212 patients, 61% were still on treatment at data cut-off. Of the 83 patients that discontinued treatment, the primary reason was disease progression (86%). Of the mel prior IO cohort, 67% of patients had M1C disease without brain metastasis, 15% had lactate dehydrogenase (LDH) $\geq 2 \times$ upper limit of normal (ULN), and 20% had liver metastasis. Figure 4.

[0383] Patients in the mel prior IO cohort were heavily pretreated. Figure 5. Of 55 patients, 76% had ≥ 2 prior therapies; 40 % of patients had progressive disease (PD) as best response to prior anti-PD1/PD-L1 therapy.

[0384] Figure 6 shows the LAG-3 expression status of the first 40 IO experienced melanoma samples. 40% (16/40) of the samples were scored as LAG-3 positive using $\geq 1\%$ cut-off in a monoplex IHC assay.

[0385] Efficacy in the melanoma prior IO cohort. Median duration of follow-up for all efficacy-evaluable patients (n = 48; all progressed on prior anti–PD-1/PD-L1 therapy) was 14 weeks (range, 4.1–41 weeks). Response by investigator assessment is shown in Figure 7. Overall response rate (ORR) was 13% and 6 patients had PR (2 of who had PD as best response to prior anti-PD1/PD-L1 therapy). 15 patients had reduction in tumor burden from baseline; reduction > 30% was observed in 7 patients (Figure 8). As shown in Figure 8, LAG-3 expression enriches for response. Figure 9 shows the depth and duration of response LEG-3 $\geq 1\%$, LAG-3 $< 1\%$, and LAG-3 unknown patients.

[0386] Figure 10 shows the duration of progression-free survival. Of 48 evaluable patients, 46% (22/48) of patients remain on treatment without progression at data cutoff.

[0387] As shown in Figure 11, there was nearly a 3-fold increase in ORR for patients with LAG-3 expression $\geq 1\%$ (20%) vs LAG-3 expression $< 1\%$ (7.1%). PD-L1 expression did not appear to enrich for response.

[0388] Updated results from the clinical trial are shown in Figures 16-23. As of August 2017, 262 patients were treated, including 68 patients with melanoma who progressed on prior anti-PD1/PD-L1 therapy (mel prior IO). Updated baseline demographics and disease characteristics are shown in Figure 17. Of the mel prior IO cohort, 68% of patients had M1C

disease without brain metastasis, 13% had lactate dehydrogenase (LDH) ≥ 2 × upper limit of normal (ULN), and 25% had liver metastasis.

[0389] Figure 18 shows the updated prior treatment history of the mel prior IO cohort. Of 68 patients, 77% had ≥ 2 prior therapies; 46 % of patients had progressive disease (PD) as best response to prior anti-PD1/PD-L1 therapy. Most patients (57%) also received prior anti–CTLA-4 therapy. 46% of patients had a best response of PD to prior anti–PD-1/PD-L1 therapy.

[0390] Figure 19 shows the updated efficacy data for the mel prior IO cohort. ORR was 11.5% and DCR was 49%. LAG-3 expression (≥ 1%) appeared to enrich for response. Median duration of response was not reached (range, 0.1+–39.3+).

[0391] Figure 20 shows the response by baseline characteristics and LAG-3 expression observed in the mel prior IO cohort. LAG-3 expression (≥ 1%) enriched for response irrespective of PD-L1 expression.

[0392] Figures 21 and 22 show the best change in target lesion size by LAG-3 and PD-L1 expression and the depth and duration of response by LAG-3 and PD-L1 expression, respectively, observed in the mel prior IO cohort. Responses were more likely in patients with LAG-3 expression ≥ 1%. PD-L1 expression did not appear to enrich for response.

[0393] Figure 23 shows the duration of progression-free survival. Of 61 evaluable patients, 34% (21/61) of patients had not progressed at data cutoff. Of 33 evaluable LAG-3 ≥ 1% patients, 55% (18/33) of patients had not progressed at data cutoff. Of 20 evaluable LAG-3 < 1% patients, 5% (1/20) of patients had not progressed at data cutoff.

EXAMPLE 3

Preliminary efficacy and biomarker enrichment across several advanced solid tumors in a Phase 1/2a study of a combination of anti-LAG-3 and anti-PD-1 monoclonal antibody

[0394] LAG-3 is a transmembrane receptor that negatively regulates T-cell activation. Signaling through LAG-3 and other T-cell inhibitory receptors, including programmed death-1 (PD-1), can lead to T-cell exhaustion and is a mechanism of immune escape for tumors. Simultaneous blockade of LAG-3 and PD-1 may function synergistically to restore T-cell activation and enhance antitumor immunity. In a phase 1/2a study, BMS-986016 (IgG4 mAb targeting LAG-3) ± nivolumab (IgG4 mAb targeting PD-1) demonstrated tolerability, peripheral T-cell activation, and preliminary clinical activity (NCT01968109; Lipson et al. *J Immunother Cancer*. 2016;4(suppl):173 [abstract P232]). Efficacy of BMS-986016 + nivolumab across

several advanced solid tumor expansion cohorts was evaluated in both all-comer and biomarker-enriched populations.

[0395] All patients (n = 204 as of April 7, 2017) were treated with BMS-986016 80 mg + nivolumab 240 mg Q2W in 56-day cycles until disease progression, confirmed complete response, completion of 12 cycles, or prohibitive toxicity. Most cohorts focused on immuno-oncology–naive patients with progression on/after at least 1 other prior therapy and included patients with advanced gastric/gastroesophageal junction cancer, squamous cell carcinoma of the head and neck, hepatocellular carcinoma, renal cell carcinoma, and NSCLC. One other cohort included patients with NSCLC who progressed on/after prior anti–PD-1/PD-L1 as their most recent therapy. Biomarker-defined patient subsets were described based on PD-L1 and LAG-3 immunohistochemical scoring in tumor biopsies.

[0396] Figure 12 shows LAG-3 expression status of immuno-oncology–naive gastric tumor samples. 48% (10/21) of the samples were scored as LAG-3 positive using a ≥1% cut-off in a monoplex IHC assay.

[0397] Figure 13 shows change in target lesion size in immuno-oncology–naive gastric cancer patients in response to treatment with a combination of anti-LAG-3 and anti-PD-1 antibody. LAG-3 positive tumors were enriched among the patients that were responsive to the treatment. Tumor response was determined according to RECIST. The group of patients shown have not been previously exposed to anti-PD-1/PD-L1 treatment.

[0398] Figure 14 shows LAG-3 expression status of immuno-oncology–naive SCCHN, renal carcinoma, HCC, and NSCLC tumor samples as determined by a monoplex IHC assay.

EXAMPLE 4

Multitumor Profiling of LAG-3 and Association With Immune Cell Phenotypes

[0399] LAG-3 negatively regulates T-cell activation. Sierro S et al. *Expert Opin Ther Targets*. 15:91–101 (2011); Grosso JF et al. *J Clin Invest*. 117:3383–3392 (2007). LAG-3 and programmed death-1 (PD-1) receptors are overexpressed and co-expressed on tumor-infiltrating lymphocytes (TILs). Goding SR et al. *J Immunol*. 190:4899–4909 (2013). LAG-3 and PD-1 overexpression may limit treatment response to anti-PD-1 therapy and lead to tumor progression. Ascierto P et al. Poster 9520 presented at the 53rd Annual Meeting of the American Society of Clinical Oncology; June 2–6, 2017; Chicago, IL; Wherry, *Nat Immunol*. 12(6):492–9 (2011); Woo SR et al. Cancer Res. 72:917–927 (2012); Huang CT et al. Immunity. 21:503–513 (2004). BMS-986016 is a fully human IgG4 antibody that targets LAG-3, blocking binding to its ligand,

major histocompatibility complex class II (MHC II) (Figure 24). Huard B et al. *Proc Natl Acad Sci U S A*. 94:5744–5749 (1997). BMS-986016 combined with nivolumab (anti-PD-1) may restore T-cell activation and tumor response in patients whose disease progressed on anti-PD-1 monotherapy. Ascierto P et al. Poster 9520 presented at the 53rd Annual Meeting of the American Society of Clinical Oncology; June 2–6, 2017; Chicago, IL. This dual inhibition may also enhance the durability of response in patients not previously treated with anti-PD-1 therapy. Simultaneous blockade of LAG-3 and PD-1 by BMS-986016 and nivolumab, respectively, produced peripheral T-cell activation and showed clinical activity and manageable safety in patients with advanced solid tumors. Ascierto P et al. Poster 9520 presented at the 53rd Annual Meeting of the American Society of Clinical Oncology; June 2–6, 2017; Chicago, IL; Lipson E et al. J Immunother Cancer. 4(suppl 1):173 (2016). To further understand the association between LAG-3 and markers of resistance across tumors, a comprehensive profiling of commercially sourced tumor specimens to investigate and characterize expression of LAG-3 and MHC II in the context of inflammatory biomarkers has been performed.

Methods

[0400] **Quantitative Immunohistochemistry (IHC)** Solid tumor specimens were profiled from patients with renal cell carcinoma (RCC), gastric carcinoma, non-small cell lung carcinoma (NSCLC), melanoma, squamous cell carcinoma of the head and neck (SCCHN), and urothelial carcinoma. Slide sections were stained by IHC for LAG-3, CD8, FOXP3, CD68, CD163, PD-L1, and MHC II using the Leica Bond Rx or Dako Link 48 platforms. For immune cell markers (LAG-3, CD8, FOXP3, CD68, CD163), the percent positivity was determined using Aperio image analysis software by defining the proportion of total nucleated cells expressing the biomarker in the tumor microenvironment. MHC II and PD-L1 expression by IHC on tumor cells were scored manually. Unsupervised clustering (Ward's method) was performed on the IHC data to identify associations between LAG-3 and other immune biomarkers. To determine MHC II+ and LAG-3+ colocalization, MHC II-high (>70% MHC II+) or MHC II-low (<10% MHC II+) tumor cell regions were assessed for the number of LAG-3 stained cells (average of three 20× fields of view each for positive and negative regions).

[0401] **mRNA Analysis** In patients with RCC and melanoma, changes in LAG-3 mRNA levels were determined by differential gene expression analyses of Affymetrix (RCC) or RNA-sequencing (melanoma) data from tumor biopsy samples collected at screening and 2–4 weeks post-immunotherapy initiation.

[0402] **Statistical Analyses** Correlations between LAG-3 expression and other immune biomarkers were assessed by Spearman's correlation, r. Mann–Whitney test was conducted to assess statistical differences. Differential gene expression analyses were performed using generalized linear models that included treatment group and time as factors..

Results

[0403] **LAG-3 Expression in Tumors.** For tumor specimens analyzed across 6 different solid tumor types (n = 245: RCC, 43; gastric, 41; NSCLC, 41; melanoma, 40; SCCHN, 40; urothelial, 40) a range of low to high LAG-3 expression was observed (0.01% to 33% of total nucleated cells). LAG-3 expression may be localized to the perinuclear, membrane, or cytoplasmic regions of lymphocytes, as shown by IHC staining (Figure 25).

[0404] **LAG-3 Association With Immune and Inflammatory Biomarkers.** A moderate correlation of LAG-3 expression with CD8, FOXP3, CD163, and CD68 (n = 237: RCC, 43; gastric, 39; NSCLC, 39; melanoma, 39; SCCHN, 40; urothelial, 37) was observed (Figure 26A–D, r = 0.49–0.65); no correlation of LAG-3 with PD-L1 and MHC II tumor expression was observed (Figure 26E and 26F, r = 0.28–0.30). MHC II expression in tumor cells (≥1%) was frequently observed, ranging from a low of 55% (melanoma) to a high of 82% (gastric carcinoma).

[0405] Tumors with ≥1% MHC II expression in tumor cells showed a significant increase in the frequency of LAG-3+ TILs (Figure 27, n = 241: RCC, 43; gastric, 40; NSCLC, 40; melanoma, 38; SCCHN, 40; urothelial, 40).

[0406] Unsupervised clustering of samples by tumor type revealed clusters of tumors with a range of inflammation from low to high in the 6 tumor types analyzed (examples in Figure 28A, urothelial carcinoma, n = 37; and 28B, gastric carcinoma, n = 39).

[0407] Increased MHC II tumor expression was frequently observed in inflammation high tumors, but was also observed in tumors with lower levels of inflammation (example in Figure 28A, urothelial carcinoma). Of those specimens that stained positively for tumor-cell MHC II expression, the level of MHC II expression was correlated with the level of LAG-3+ TILs in some tumor types (examples in Figure 28A and 298, urothelial and gastric carcinoma). The majority of tumors with high MHC II expression had low PD-L1 expression (Figure 28C, n = 229: RCC, 43; gastric, 39; NSCLC, 38; melanoma, 33; SCCHN, 39; urothelial, 37).

[0408] **Heterogeneous MHC II Tumor Cell Expression and LAG-3+ TILs.** In a subset of tumor specimens tested (n = 6), heterogeneous MHC II tumor cell expression was observed,

ranging from low (<10%) to high (>70%) (Figure 29A, urothelial carcinoma, n = 4; gastric carcinoma, n = 2). In this subset, a significant increase in the number of LAG-3+ TILs was observed in tumor regions with high MHC II expression vs low MHC II expression (Figure 29A-C).

[0409] **Changes in LAG-3 mRNA Level During Anti-PD-1 Monotherapy.** In an analysis of tissue samples from patients with metastatic melanoma (NCT01621490/CheckMate 038) or metastatic RCC (NCT01358721/ CheckMate 009), a significant increase in LAG-3 mRNA levels between screening and week 2–4 of treatment with nivolumab was observed (Figure 30).

[0410] LAG-3 expression was associated with cellular inflammation in the tumor microenvironment, as shown by IHC. MHC II tumor cell expression was frequently observed across the 6 tumor types analyzed; LAG-3 expression in immune cells was enriched in tumors with expression of MHC II in tumor cells. Higher frequency of LAG-3+ TILs was observed in MHC II high/positive tumor regions vs MHC II low/negative tumor regions within individual tumor specimens, raising the possibility that co-localization of LAG-3 and MHC II expression in tumor cells may serve as a mechanism of LAG-3 checkpoint activation in certain tumors. These findings, and the observation that nivolumab may induce LAG-3 expression, support the use of LAG-3 as a predictive biomarker for BMS-986016 therapy in patients whose disease progressed following treatment with anti-PD-1 therapy.

SEQUENCES

SEQ ID NO:1 Heavy Chain Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

QVQLQQWGAGLLKPSETLSLTCAVYGGSFSDYYWNWIRQPPGKGLEWIGEINHRGSTNSNPSLKS
RVTLSLDTSKNQFSLKLRSVTAADTAVYYCAFGYSDYEYNWFDPWGQGTLVTVSSASTKGPSVFP
LAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSS
LGTKTYTCNVDHKPSNTKVDKRVESKYGPPCPPCPAPEFLGGPSVFLFPPKPKDTLMISRTPEVT
CVVVDVSQEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSN
KGLPSSIEKTISKAKGQPREPQVYTLPPSQEEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENN
YKTTPPVLDSDGSFFLYSRLTVDKSRWQEGNVFSCSVMHEALHNHYTQKSLSLSLGK

SEQ ID NO:2 Light Chain Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

EIVLTQSPATLSLSPGERATLSCRASQSISSYLAWYQQKPGQAPRLLIYDASNRATGIPARFSGS
GSGTDFTLTISSLEPEDFAVYYCQQRSNWPLTFGQGTNLEIKRTVAAPSVFIFPPSDEQLKSGTA
SVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYACE
VTHQGLSSPVTKSFNRGEC

SEQ ID NO:3 Heavy Chain Variable Region (VH) Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

QVQLQQWGAGLLKPSETLSLTCAVYGGSFSDYYWNWIRQPPGKGLEWIGEINHRGSTN
SNPSLKSRVTLSLDTSKNQFSLKLRSVTAADTAVYYCAFGYSDYEYNWFDPWGQGTLV
TVSS

SEQ ID NO:4 Heavy Chain Variable Region (VH) Nucleotide Sequence; Anti-LAG-3 mAb (BMS-986016)

caggtgcagctacagcagtggggcgcaggactgttgaagccttcggagaccctgtccctcacctg
cgctgtctatggtgggtccttcagtgattactactggaactggatccgccagcccccagggaagg
ggctggagtggattggggaaatcaatcatcgtggaagcaccaactccaacccgtccctcaagagt
cgagtcaccctatcactagacacgtccaagaaccagttctccctgaagctgaggtctgtgaccgc
cgcggacacggctgtgtattactgtgcgtttggatatagtgactacgagtacaactggttcgacc
cctggggccagggaaccctggtcaccgtctcctca

SEQ ID NO:5 Light Chain Variable Region (VL) Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

EIVLTQSPATLSLSPGERATLSCRASQSISSYLAWYQQKPGQAPRLLIYDASNRATGIPARFSGS
GSGTDFTLTISSLEPEDFAVYYCQQRSNWPLTFGQGTNLEIK

SEQ ID NO:6 Light Chain Variable Region (VL) Nucleotide Sequence; Anti-LAG-3 mAb (BMS-986016)

gaaattgtgttgacacagtctccagccaccctgtctttgtctccaggggaaagagccaccctctc
ctgcagggccagtcagagtattagcagctacttagcctggtaccaacagaaacctggccaggctc
ccaggctcctcatctatgatgcatccaacagggccactggcatcccagccaggttcagtggcagt

gggtctgggacagacttcactctcaccatcagcagcctagagcctgaagattttgcagtttatta
ctgtcagcagcgtagcaactggcctctcacttttggccaggggaccaacctggagatcaaa

SEQ ID NO:7 Heavy Chain CDR1 Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

DYYWN

SEQ ID NO:8 Heavy Chain CDR2 Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

EINHRGSTNSNPSLKS

SEQ ID NO:9 Heavy Chain CDR3 Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

GYSDYEYNWFDP

SEQ ID NO:10 Light Chain CDR1 Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

RASQSISSYLA

SEQ ID NO:11 Light Chain CDR2 Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

DASNRAT

SEQ ID NO:12 Light Chain CDR3 Amino Acid Sequence; Anti-LAG-3 mAb (BMS-986016)

QQRSNWPLT

SEQ ID NO:13 Human LAG-3 Amino Acid Sequence

MWEAQFLGLLFLQPLWVAPVKPLQPGAEVPVVWAQEGAPAQLPCSPTIPLQDLSLLRRAGVTWQH
QPDSGPPAAAPGHPLAPGPHPAAPSSWGPRPRRYTVLSVGPGGLRSGRLPLQPRVQLDERGRQRG
DFSLWLRPARRADAGEYRAAVHLRDRALSCRLRLRLGQASMTASPPGSLRASDWVILNCSFSRPD
RPASVHWFRNRGQGRVPVRESPHHHLAESFLFLPQVSPMDSGPWGCILTYRDGFNVSIMYNLTVL
GLEPPTPLTVYAGAGSRVGLPCRLPAGVGTRSFLTAKWTPPGGGPDLLVTGDNGDFTLRLEDVSQ
AQAGTYTCHIHLQEQQLNATVTLAIITVTPKSFGSPGSLGKLLCEVTPVSGQERFVWSSLDTPSQ
RSFSGPWLEAQEAQLLSQPWQCQLYQGERLLGAAVYFTELSSPGAQRSGRAPGALPAGHLLLFLT
LGVLSLLLLVTGAFGFHLWRRQWRPRRFSALEQGIHPPQAQSKIEELEQEPEPEPEPEPEPEPEP
EPEQL

SEQ ID NO:14 LAG-3 Epitope

PGHPLAPG

SEQ ID NO:15 LAG-3 Epitope

HPAAPSSW

SEQ ID NO:16 LAG-3 Epitope

PAAPSSWG

SEQ ID NO:17 Heavy Chain Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

QVQLVESGGGVVQPGRSLRLDCKASGITFSNSGMHWVRQAPGKGLEWVAVIWYDGSKRYYADSVK
GRFTISRDNSKNTLFLQMNSLRAEDTAVYYCATNDDYWGQGTLVTVSSASTKGPSVFPLAPCSRS
TSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTKTYT
CNVDHKPSNTKVDKRVESKYGPPCPPCPAPEFLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVS
QEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKGLPSSI
EKTISKAKGQPREPQVYTLPPSQEEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPV
LDSDGSFFLYSRLTVDKSRWQEGNVFSCSVMHEALHNHYTQKSLSLSLGK

SEQ ID NO:18 Light Chain Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

EIVLTQSPATLSLSPGERATLSCRASQSVSSYLAWYQQKPGQAPRLLIYDASNRATGIPARFSGS
GSGTDFTLTISSLEPEDFAVYYCQQSSNWPRTFGQGTKVEIKRTVAAPSVFIFPPSDEQLKSGTA
SVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYSLSSTLLSKADYEKHKVYACEV
THQGLSSPVTKSFNRGEC

SEQ ID NO:19 Heavy Chain Variable Region (VH) Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

QVQLVESGGGVVQPGRSLRLDCKASGITFSNSGMHWVRQAPGKGLEWVAVIWYDGSKRYYADSVK
GRFTISRDNSKNTLFLQMNSLRAEDTAVYYCATNDDYWGQGTLVTVSS

SEQ ID NO:20 Heavy Chain Variable Region (VH) Nucleotide Sequence; Anti-PD-1 mAb (BMS936558)

caggtgcagctggtggagtctgggggaggcgtggtccagcctgggaggtccctgagactcgactg
taaagcgtctggaatcaccttcagtaactctggcatgcactgggtccgccaggctccaggcaagg
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agccgaggacacggctgtgtattactgtgcgacaaacgacgactactggggccagggaaccctgg
tcaccgtctcctca

SEQ ID NO:21 Light Chain Variable Region (VL) Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

EIVLTQSPATLSLSPGERATLSCRASQSVSSYLAWYQQKPGQAPRLLIYDASNRATGIPARFSGS
GSGTDFTLTISSLEPEDFAVYYCQQSSNWPRTFGQGTKVEIK

SEQ ID NO:22 Light Chain Variable Region (VL) Nucleotide Sequence; Anti-PD-1 mAb (BMS936558)

gaaattgtgttgacacagtctccagccaccctgtctttgtctccaggggaaagagccaccctctc
ctgcagggccagtcagagtgttagtagttacttagcctggtaccaacagaaacctggccaggctc
ccaggctcctcatctatgatgcatccaacagggccactggcatcccagccaggttcagtggcagt
gggtctgggacagacttcactctcaccatcagcagcctagagcctgaagattttgcagtttatta
ctgtcagcagagtagcaactggcctcggacgttcggccaagggaccaaggtggaaatcaaa

SEQ ID NO:23 Heavy Chain CDR1 Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

NSGMH

SEQ ID NO:24 Heavy Chain CDR2 Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

VIWYDGSKRYYADSVKG

SEQ ID NO:25 Heavy Chain CDR3 Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

NDDY

SEQ ID NO:26 Light Chain CDR1 Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

RASQSVSSYLA

SEQ ID NO:27 Light Chain CDR2 Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

DASNRAT

SEQ ID NO:28 Light Chain CDR3 Amino Acid Sequence; Anti-PD-1 mAb (BMS936558)

QQSSNWPRT

SEQ ID NO:29 Complete Homo sapiens PD-1 sequence

agtttcccttccgctcacctccgcctgagcagtggagaaggcggcactctggtggggctgctcca
ggcatgcagatcccacaggcgccctggccagtcgtctgggcggtgctacaactgggctggcggcc
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ggccagcaggcacctgagtggctgggacaagggatccccccttccctgtggttctattatattata
attataattaaatatgagagcatgct

SEQ ID NO:30 Heavy Chain Nucleotide Sequence; Anti-LAG-3 mAb (BMS-986016)

caggtgcagctacagcagtggggcgcaggactgttgaagccttcggagaccctgtccctcacctg
cgctgtctatggtgggtccttcagtgattactactggaactggatccgccagcccccagggaagg
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cgagtcaccctatcactagacacgtccaagaaccagttctccctgaagctgaggtctgtgaccgc
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SEQ ID NO:31 Light Chain Nucleotide Sequence; Anti-LAG-3 mAb (BMS-986016)

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SEQ ID NO:32 Motif

MYPPPY

What is claimed is:

1. A method of selecting a malignant tumor in a human patient for immunotherapy, comprising:

   (a) determining the level of LAG-3 expression in a tumor sample; and

   (b) selecting the tumor for immunotherapy if the tumor is a LAG-3 positive tumor.

2. A method of identifying a malignant tumor in a human patient as eligible for immunotherapy, comprising:

   (a) determining the level of LAG-3 expression in a tumor sample; and

   (b) identifying the tumor as eligible for immunotherapy if the tumor is a LAG-3 positive tumor.

3. A method of identifying a malignant tumor in a human patient that is likely to be responsive to an immunotherapy, the method comprising:

   (a) determining the level of LAG-3 expression in a tumor sample; and

   (b) identifying the tumor as likely to be responsive to treatment if the tumor is a LAG-3 positive tumor.

4. A method of classifying a malignant tumor in a human patient as likely to be responsive to an immunotherapy, the method comprising:

   (a) determining the level of LAG-3 expression in a tumor sample; and

   (b) classifying the tumor as likely to be responsive to immunotherapy if the tumor is a LAG-3 positive tumor.

5. The method of any one of claims 1 to 4, further comprising determining the level of PD-L1 expression in the tumor sample.

6. The method of any one of claims 1 to 5, wherein the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor.

7. The method of any one of claims 1 to 5, wherein the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor.

8. The method of any one of claims 1 to 5, wherein the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor.

9. The method of any one of claims 1 to 5, wherein the immunotherapy comprises contacting the tumor with a therapeutically effective amount of an anti-CTLA-4 antibody.

10. The method of any one of claims 1 to 5, wherein the immunotherapy comprises contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor.

11. The method of any one of claims 1 to 5, comprising contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor.

12. The method of any one of claims 1 to 5, comprising contacting the tumor with a therapeutically effective amount of a LAG-3 inhibitor.

13. The method of any one of claims 1 to 5, comprising contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor.

14. The method of any one of claims 1 to 5, comprising contacting the tumor with a therapeutically effective amount of an anti-CTLA-4 antibody.

15. The method of any one of claims 1 to 5, comprising contacting the tumor with a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor.

16. The method of any one of claims 1 to 5, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor.

17. The method of any one of claims 1 to 5, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor.

18. The method of any one of claims 1 to 5, comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor.

19. The method of any one of claims 1 to 5, comprising administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody.

20. The method of any one of claims 1 to 5, comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor.

21. A method of identifying a patient with a malignant tumor who is likely to respond to an immunotherapy, the method comprising:

(a) determining the level of LAG-3 expression in a tumor sample; and

(b) identifying the patient who is likely to respond to treatment if the tumor is a LAG-3 positive tumor.

22. A method of selecting a patient with a malignant tumor for immunotherapy, the method comprising:

(a) determining the level of LAG-3 expression in a tumor sample; and

(b) selecting the patient for immunotherapy if the tumor is a LAG-3 positive tumor.

23. The method of claim 21 or claim 22, further comprising determining the level of PD-L1 expression in the tumor sample.

24. The method of any one of claims 21 to 23, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor.

25. The method of any one of claims 21 to 23, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor.

26. The method of any one of claims 21 to 23, comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor.

27. The method of any one of claims 21 to 23, comprising administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody.

28. The method of any one of claims 21 to 23, comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor.

29. A method of treating a malignant tumor in a human patient, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor; wherein the patient is predicted to respond to treatment with the LAG-3 inhibitor and PD-1 pathway inhibitor based upon LAG-3 expression in a sample of the patient's tumor.

30. A method of treating a malignant tumor in a human patient, comprising: administering to the patient a therapeutically effective amount of a LAG-3 inhibitor; wherein the patient is predicted to respond to treatment with the LAG-3 inhibitor based upon LAG-3 expression in a sample of the patient's tumor.

31. A method of treating a malignant tumor in a human patient, comprising: administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor; wherein the patient is predicted to respond to treatment with the PD-1 pathway inhibitor based upon LAG-3 expression in a sample of the patient's tumor.

32. A method of treating a malignant tumor in a human patient, comprising: administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody; wherein the patient is predicted to respond to treatment with the anti-CTLA-4 antibody based upon LAG-3 expression in a sample of the patient's tumor.

33. A method of treating a malignant tumor in a human patient, comprising: administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor; wherein the patient is predicted to respond to treatment with the PD-1 pathway inhibitor and an immune checkpoint inhibitor based upon LAG-3 expression in a sample of the patient's tumor.

34. The method of any one of claims 29 to 33, wherein the patient is predicted to respond to the treatment based upon LAG-3 and PD-L1 expression in a sample of the patient's tumor.

35. A method of treating a malignant tumor in a human patient in need thereof, comprising:

(a) determining the level of LAG-3 expression in a tumor sample; and

(b) administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor if the tumor is a LAG-3 positive tumor.

36. A method of treating a malignant tumor in a human patient in need thereof, comprising:

(a) determining the level of LAG-3 expression in a tumor sample; and

(b) administering to the patient a therapeutically effective amount of a LAG-3 inhibitor if the tumor is a LAG-3 positive tumor.

37. A method of treating a malignant tumor in a human patient in need thereof, comprising:

(a) determining the level of LAG-3 expression in a tumor sample; and

(b) administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor if the tumor is a LAG-3 positive tumor.

38. A method of treating a malignant tumor in a human patient in need thereof, comprising:

(a) determining the level of LAG-3 expression in a tumor sample; and

(b) administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody if the tumor is a LAG-3 positive tumor.

39. A method of treating a malignant tumor in a human patient in need thereof, comprising:

(a) determining the level of LAG-3 expression in a tumor sample; and

(b) administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor if the tumor is a LAG-3 positive tumor.

40. The method of any one of claim 35 to 39, further comprising determining the level of PD-L1 expression in the tumor sample.

41. A method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration.

42. A method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor,

wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration.

43. A method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration.

44. A method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration.

45. A method for treating a malignant tumor in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration.

46. The method of any one of claims 41 to 45, wherein the patient is identified as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration.

47. The method of any one of claims 41 to 45, wherein the patient is identified as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration

48. A method for treating a malignant tumor in a human patient in need thereof, comprising:

(a) identifying the patient as having a LAG-3 positive malignant tumor; and

(b) administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor.

49. A method for treating a malignant tumor in a human patient in need thereof, comprising:

(a) identifying the patient as having a LAG-3 positive malignant tumor; and

(b) administering to the patient a therapeutically effective amount of a LAG-3 inhibitor.

50. A method for treating a malignant tumor in a human patient in need thereof, comprising:

(a) identifying the patient as having a LAG-3 positive malignant tumor; and

(b) administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor.

51. A method for treating a malignant tumor in a human patient in need thereof, comprising:

(a) identifying the patient as having a LAG-3 positive malignant tumor; and

(b) administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody.

52. A method for treating a malignant tumor in a human patient in need thereof, comprising:

(a) identifying the patient as having a LAG-3 positive malignant tumor; and

(b) administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor.

53. The method of any one of claims 48 to 52, further comprising identifying the patient as having a LAG-3 positive, PD-L1 positive malignant tumor.

54. The method of any one of claims 48 to 52, further comprising identifying the patient as having a LAG-3 positive, PD-L1 negative malignant tumor.

55. A method for extending a progression-free survival period for over 12 months in a human patient afflicted with a malignant tumor comprising administering to the patient a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months.

56. A method for extending a progression-free survival period for over 12 months in a human patient afflicted with a malignant tumor comprising administering to the patient a LAG-3 inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months.

57. A method for extending a progression-free survival period for over 12 months in a human patient afflicted with a malignant tumor comprising administering to the patient a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months.

58. A method for extending a progression-free survival period for over 12 months in a human patient afflicted with a malignant tumor comprising administering to the patient an anti-CTLA-4 antibody, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months.

59. A method for extending a progression-free survival period for over 12 months in a human patient afflicted with a malignant tumor comprising administering to the patient a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months.

60. The method of any one of claims 55 to 59, wherein the patient is identified as having a LAG-3 positive, PD-L1 positive malignant tumor prior to the administration.

61. The method of any one of claims 55 to 59, wherein the patient is identified as having a LAG-3 positive, PD-L1 negative malignant tumor prior to the administration

62. The method of any one of claims 55 to 61, wherein the progression-free survival of the patient is extended after the administration for over about 13 months, about 14 months, about 15 months, about 16 months, about 17 months, about 18 months, about 2 years, about 3 years, about 4 years, about 5 years, about 6 years, about 7 years, about 8 years, about 9 years, or about 10 years.

63. The method of claim 62, wherein the progression-free survival of the patient is extended for over 14 months.

64. A method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration.

65. A method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration.

66. A method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration.

67. A method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration.

68. A method for reducing a tumor size at least by 10% in a human patient afflicted with a malignant tumor comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein the patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the administration reduces the tumor size at least about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, or 100% compared to the tumor size prior to the administration.

69. The method of any one of claims 64 to 68, wherein the patient is identified as having a LAG-3 positive, PD-L1 positive malignant tumor prior to the administration.

70. The method of any one of claims 64 to 68, wherein the patient is identified as having a LAG-3 positive, PD-L1 negative malignant tumor prior to the administration.

71. The method of any one of claims 55 to 70, further comprising identifying the patient as having a LAG-3 positive malignant tumor prior to the administration.

72. The method of any one of claims 55 to 71, further comprising identifying the patient as having a LAG-3 positive, PD-L1 positive malignant tumor prior to the administration.

73. The method of any one of claims 55 to 71, further comprising identifying the patient as having a LAG-3 positive, PD-L1 negative malignant tumor prior to the administration.

74. The method of any one of claims 55 to 73, wherein the patient experiences (i) extended progression-free survival for over 12 months, (ii) tumor size reduction at least about 10%, about 20%, about 30%, about 40%, or about 50% compared to the tumor size prior to the administration, or (iii) both.

75. A method for increasing an objective response rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

76. A method for increasing an objective response rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

77. A method for increasing an objective response rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

78. A method for increasing an objective response rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

79. A method for increasing an objective response rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

80. A method for increasing a disease control rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

81. A method for increasing a disease control rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

82. A method for increasing a disease control rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

83. A method for increasing a disease control rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of an anti-CTLA-4 antibody, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

84. A method for increasing a disease control rate to a cancer treatment to be higher than about 50% in a human patient population, each of whom is afflicted with a malignant tumor, comprising administering to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than about 55%, about 60%, about 65%, about 70%, or about 75%.

85. The method of any one of claims 75 to 84, wherein each patient is identified as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration.

86. The method of any one of claims 75 to 84, wherein each patient is identified as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration .

87. The method of any one of claims 75 to 86, wherein the median duration of response is ≥ 3 month, ≥ 6 month, ≥ 12 month, or ≥ 18 month.

88. The method of any one of claims 75 to 87, further comprising identifying each patient of the patient population as having a LAG-3 positive malignant tumor prior to the administration.

89. The method of any one of claims 75 to 88, further comprising identifying each patient of the patient population as having a LAG-3 positive PD-L1 positive malignant tumor prior to the administration.

90. The method of any one of claims 75 to 88, further comprising identifying each patient of the patient population as having a LAG-3 positive PD-L1 negative malignant tumor prior to the administration.

91. The method of any one of claims 75 to 90, wherein each patient of the patient population is further characterized by (i) extended progression-free survival for over 12 months, (ii) tumor size reduction at least about 10%, about 20%, about 30%, about 40%, or about 50% compared to the tumor size prior to the administration, or (iii) both.

92. The method of any one of claims 75 to 91, wherein the patient population comprises at least about 100, 200, 300, 400, 500, 600, 700, 800, 900, or 1000 patients having a LAG-3 positive malignant tumor.

93. A method for selecting a human patient suitable for a combination therapy comprising:

(a) identifying a patient as having a LAG-3 positive malignant tumor; and

(b) instructing a healthcare provider to administer to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor.

94. A method for selecting a human patient suitable for a combination therapy comprising:

(a) identifying a patient as having a LAG-3 positive malignant tumor; and

(b) instructing a healthcare provider to administer to the patient a therapeutically effective amount of a LAG-3 inhibitor.

95. A method for selecting a human patient suitable for a combination therapy comprising:

(a) identifying a patient as having a LAG-3 positive malignant tumor; and

(b) instructing a healthcare provider to administer to the patient a therapeutically effective amount of a PD-1 pathway inhibitor.

96. A method for selecting a human patient suitable for a combination therapy comprising:

(a) identifying a patient as having a LAG-3 positive malignant tumor; and

(b) instructing a healthcare provider to administer to the patient a therapeutically effective amount of an anti-CTLA-4 antibody.

97. A method for selecting a human patient suitable for a combination therapy comprising:

(a) identifying a patient as having a LAG-3 positive malignant tumor; and

(b) instructing a healthcare provider to administer to the patient a therapeutically effective amount of a PD-1 pathway inhibitor and an immune checkpoint inhibitor.

98. The method of any one of claims 93 to 97, further comprising identifying the patient as having a LAG-3 positive, PD-L1 positive malignant tumor.

99. The method of any one of claims 93 to 97, further comprising identifying the patient as having a LAG-3 positive, PD-L1 negative malignant tumor.

100. The method of any one of claims 93 to 99, wherein the administration treats the malignant tumor.

101. The method of any one of claims 93 to 100, wherein identifying the patient as having a LAG-3 positive malignant tumor comprises determining LAG-3 expression in the malignant tumor.

102. The method of any one of claims 93 to 101, wherein identifying the patient as having a LAG-3 positive PD-L1 positive malignant tumor comprises determining PD-L1 expression in the malignant tumor.

103. The method of any one of claims 93 to 101, wherein identifying the patient as having a LAG-3 positive PD-L1 negative malignant tumor comprises determining PD-L1 expression in the malignant tumor.

104. The method of any one of claims 93 to 103, wherein LAG-3 expression is determined by reviewing the results of an assay capable of determining LAG-3 expression.

105. The method of any one of claims 93 to 103, wherein LAG-3 expression is determined by reviewing the results of an immunohistochemistry assay capable of detecting LAG-3 expression.

106. The method of any one of claims 93 to 105, wherein PD-L1 expression is determined by reviewing the results of an assay capable of determining PD-L1 expression.

107. The method of any one of claims 93 to 105, wherein PD-L1 expression is determined by reviewing the results of an immunohistochemistry assay capable of detecting PD-L1 expression.

108. The method of any one of claims 1 to 107, wherein a LAG-3 positive tumor comprises at least about 1%, at least about 2%, at least about 3%, at least about 4%, at least about 5%, at least about 7%, at least about 10%, at least about 15%, at least about 20%, at least about 25%, at least about 30%, at least about 40%, at least about 50%, at least about 60%,

at least about 70%, at least about 80%, at least about 90%, or 100% cells expressing LAG-3.

109. The method of any one of claims 1 to 107, wherein a LAG-3 positive tumor comprises at least about 1% cells expressing LAG-3.

110. The method of any one of claims 1 to 107, wherein a LAG-3 positive tumor comprises at least about 5% cells expressing LAG-3.

111. The method of any one of claims 108 to 110, wherein the cells expressing LAG-3 comprise tumor infiltrating lymphocytes.

112. The method of any one of claims 109-110, wherein the cells expressing LAG-3 are the total number of cells.

113. The method of any one of claims 109-112, wherein the cells express LAG-3 on the cell surface.

114. The method of any one of claims 1 to 113, wherein the malignant tumor is selected from the group consisting of a liver cancer, bone cancer, pancreatic cancer, skin cancer, oral cancer, cancer of the head or neck, breast cancer, lung cancer, including small cell and non-small cell lung cancer, cutaneous or intraocular malignant melanoma, renal cancer, uterine cancer, ovarian cancer, colorectal cancer, colon cancer, rectal cancer, cancer of the anal region, stomach cancer, testicular cancer, uterine cancer, carcinoma of the fallopian tubes, carcinoma of the endometrium, carcinoma of the cervix, carcinoma of the vagina, carcinoma of the vulva, non-Hodgkin's lymphoma, cancer of the esophagus, cancer of the small intestine, cancer of the endocrine system, cancer of the thyroid gland, cancer of the parathyroid gland, cancer of the adrenal gland, sarcoma of soft tissue, cancer of the urethra, cancer of the penis, cancers of the childhood, lymphocytic lymphoma, cancer of the bladder, cancer of the kidney or ureter, carcinoma of the renal pelvis, neoplasm of the central nervous system (CNS), primary CNS lymphoma, tumor angiogenesis, spinal axis tumor, brain stem glioma, pituitary adenoma, Kaposi's sarcoma, epidermoid cancer, squamous cell cancer, environmentally induced cancers including those induced by asbestos, hematologic malignancies including, for example, multiple myeloma, B-cell lymphoma, Hodgkin lymphoma/primary mediastinal B-cell lymphoma, non-Hodgkin's lymphomas, acute myeloid lymphoma, chronic myelogenous leukemia, chronic lymphoid leukemia, follicular lymphoma, diffuse large B-cell lymphoma, Burkitt's lymphoma, immunoblastic large cell lymphoma, precursor B-lymphoblastic lymphoma, mantle cell lymphoma, acute lymphoblastic leukemia, mycosis fungoides, anaplastic large cell lymphoma, T-cell lymphoma, and precursor T-lymphoblastic lymphoma, and any combination thereof.

115. The method of any one of claims 1 to 113, wherein the malignant tumor is chosen from melanoma, non-small cell lung cancer (NSCLC), human papilloma virus (HPV)-related tumor, and gastric adenocarcinoma.

116. The method of any one of claims 1 to 113, wherein the malignant tumor is non-small cell lung cancer (NSCLC), a virally-related cancer related tumor, or gastric adenocarcinoma.

117. The method of any one of claims 1 to 113, wherein the malignant tumor is melanoma, gastric cancer, gastroesophageal junction cancer, non-small cell lung cancer, bladder cancer, head and neck squamous cell carcinoma, or renal cell cancer.

118. The method of any one of claims 1 to 113, wherein the malignant tumor is lung cancer, melanoma, squamous cell carcinoma of the head and neck, renal cancer, gastric cancer, or hepatocellular carcinoma.

119. The method of any one of claims 1 to 113, wherein the LAG-3 positive malignant tumor is a melanoma tumor comprising about 1% or more cells expressing LAG-3.

120. The method of any one of claims 1 to 113, wherein the LAG-3 positive malignant tumor is a gastric cancer tumor comprising about 1% or more cells expressing LAG-3.

121. The method of any one of claims 1 to 120, wherein the malignant tumor is refractory to treatment with an immune checkpoint inhibitor.

122. The method of any one of claims 1 to 121, wherein the malignant tumor is refractory to treatment with an anti-PD-1 antibody.

123. The method of any one of claims 1 to 122, wherein the malignant tumor is refractory to treatment with an anti-PD-L1 antibody.

124. A method for treating melanoma in a human patient, comprising:

(a) identifying the patient as having a LAG-3 positive melanoma; and

(b) administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor.

125. The method of claim 124, wherein identifying the patient as having a LAG-3 positive melanoma comprises determining LAG-3 expression in the melanoma tumor.

126. The method of claim 125, wherein LAG-3 expression is determined by reviewing the results of an assay capable of determining LAG-3 expression.

127. The method of claim 125, wherein LAG-3 expression is determined by an immunohistochemistry assay capable of detecting LAG-3 expression.

128. The method of any one of claims 124 to 127, further comprising identifying the patient as having a LAG-3 positive PD-L1 positive malignant tumor.

129. The method of any one of claims 124 to 127, further comprising identifying the patient as having a LAG-3 positive PD-L1 negative malignant tumor.

130. A method for treating a melanoma in a human patient in need thereof comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive melanoma prior to the administration.

131. A method for extending a progression-free survival period to over 12 months in a human patient afflicted with a melanoma comprising administering to the patient a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein the patient is identified as having a LAG-3 positive melanoma prior to the administration and wherein the patient demonstrates progression-free survival for over 12 months.

132. The method of claim 130 or claim 131, wherein the patient is identified as having a LAG-3 positive, PD-L1 positive melanoma prior to the administration.

133. The method of claim 130 or claim 131, wherein the patient is identified as having a LAG-3 positive, PD-L1 negative melanoma prior to the administration.

134. A method for increasing an objective response rate to a cancer treatment to be higher than 15% in a human patient population, each of whom is afflicted with melanoma, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive malignant tumor prior to the administration and wherein the objective response rate is higher than 15%.

135. A method for increasing a disease control rate to a cancer treatment to be higher than 70% in a human patient population, each of whom is afflicted with melanoma, comprising administering to the patient a therapeutically effective amount of a LAG-3 inhibitor and a PD-1 pathway inhibitor, wherein each patient is identified as having a LAG-3 positive melanoma prior to the administration and wherein the objective response rate is higher than 70%.

136. The method of claim 134 or claim 135, further comprising identifying each patient of the patient population as having a LAG-3 positive melanoma prior to the administration.

137. The method of any one of claims 134 to 136, wherein the median duration of response is $\geq$ 3 month, $\geq$ 6 month, $\geq$ 12 month, or $\geq$ 18 month.

138. The method of any one of claims 134 to 137, wherein each patient is identified as having a LAG-3 positive, PD-L1 positive melanoma prior to the administration.

139. The method of any one of claims 134 to 137, wherein each patient is identified as having a LAG-3 positive, PD-L1 negative melanoma prior to the administration.

140. The method of any one of claims 124 to 139, wherein the melanoma is refractory to treatment with an immune checkpoint inhibitor.

141. The method of any one of claims 124 to 140, wherein the melanoma is refractory to treatment with an anti-PD-1 antibody or an anti-PD-L1 antibody.

142. The method of any one of claims 1 to 141, wherein determining the level of LAG-3 and/or PD-L1 expression comprises providing a test tissue sample obtained from the patient, the test tissue sample comprising tumor cells and/or tumor-infiltrating immune cells.

143. The method of claim 142, wherein the test tissue sample is a tumor biopsy.

144. The method of claim 142 or claim 143, wherein the test tissue sample is a formalin-fixed paraffin embedded (FFPE) sample.

145. The method of any one of claims 1 to 144, wherein determining comprises detecting LAG-3 and/or PD-L1 protein or RNA expression in the test tissue sample.

146. The method of claim 145, wherein LAG-3 and/or PD-L1 expression is detected by an assay capable of detecting the level of LAG-3 and/or PD-L1 protein, respectively, in the test tissue sample.

147. The method of claim 142, wherein LAG-3 and/or PD-L1 expression is detected by an immunohistochemistry assay.

148. The method of claim 147, wherein the immunohistochemistry assay is a monoplex assay.

149. The method of claim 147, wherein the immunohistochemistry assay is a multiplex assay.

150. The method of any one of claims 147 to 149, wherein the immunohistochemistry assay comprises contacting the tumor sample with the 17B4, SP346, 11E3, 874501, or EPR4392(2) anti-human LAG-3 monoclonal antibody.

151. The method of any one of claims 147 to 149, wherein the immunohistochemistry assay comprises contacting the tumor sample with an anti-LAG-3 antibody comprising heavy and light chain variable regions comprising the sequences set forth in SEQ ID NOs:3 and 5, respectively.

152. The method of any one of claims 145 to 151, wherein the immunohistochemistry assay uses a black or brown chromogen.

153. The method of any one of claims 145 to 151, wherein the immunohistochemistry assay uses a red chromogen.

154. The method of any one of claims 145 to 151, wherein the immunohistochemistry assay uses a blue chromogen.

155. The method of any one of claims 145 to 151, wherein the immunohistochemistry assay uses a green chromogen.

156. The method of any one of claims 145 to 151, wherein the immunohistochemistry assay uses a purple chromogen.

157. The method of any one of claims 145 to 156, wherein the immunohistochemistry assay is scored at a low magnification.

158. The method of claim 145, wherein low magnification is about 20X.

159. The method of any one of claims 145 to 156, wherein the immunohistochemistry assay is scored at high magnification.

160. The method of claim 159, wherein high magnification is about 40X.

161. The method of any one of claims 145 to 160, wherein the immunohistochemistry assay is scored by an image analysis software.

162. The method of any one of claims 145 to 160, wherein the immunohistochemistry assay is scored by pathologist visual immune score.

163. The method of any one of claims 145 to 160, wherein the immunohistochemistry assay is scored manually.

164. The method of any one of claims 145 to 163, wherein scoring the immunohistochemistry assay comprises assessing the proportion of cells in the test tissue sample that express LAG-3 and/or assessing the proportion of cells in the test tissue sample that express PD-L1.

165. The method of any one of claims 145 to 163, wherein scoring the immunohistochemistry assay comprises assessing the proportion of tumor cells in the test tissue sample that express LAG-3 and/or assessing the proportion of tumor cells in the test tissue sample that express PD-L1.

166. The method of any one of claims 145 to 163, wherein scoring the immunohistochemistry assay comprises assessing the proportion of immune cells in the test tissue sample that express LAG-3 and/or assessing the proportion of immune cells in the test tissue sample that express PD-L1.

167. The method of any one of claims 145 to 163, wherein scoring the immunohistochemistry assay comprises assessing the proportion of T cells in the test tissue sample that express LAG-3 and/or assessing the proportion of T cells in the test tissue sample that express PD-L1.

168. The method of any one of claims 145 to 163, wherein scoring the immunohistochemistry assay comprises assessing the proportion of CD8+ T cells in the test tissue sample that express LAG-3 and/or assessing the proportion of CD8+ T cells in the test tissue sample that express PD-L1.

169. The method of any one of claims 145 to 163, wherein scoring the immunohistochemistry assay comprises assessing the proportion of CD4+ T cells in the test tissue sample that express LAG-3 and/or assessing the proportion of CD4+ T cells in the test tissue sample that express PD-L1.

170. The method of any one of claims 145 to 163, wherein scoring the immunohistochemistry assay comprises assessing the proportion of FOXP3+ T cells in the test tissue sample that express LAG-3 and/or assessing the proportion of FOXP3+ T cells in the test tissue sample that express PD-L1.

171. The method of any one of claims 145 to 170, wherein cells with partial membrane/cytoplasmic LAG-3 localization are scored as LAG-3 expressing cells.

172. The method of any one of claims 145 to 170, wherein cells with dot-like LAG-3 localization are scored as LAG-3 expressing cells.

173. The method of any one of claims 145 to 170, wherein cells with complete membrane/cytoplasmic LAG-3 localization are scored as LAG-3 expressing cells.

174. The method of any one of claims 145 to 170, wherein cells with any LAG-3 localization pattern are scored as LAG-3 expressing cells.

175. The method of any one of claims 145 to 174, wherein the immunohistochemistry assay is a multiplex assay that further comprises detecting the expression of MHC Class II by the tumor cells.

176. The method of any one of claims 145 to 175, wherein scoring the immunohistochemistry assay comprises assessing the proportion of cells in the test tissue sample that expresses MHC Class II.

177. The method of any one of claims 145 to 175, wherein scoring the immunohistochemistry assay comprises assessing the proportion of non-immune cells in the test tissue sample that expresses MHC II.

178. The method of claim 144, wherein LAG-3 and/or PD-L1 protein expression is detected by flow cytometry.

179. The method of claim 178, wherein the test tissue sample obtained from the patient comprises tumor infiltrating immune cells.

180. The method of claim 179, wherein the malignant tumor is a hematological malignancy and the tissue sample comprises circulating lymphocytes.

181. The method of any one of claims 178 to 180, wherein the flow cytometry is a multiplex assay.

182. The method of claim 181, wherein the flow cytometry comprises detecting the expression of markers comprising LAG-3, PD-L1, CD4, CD8, FOXP3, and any combination thereof.

183. The method of claim 182, wherein scoring the flow cytometry comprises assessing the proportion of T cells in the test tissue sample that express LAG-3.

184. The method of claim 182, wherein scoring the flow cytometry comprises assessing the proportion of CD8+ T cells in the test tissue sample that express LAG-3.

185. The method of claim 182, wherein scoring the flow cytometry comprises assessing the proportion of CD4+ T cells in the test tissue sample that express LAG-3.

186. The method of claim 182, wherein scoring the flow cytometry comprises assessing the proportion of FOXP3+ T cells in the test tissue sample that express LAG-3.

187. The method of claim 143, wherein LAG-3 and/or PD-L1 expression is detected by an assay capable of detecting the level of LAG-3 and/or PD-L1, respectively, RNA in the tumor sample.

188. The method of claim 187, wherein LAG-3 and/or PD-L1 expression is detected by an RT-PCR based assay.

189. The method of claim 134, wherein scoring the RT-PCR based assay comprises assessing the level of LAG-3 and/or PD-L1 RNA expression in the test tissue sample relative to a predetermined level.

190. The method of any one of claims 1 to 189, wherein the LAG-3 inhibitor is an anti-LAG-3 antibody or antigen-binding fragment thereof.

191. The method of claim 190, wherein the anti-LAG-3 antibody is a bispecific antibody.

192. The method of claim 190 or 191, wherein the anti-LAG-3 antibody or antigen-binding fragment thereof comprises (a) a heavy chain variable region CDR1 comprising the sequence set forth in SEQ ID NO:7; (b) a heavy chain variable region CDR2 comprising the sequence set forth in SEQ ID NO:8; (c) a heavy chain variable region CDR3 comprising the sequence set forth in SEQ ID NO:9; (d) a light chain variable region CDR1 comprising the sequence set forth in SEQ ID NO:10; (e) a light chain variable region CDR2 comprising the sequence set forth in SEQ ID NO:11; and (f) a light chain variable region CDR3 comprising the sequence set forth in SEQ ID NO:12.

193. The method of any one of claims 190 to 192, wherein the anti-LAG-3 antibody or antigen-binding fragment thereof comprises heavy and light chain variable regions comprising the sequences set forth in SEQ ID NOs:3 and 5, respectively.

194. The method of claim 190, wherein the anti-LAG-3 antibody is MK-4280 (28G-10), REGN3767, GSK2837781, IMP731 (H5L7BW), BAP050, IMP-701 (LAG-525), IMP321, FS-118, Sym022, TSR-033, MGD013, FS118, or GSK2831781.

195. The method of any one of claims 1 to 194, wherein the PD-1 pathway inhibitor is an anti-PD-1 antibody or antigen-binding fragment thereof.

196. The method of claim 195, wherein the anti-PD-1 antibody or antigen-binding fragment thereof comprises (a) a heavy chain variable region CDR1 comprising the sequence set forth in SEQ ID NO:23; (b) a heavy chain variable region CDR2 comprising the sequence set forth in SEQ ID NO:24; (c) a heavy chain variable region CDR3 comprising the sequence set forth in SEQ ID NO:25; (d) a light chain variable region CDR1 comprising the sequence set forth in SEQ ID NO:26; (e) a light chain variable region CDR2 comprising the sequence set forth in SEQ ID NO:27; and (f) a light chain variable region CDR3 comprising the sequence set forth in SEQ ID NO:28.

197. The method of claim 196, wherein the anti-PD-1 antibody or antigen-binding fragment thereof comprises heavy and light chain variable regions comprising the sequences set forth in SEQ ID NOs:19 and 21, respectively.

198. The method of claim 197, wherein the anti-PD-1 antibody or antigen-binding fragment thereof comprises heavy and light chains comprising the sequences set forth in SEQ ID NOs:17 and 18, respectively.

199. The method of claim 195, wherein the anti-PD-1 antibody is pembrolizumab (KEYTRUDA; MK-3475), pidilizumab (CT-011), or nivolumab (OPDIVO; BMS-936558).

200. The method of any one of claims 1 to 194, wherein the PD-1 pathway inhibitor is an anti-PD-L1 antibody or antigen-binding fragment thereof.

201. The method of claim 200, wherein the anti-PD-L1 antibody is atezolizumab (Tecentriq or RG7446), durvalumab (Imfinzi or MEDI4736), avelumab (Bavencio) or BMS-936559.

202. The method of any one of claims 1 to 194, wherein the PD-1 pathway inhibitor is an anti-PD-L2 antibody or antigen-binding fragment thereof.

203. The method of any one of claims 1 to 194, wherein the anti-CTLA-4 antibody is ipilimumab or an antigen-binding fragment thereof.

204. The method of any one of claims 1 to 194, wherein the immune checkpoint inhibitor is a CTLA-4 antagonist, a CD80 antagonist, a CD86 antagonist, a Tim-3 antagonist, a TIGIT antagonist, a CD20 antagonist, a CD96 antagonist, a IDO1 antagonist, a STING antagonist, a GARP antagonist, a CD40 antagonist, A2aR antagonist, a CEACAM1 (CD66a) antagonist, a CEA antagonist, a CD47 antagonist a PVRIG antagonist, a TDO antagonist, a VISTA antagonist, or a KIR antagonist.

205. The method of any one of claims 190 to 204, wherein the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycle, four doses of the anti-LAG-3 antibody are administered at a dose of 3, 20, 80, 160, or 240 mg.

206. The method of any one of claims 190 to 205, wherein the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycle, four doses of the anti-PD-1 antibody are administered at a dose of 80 or 240 mg.

207. The method of any one of claims 190 to 205, wherein the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycle, four doses of the anti-PD-L1 antibody are administered at a dose of 3, 20, 80, 160, or 240 mg.

208. The method of any one of claims 190 to 207, wherein the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycle, four doses of the anti-CTLA-4 antibody are administered at a dose of 3, 20, 80, 160, or 240 mg.

209. The method of any one of claims 190 to 208, wherein the method comprises at least one administration cycle, wherein the cycle is a period of eight weeks, wherein for each of the at least one cycle, four doses of the anti-LAG-3 antibody are administered at a dose of 3,

20, 80, 160, or 240 mg and four doses of the anti-PD-1 antibody are administered at a dose of 80 or 240 mg.

210. The method of any one of claims 190 to 209, wherein the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the following doses: (a) 3 mg of anti-LAG-3 antibody and 80 mg of anti-PD-1 antibody; (b) 3 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody; (c) 20 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody; (d) 80 mg of anti-LAG-3 antibody and 160 mg of anti-PD-1 antibody; (e) 80 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody; (f) 160 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody, or (g) 240 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody.

211. The method of claim 210, wherein the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the dose of 80 mg of anti-LAG-3 antibody and 160 mg of anti-PD-1 antibody.

212. The method of claim 210, wherein the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the dose of 80 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody.

213. The method of claim 210, wherein the anti-LAG-3 antibody and anti-PD-1 antibody are administered at the dose of 160 mg of anti-LAG-3 antibody and 240 mg of anti-PD-1 antibody.

214. The method of any one of claims 190 to 213, wherein the anti-PD-1 and anti-LAG-3 antibodies or antigen-binding fragments thereof are formulated for intravenous administration.

215. The method of any one of claims 190 to 214, wherein the anti-PD-1 and anti-LAG-3 antibodies or antigen-binding fragments thereof are formulated together.

216. The method of any one of claims 190 to 214, wherein the anti-PD-1 and anti-LAG-3 antibodies or antigen-binding fragments thereof are formulated separately.

217. The method of any one of claims 205 to 216, wherein the treatment consists of up to 12 cycles.

218. The method of any one of claims 205 to 217, wherein anti-PD-1 antibody or antigen-binding fragment thereof is administered on Days 1, 15, 29, and 43 of each cycle.

219. The method of any one of claims 205 to 217, wherein anti-LAG-3 antibody or antigen-binding fragment thereof is administered on Days 1, 15, 29, and 43 of each cycle.

220. The method of any one of claims 205 to 217, wherein anti-CTLA-4 antibody or antigen-binding fragment thereof is administered on Days 1, 15, 29, and 43 of each cycle.

221. The method of any one of claims 205 to 217, wherein the anti-PD-1 antibody or antigen-binding fragment thereof is administered prior to administration of the anti-LAG-3 antibody or antigen-binding fragment thereof.

222. The method of any one of claims 190 to 221, wherein the anti-LAG-3 antibody or antigen-binding fragment thereof is administered within about 30 minutes prior to administration of the anti-PD-1 antibody or antigen-binding fragment thereof.

223. The method of any one of claims 190 to 222, wherein the anti-PD-1 antibody or antigen-binding fragment thereof is administered after administration of the anti-LAG-3 antibody or antigen-binding fragment thereof.

224. The method of any one of claims 190 to 222, wherein the anti-PD-1 antibody or antigen-binding fragment thereof is administered before administration of the anti-LAG-3 antibody or antigen-binding fragment thereof.

225. The method of any one of claims 190 to 222, wherein the anti-PD-1 antibody or antigen-binding fragment thereof is administered concurrently with the anti-LAG-3 antibody or antigen-binding fragment thereof.

226. The method of any one of claims 190 to 225, wherein the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor are administered as a first line of treatment.

227. The method of any one of claims 190 to 225, wherein the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor are administered as a second line of treatment.

228. The method of any one of claims 190 to 227, further comprising the administration of at least one additional therapeutic agent.

229. The method of claim 228, wherein the at least one additional therapeutic agent is a chemotherapeutic agent.

230. The method of claim 229, wherein the at least one additional therapeutic agent is an immune checkpoint inhibitor.

231. The method of any one of claims 1 to 230, wherein the method produces at least one therapeutic effect chosen from a reduction in size of a tumor, reduction in number of metastatic lesions over time, complete response, partial response, and stable disease.

232. The method of any one of claims 1 to 231, wherein administering the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor activates the patient's T cells.

233. The method of any one of claims 1 to 232, wherein administering the anti-LAG-3 antibody or antigen-binding fragment thereof and PD-1 pathway inhibitor induces the expression activation markers by the patient's T cells.

234. The method of any one of claims 1 to 233, wherein administering the anti-LAG-3 antibody or antigen-binding fragment thereof results in the occupancy of at least about 10%, at least about 20%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, at least about 95%, or about 100% of the LAG-3 receptors on the patient's T cells.

235. The method of claim 234, wherein the T cells are CD8+ T cells.

236. The method of claim 234, wherein the T cells are tumor infiltrating T cells.

237. The method of any one of claims 219 to 224, wherein the PD-1 pathway inhibitor comprises an anti-PD-1 antibody or antigen-binding fragment thereof.

238. A kit for treating a patient afflicted with a malignant tumor, the kit comprising:

(a) a dosage ranging from about 0.1 to about10 mg/kg body weight of an anti-LAG-3 antibody or an antigen-binding fragment thereof;

(b) a dosage ranging from about 0.1 to about 10 mg/kg body weight of an anti-PD-1 antibody or antigen-binding fragment thereof; and

(c) instructions for using the anti-LAG-3 antibody and anti-PD-1 antibody or antigen-binding fragments thereof in the method of any one of claims 1 to 237.

239. A kit for treating a patient afflicted with a malignant tumor, the kit comprising:

(a) a dosage ranging from about 0.1 to about 10 mg/kg body weight of an anti-PD1 antibody;

(b) a dosage an immune checkpoint inhibitor; and

(c) instructions for using the anti-PD-1 antibody or antigen-binding fragment thereof and immune checkpoint inhibitor in the method of any one of claims 1 to 237.

240. A kit for treating a patient afflicted with a malignant tumor, the kit comprising:

(a) a dosage ranging from about 0.1 to about 10 mg/kg body weight of an anti-LAG-3 antibody or antigen-binding fragment thereof; and

(b) instructions for using the anti-LAG-3 antibody or antigen-binding fragment thereof in the method of any one of claims 1 to 237.

241. A kit for treating a patient afflicted with a malignant tumor, the kit comprising:

(a) a dosage ranging from 0.1 to 10 mg/kg body weight of an anti-PD-1 antibody or antigen-binding fragment thereof; and

(b) instructions for using the anti-PD-1 antibody or antigen-binding fragment thereof in the method of any one of claims 1 to 237.

242. A method of identifying a patient that is refractory to treatment with a PD-1 antagonist, the method comprising determining the level of LAG-3 expression, wherein an increased level of LAG-3 expression following treatment with the PD-1 antagonist, relative to the level of LAG-3 expression prior to treatment with the PD-1 antagonist, indicates that a patient is refractory to PD-1 antagonist therapy.

243. A method of identifying a patient that is at risk of becoming refractory to treatment with a PD-1 antagonist, the method comprising determining the level of LAG-3 expression, wherein an increased level of LAG-3 expression following treatment with the PD-1 antagonist, relative to the level of LAG-3 expression prior to treatment with the PD-1 antagonist, indicates that a patient is at risk of becoming refractory to PD-1 antagonist therapy.

244. A method of identifying a patient who is likely to respond to a LAG-3 therapy, the method comprising determining the level of LAG-3 expression in the patient, wherein an increased level of LAG-3 expression following treatment with a PD-1 antagonist, relative to the level of LAG-3 expression prior to treatment with the PD-1 antagonist, indicates that a patient is likely to respond to a LAG-3 therapy.

245. A method of selecting a patient for treatment with a LAG-3 therapy, the method comprising determining the level of LAG-3 expression in the patient, wherein an increased level of LAG-3 expression following treatment with a PD-1 antagonist, relative to the level of LAG-3 expression prior to treatment with the PD-1 antagonist, indicates that a patient is likely to respond to a LAG-3 therapy.

246. The method of any one of claims 242-245, wherein the PD-1 antagonist is a PD-1 inhibitor.

247. The method of any one of claims 242-245, wherein the PD-1 antagonist is a PD-1 antibody or antigen-binding fragment thereof.

248. The method of any one of claims 244-245, wherein the LAG-3 therapy is a LAG-3 inhibitor.

249. The method of any one of claims 244, 245 or 248, wherein the LAG-3 therapy is an anti-LAG-3 antibody or antigen-binding fragment thereof.

250. The method of any one of claims 244 or 245, wherein the LAG-3 therapy is a combination therapy.

251. The method of claim 250, wherein the LAG-3 combination therapy is a combination of an anti-LAG-3 antibody or antigen-binding fragment thereof and an anti-PD-1 antibody or antigen-binding fragment thereof.

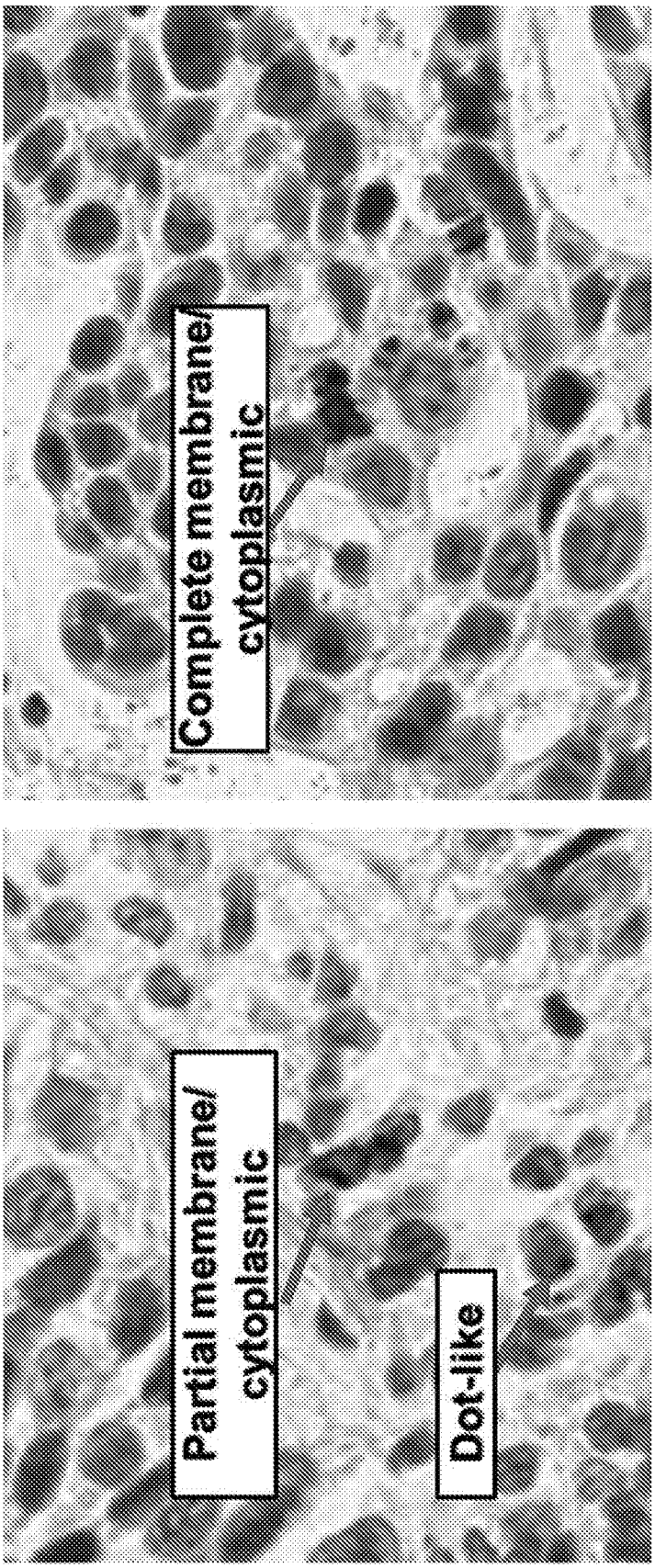

Complete membrane/
cytoplasmic
Partial membrane/
cytoplasmic
Dot-like

Figure 1

Figure 2

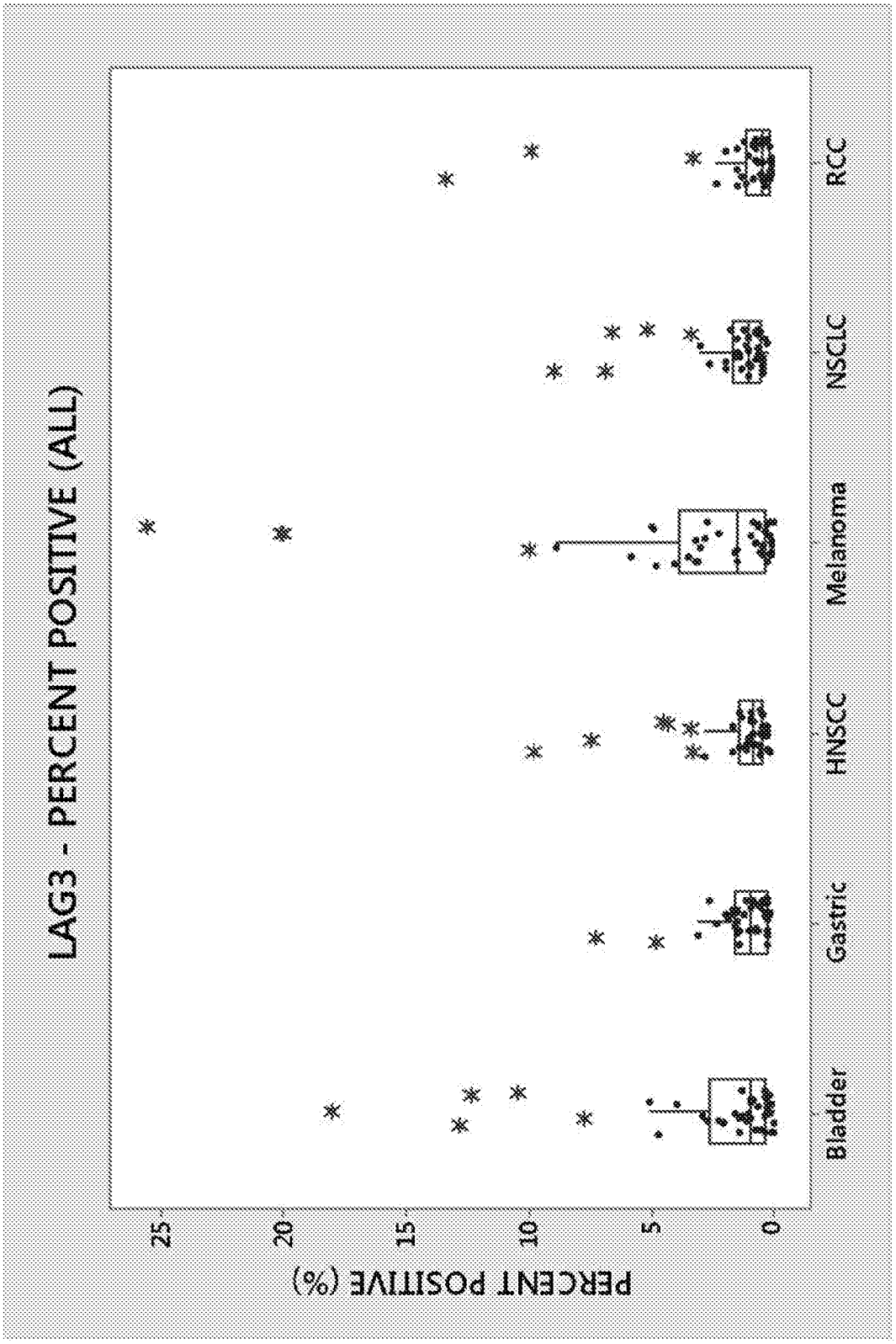
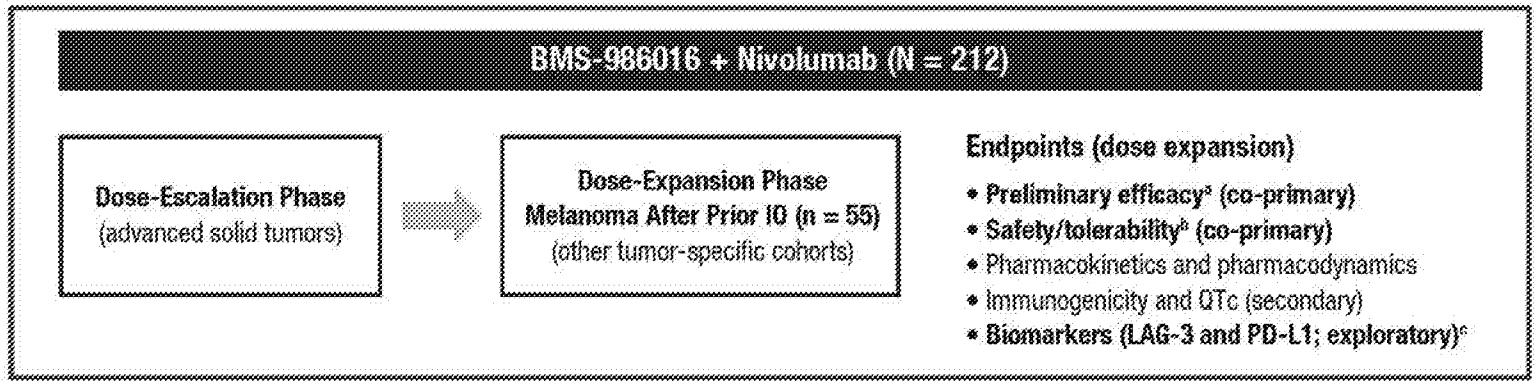

LAG3 - PERCENT POSITIVE (ALL)
PERCENT POSITIVE (%)
0
5
10
15
20
25
Bladder
Gastric
HNSCC
Melanoma
NSCLC
RCC

Figure 3A



[a]Tumor response evaluated per Response Evaluation Criteria in Solid Tumors (RECIST) v1.1[10] (investigator assessment). [b]Safety evaluated per Common Terminology Criteria for Adverse Events v4.0[11] during treatment and up to 135 days after discontinuation. [c]LAG-3 and PD-L1 expression (percent of positive cells within invasive margin, tumor, and stroma) evaluated using immunohistochemistry (IHC) assays on formalin-fixed, paraffin-embedded tumor sections. Immune cell LAG-3 expression (≥ 1% or < 1%) determined using mouse antibody clone 17B4; tumor cell PD-L1 expression (≥ 1% or < 1%) determined using Dako 28-8 PD-L1 IHC 28-8 kit.

Figure 3B

Key Eligibility Criteria for Patients in the Melanoma Prior-IO Expansion Cohort

Inclusion	Exclusion
At least 18 years of age and ECOG PS ≤ 1	Prior exposure to IOs other than anti–CTLA-4 and/or anti–PD-1/PD-L1 antibody therapy (eg, anti–PD-L2, anti–KIR, anti–CD137, and anti–OX40)
Advanced (metastatic and/or unresectable) melanoma	Uveal melanoma
Progression on/intolerance of ≥ 1 and ≤ 3 prior standard regimens	Uncontrolled brain metastases
Prior treatment with anti–PD-1/PD-L1 (± anti–CTLA-4 ± BRAF ± MEK inhibitors) required	Active autoimmune disease
Prior adjuvant or neoadjuvant therapy with cytokines (IL-2 or IFN) or anti–CTLA-4 antibodies allowed	
At least 1 measurable lesion per RECIST v1.1[10]	

CD137, cluster of differentiation 137; CTLA-4, cytotoxic T lymphocyte antigen-4; ECOG PS, Eastern Cooperative Oncology Group performance status; IFN, interferon; IL-2, interleukin-2; KIR, anti–killer-cell immunoglobulin-like receptor.

Figure 4

Table 1. **Baseline Demographics and Disease Characteristics**

	Mel Prior IO (n = 55)
Median age (range), years	59 (25–80)
< 65 years, n (%)	36 (65)
Male, n (%)	39 (71)
Race, n (%)	
White	52 (95)
Black	1 (1.8)
Asian	1 (1.8)
Other	1 (1.8)
ECOG PS, n (%)	
0	38 (69)
1	17 (31)
M stage at study entry, n (%)	
M0	7 (13)
M1A	6 (11)
M1B	4 (7.3)
M1C with brain metastasis	1 (1.8)
M1C without brain metastasis	37 (67)
LDH, n (%)	
Normal	25 (45)
Normal to < 2 × ULN	13 (24)
≥ 2 × ULN	8 (15)
Unknown	9 (16)
Liver metastases, n (%)	
Yes	11 (20)
No	44 (80)
BRAF status, n (%)	
Mutation	21 (38)
No mutation	31 (56)
Unknown	3 (5.5)

M, metastasis.

Figure 5

Table 2. Prior Therapy

Patients, n (%)	Mel Prior IO (n = 55)
Prior radiotherapy	16 (29)
Prior systemic therapy	54 (98)
Immunotherapy	54 (98)
Anti–CTLA-4[a]	32 (58)
Anti–PD-1/PD-L1[b]	52 (95)
Best response to prior anti–PD-1/PD-L1[c]	
CR	1 (1.8)
PR	12 (22)
SD	16 (29)
PD	22 (40)
BRAF inhibitors	16 (29)
MEK inhibitors	11 (20)
Number of systemic regimens	
1	12 (22)
2	17 (31)
≥ 3	25 (45)
Median (range)	2 (1–5)

CR, complete response; PR, partial response; SD, stable disease. [a]Two patients received anti–PD-1/PD-L1 + anti–CTLA-4, 5 patients received anti–CTLA-4 after anti–PD-1/PD-L1, and 29 patients received anti–CTLA-4 before anti–PD-1/PD-L1; prior anti–CTLA-4 therapy was not reported in 4 patients. [b]Twenty-five patients received nivolumab, 25 patients received pembrolizumab, and 2 patients received other therapies; 2 patients did not receive prior anti–PD-1/PD-L1; prior anti–PD-1/PD-L1 was unknown in 1 patient. [c]Response in 1 patient was reported as not applicable.

Figure 6

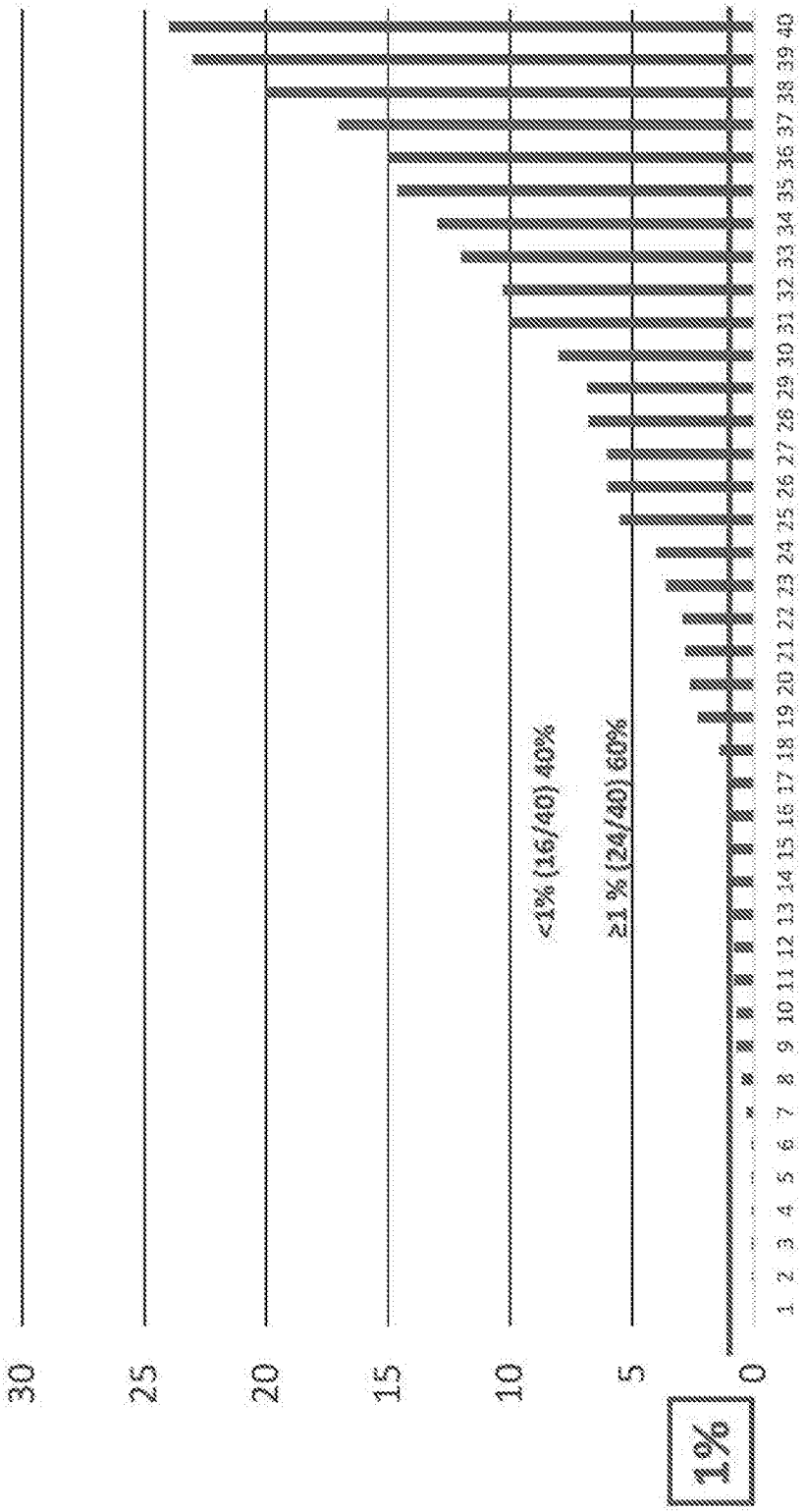

LAG-3 Monoplex % Positive: First 40 IO Experienced
Melanoma
30
25
20
15
10
5
0
1%
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40
<1% (16/40) 40%
≥1 % (24/40) 60%

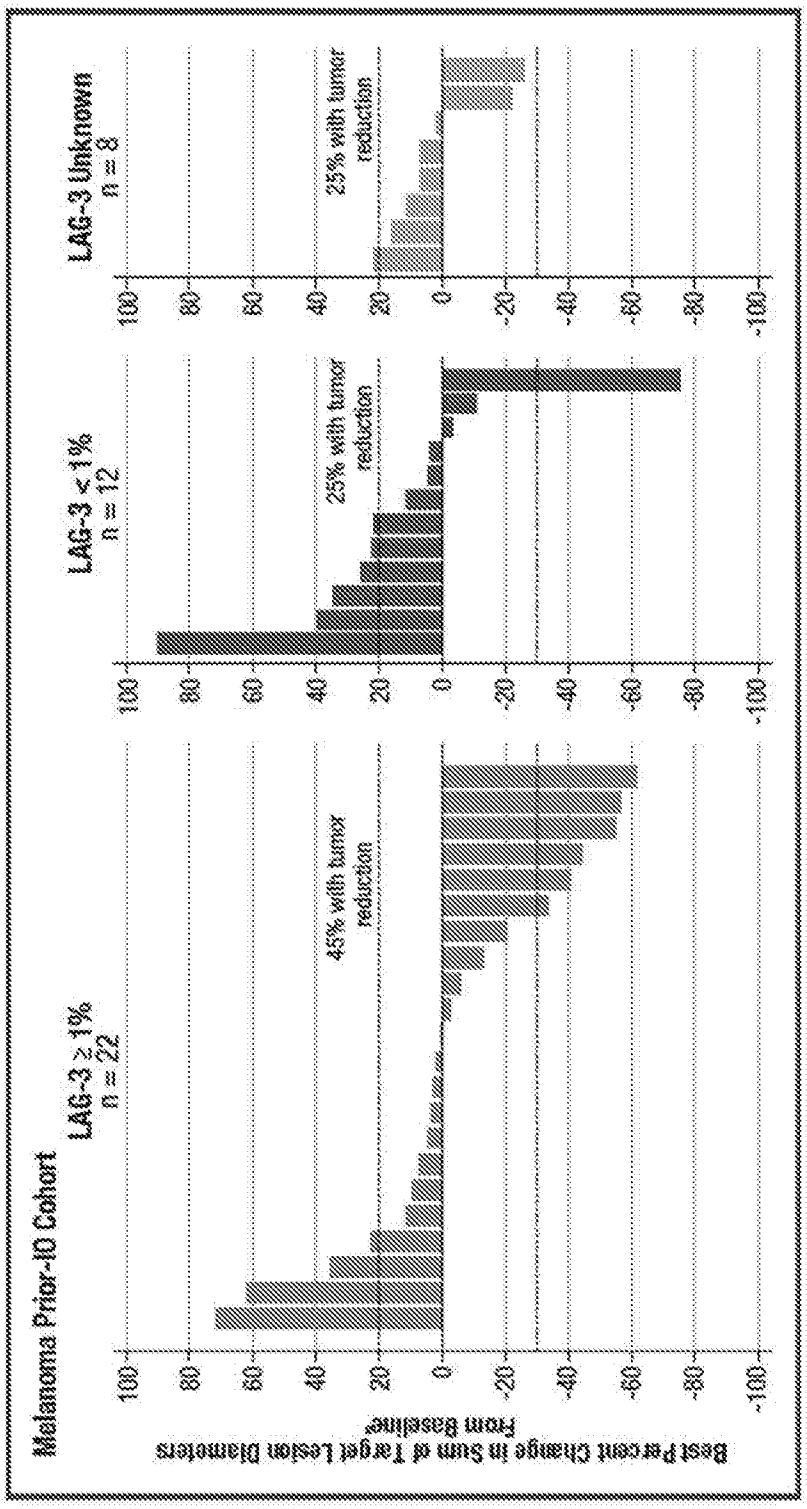
Figure 7

Table 3. **Preliminary Evidence of Antitumor Activity**

Patients, n (%)	Mel Prior IO (n = 49)
BOR	
CR	0
PR[a]	6 (12)
SD	20 (42)
PD	16 (33)
Clinical progression[c]	6 (13)
ORR, 95% CI[b]	6 (13), 4.7, 25
LAG-3 ≥ 1% (n = 25)	5 (20), 6.8, 41
LAG-3 < 1% (n = 14)	1 (7.1), 0.2, 34
DCR (CR + PR + SD)[b]	26 (54)
LAG-3 ≥ 1% (n = 25)	16 (64)
LAG-3 < 1% (n = 14)	5 (36)

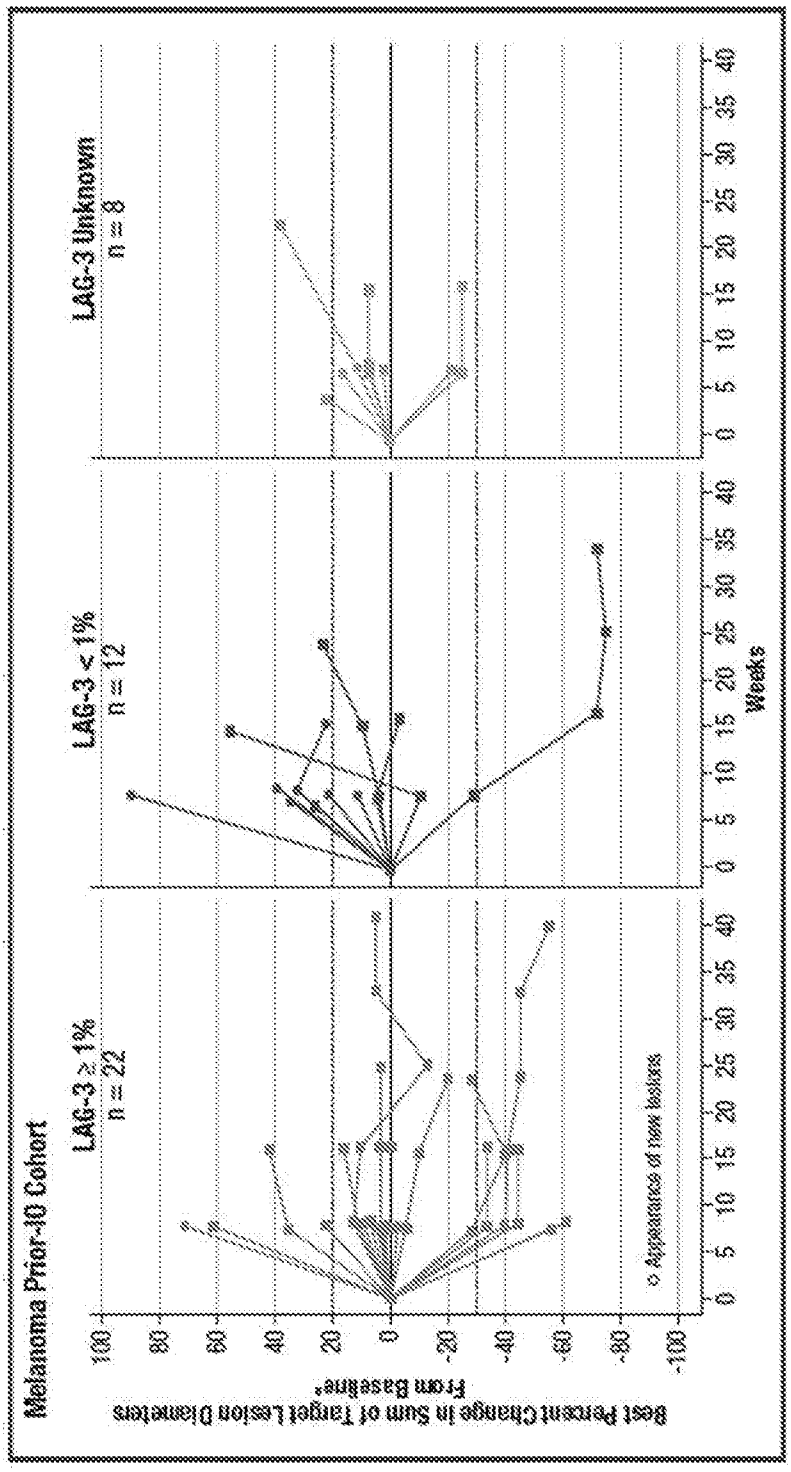
BOR, best overall response; DCR, disease control rate. [a]All response-evaluable patients; all progressed on prior anti-PD-1/PD-L1 therapy. [b]Two responses were unconfirmed. [c]Occurred prior to first radiographic scan.

Figure 8



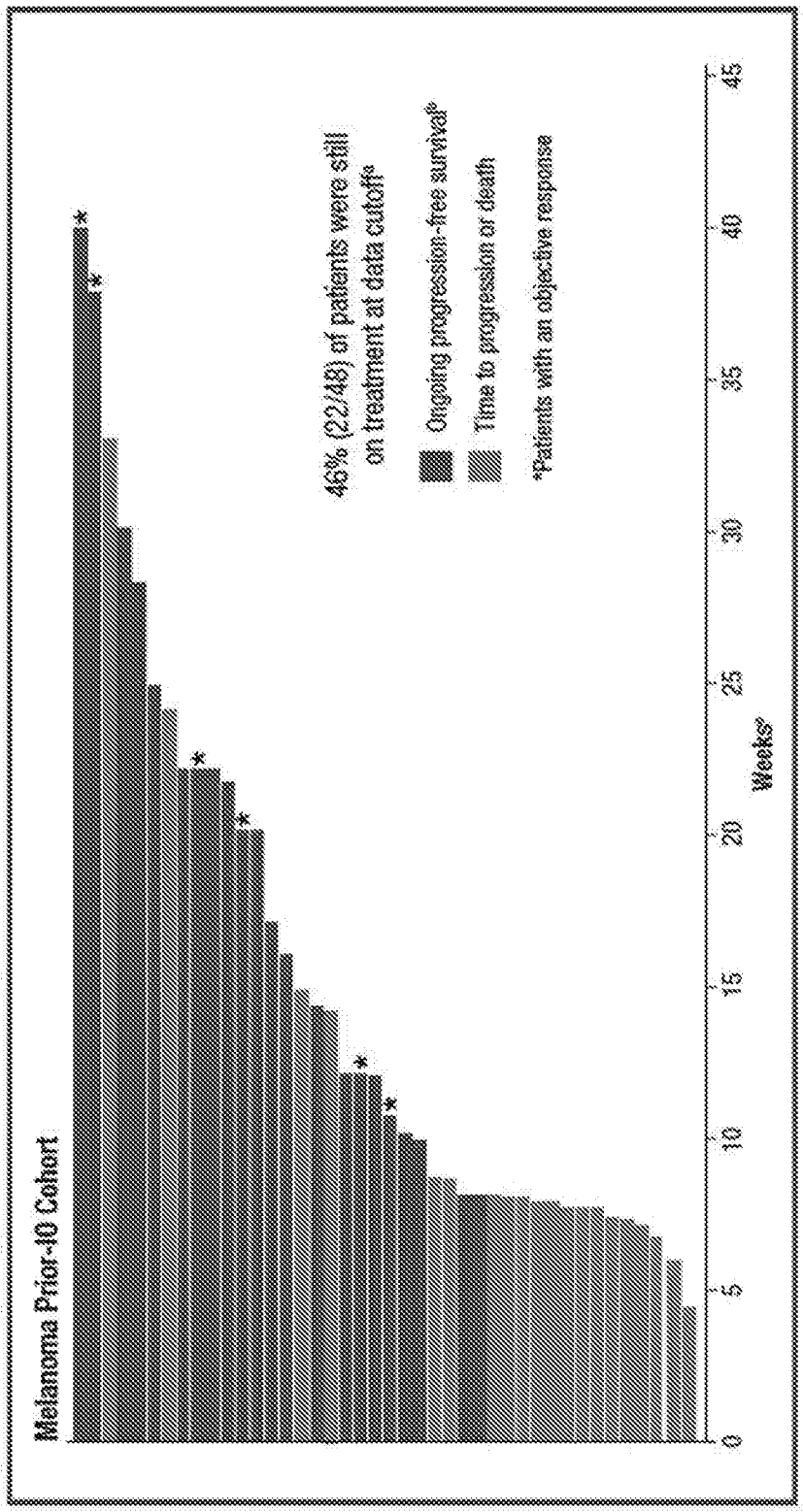
*Six patients had clinical progression prior to their first scan and are not included in the plot. One patient with best change from baseline > 30% had an unconfirmed best response of SD.

Figure 9



*Six patients had clinical progression prior to their first scan and are not included in the plot.

Figure 10



*Six patients had clinical progression prior to their first scan and are not included in the plot. *Censored on last visit. *Evaluations are planned for every 8 weeks.

Figure 11

Table 4. **Response by Baseline Characteristics (investigator assessed)**

	Mel Prior IO (n = 40*)		
		ORR	
	n	n (%)	95% CI
LAG-3 expression			
≥ 1%	25	5 (20)	6.8, 41
< 1%	14	1 (1.7)	0.2, 34
PD-L1 expression			
≥ 1%	16	2 (13)	1.6, 38
< 1%	19	4 (21)	6.1, 46
Response based upon prior BOR to anti–PD-1/PD-L1 only			
CR	1	0	–
PR	12	2 (17)	–
SD	14	1 (7.1)	–
CR + PR + SD	27	3 (11)	2.4, 29
PD	20	2 (10)	1.2, 32
M stage at study entry			
M1A	6	1 (17)	0.4, 64
M1B	3	0	0, 71
M1C with brain metastasis	1	0	0, 97.5
M1C without brain metastasis	32	4 (13)	3.5, 29
Liver metastases			
Yes	11	1 (9.1)	0.2, 41
No	37	5 (14)	4.5, 29
BRAF status			
Mutation	19	0	0, 18
No mutation	28	6 (21)	8.3, 41

*All response-evaluable patients; all progressed on prior anti–PD-1/PD-L1 therapy.

Figure 12

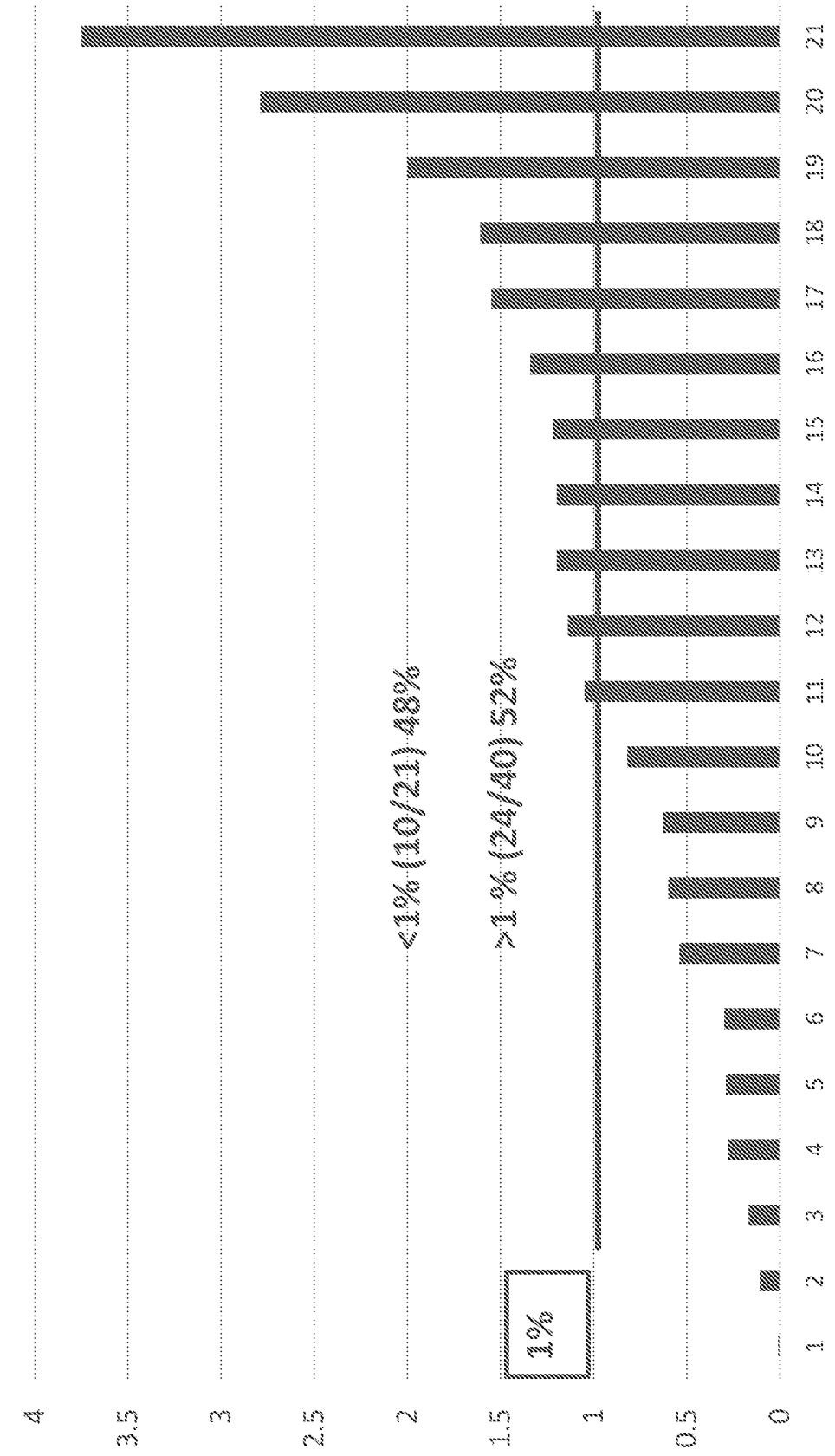

Gastric LAG3 Staining First 21 Subjects
<1% (10/21) 48%
>1 % (24/40) 52%
1%
4
3.5
3
2.5
2
1.5
1
0.5
0
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21

Figure 13

Preliminary Clinical Activity of LAG-3 + nivo in IO Naïve Gastric Patients Enriched in LAG+ Patients

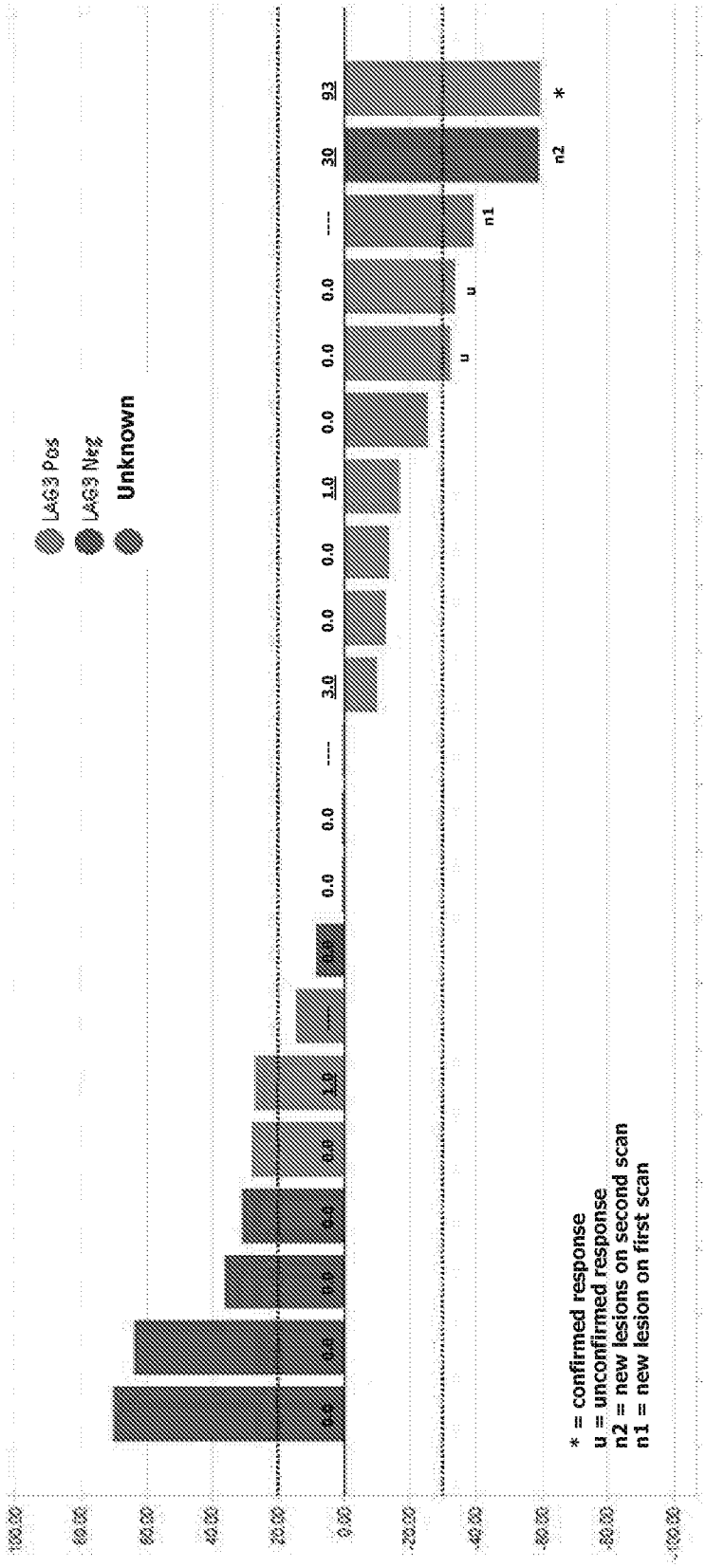
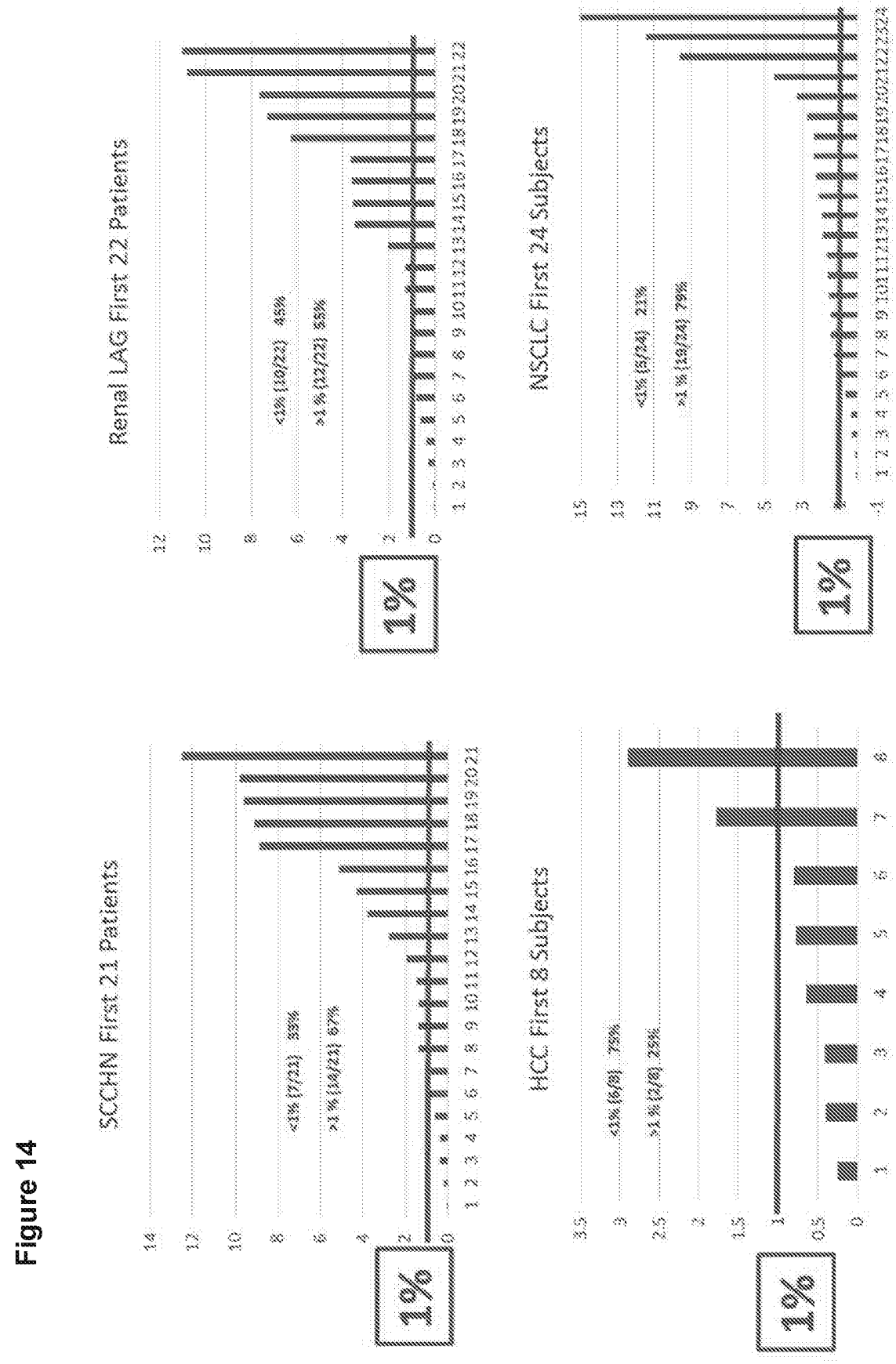
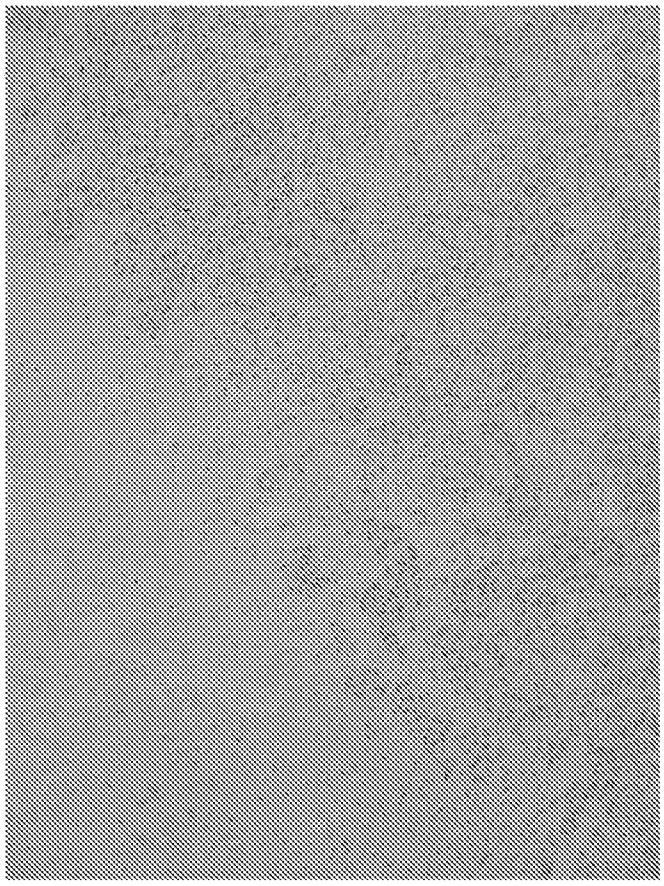
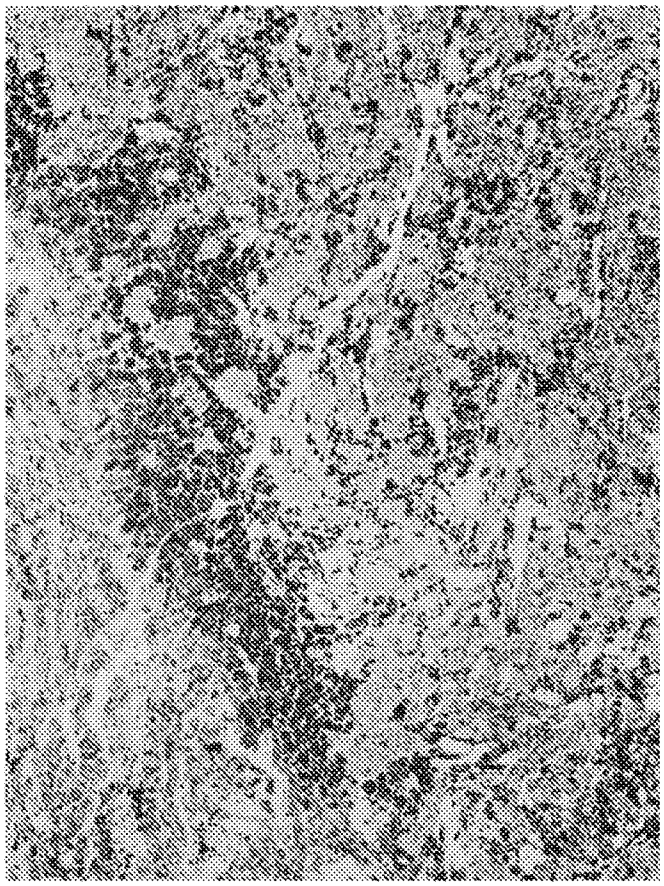


Figure 14


Renal LAG First 22 Patients
<1% (10/22) 45%
>1% (12/22) 55%
1%
SCCHN First 21 Patients
<1% (7/21) 33%
>1% (14/21) 67%
1%
NSCLC First 24 Subjects
<1% (6/24) 21%
>1% (19/24) 79%
1%
HCC First 8 Subjects
<1% (6/8) 75%
>1% (2/8) 25%
1%



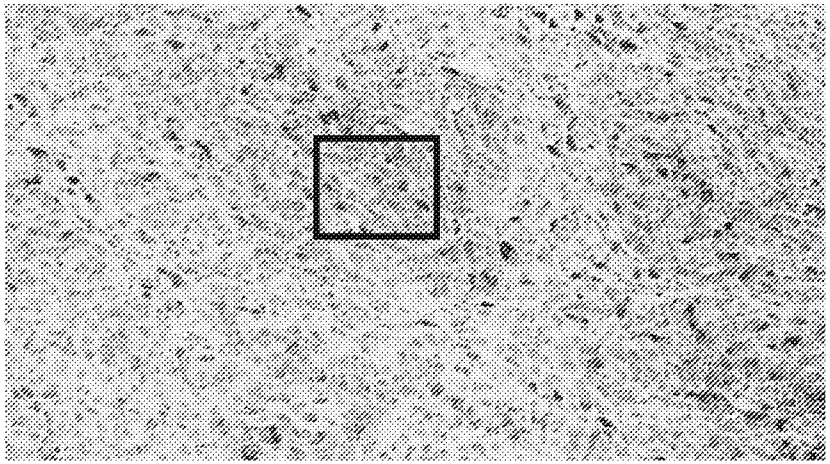
Pigmented melanoma following bleaching
(nuclei counterstained with hematoxylin)



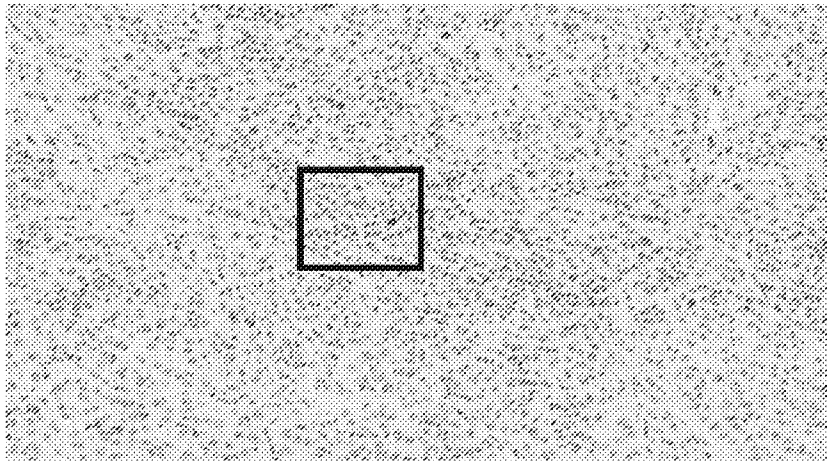
Pigmented melanoma without bleaching
(nuclei counterstained with hematoxylin)

Figure 15A

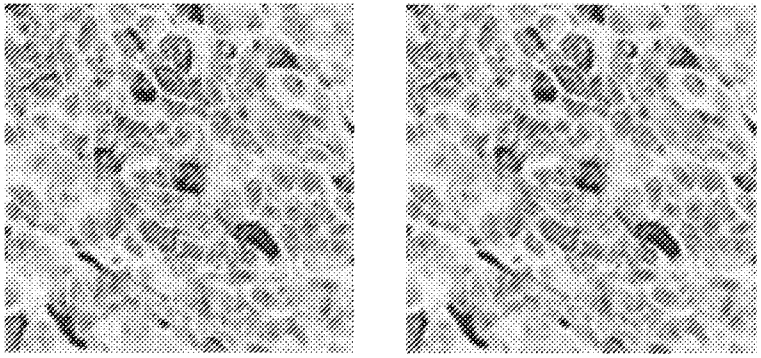
Figure 15B



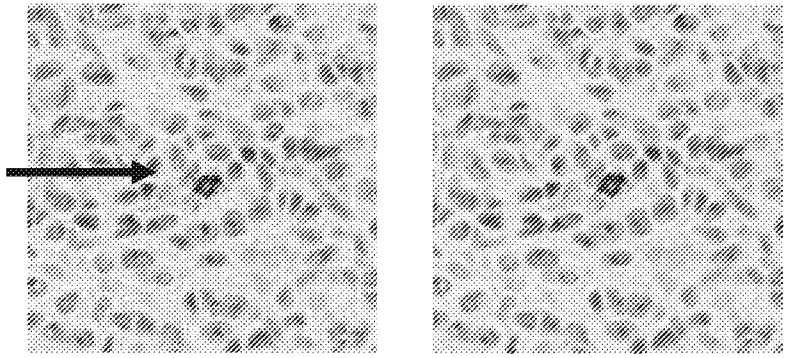
Pigmented melanoma - LAG-3 by IHC
without bleaching
(nuclei counterstained with hematoxylin)



Pigmented melanoma - LAG3 by IHC following
bleaching
(nuclei counterstained with hematoxylin)

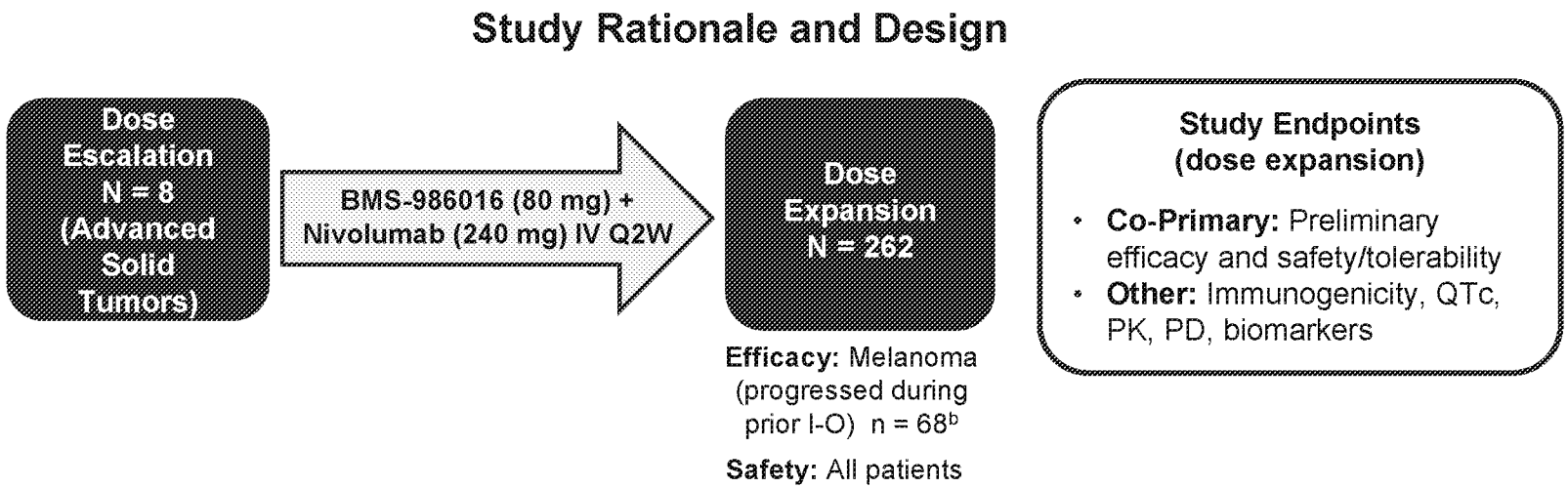


LAG-3 IHC indistinguishable from melanin



LAG-3 IHC following bleaching

Figure 16



[b]Sixty-one patients were response-evaluable.

Figure 17

Baseline Demographics and Disease Characteristics

	Mel Prior PD-(L)1 n = 68
Median age (range), years	60 (25–81)
< 65 years, n (%)	44 (65)
Male, n (%)	46 (68)
Race, n (%)	
White	65 (96)
Other	3 (4.4)
ECOG PS, n (%)	
0	44 (65)
1	24 (35)
M stage at study entry, n (%)	
M0	6 (8.8)
M1A	7 (10)
M1B	8 (12)
M1C with brain metastasis	1 (1.5)
M1C without brain metastasis	46 (68)

	Mel Prior PD-(L)1 n = 68
LDH, n (%)	
Normal	35 (51)
Normal to < 2 × ULN	15 (22)
≥ 2 × ULN	9 (13)
Unknown	9 (13)
Liver metastases, n (%)	
No	51 (75)
Yes	17 (25)
BRAF status, n (%)	
No mutation	43 (63)
Mutation	23 (34)
Unknown	2 (2.9)

ECOG PS, Eastern Cooperative Oncology Group performance status; LDH, lactate dehydrogenase; M, metastasis; Mel Prior PD-(L)1, patients with melanoma who progressed during prior anti–PD-1/PD-L1 therapy; ULN, upper limit of normal.

Figure 18

Prior Therapies

	All Prior PD-L1 n = 68
Prior radiotherapy, n (%)	19 (28)
Prior systemic therapy, n (%)	68 (100)
Immunotherapy	68 (100)
Anti–CTLA-4[a]	39 (57)
Anti–PD-1/PD-L1[b]	68 (100)
Best response to prior anti–PD-1/PD-L1[c]	
CR	1 (1.5)
PR	12 (18)
SD	20 (29)
PD	31 (46)
BRAF inhibitors	19 (28)
MEK inhibitors	13 (19)
Number of systemic regimens	
1	16 (24)
2	21 (31)
≥ 3	31 (46)
Median (range)	2 (1–5)

CR, complete response; CTLA-4, cytotoxic T lymphocyte antigen-4;
PD, progressive disease; PR, partial response; SD, stable disease.
[a]Four patients received anti–PD-1/PD-L1 + anti–CTLA-4, 8 patients received anti–CTLA-4 after anti–PD-1/PD-L1, and 33 patients received anti–CTLA-4 before anti–PD-1/PD-L1; prior anti–CTLA-4 therapy was not reported in 2 patients [b]Thirty-three patients received nivolumab, 33 patients received pembrolizumab, and 2 patients received other therapies. [c]Response in 1 patient was reported as not applicable.

Figure 19

Antitumor Activity of BMS-986016 + Nivolumab

	Mel Prior PD-(L)1[a]	
	All n = 61	LAG-3 ≥ 1%[b] n = 33
ORR,[c] n (%) 95% CI	7 (11.5)[d] 4.7, 22	6 (18)[d] 7, 36
BOR,[c] n (%)		
CR	1 (1.6)	1 (3.0)
PR	6 (9.8)[d]	5 (15)[d]
SD	23 (38)	15 (45)
PD	25 (41)	8 (24)
Clinical progression[e]	6 (9.8)	4 (12)
DCR (CR + PR + SD),[c] n (%) 95% CI	30 (49) 36, 62	21 (64) 45, 80

BOR, best overall response.

[a]Response-evaluable patients; all progressed during prior anti–PD-1/PD-L1 therapy. [b]Immune-cell LAG-3 expression (percent of positive cells within invasive margin, tumor, and stroma) evaluated by IHC in tumor sections with antibody clone 17B4. [c]Tumor response evaluated by investigator per Response Evaluation Criteria in Solid Tumors v1.1. [d]One response was unconfirmed. [e]Occurred prior to first radiographic scan.

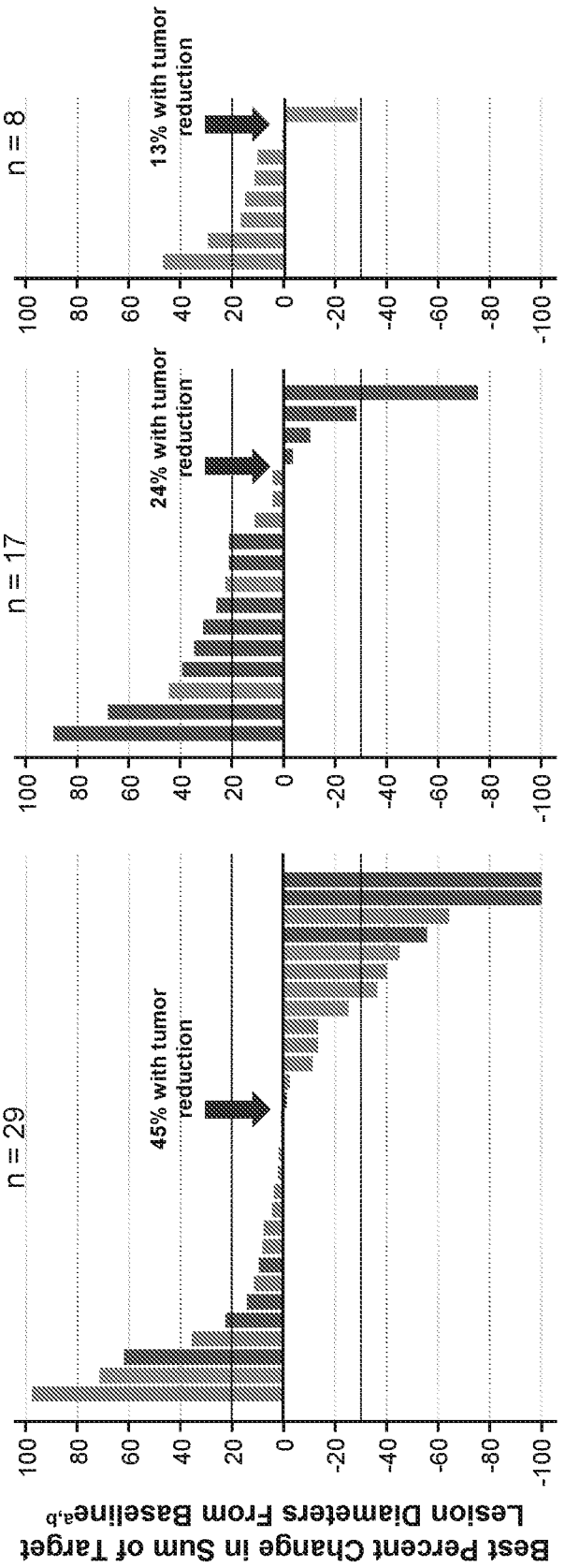
Figure 20

Response by Baseline Characteristics and LAG-3 Expression

	Met Prior PD-(L)1[a]			
	LAG-3 Expression (N = 53)			
	LAG-3 ≥ 1%		LAG-3 < 1%	
	N	ORR, n (%)	N	ORR, n (%)
PD-L1 expression[b] (N = 44)				
≥ 1%	16	1 (6.3)	4	0
< 1%	11	3 (27)	13	1 (7.7)
BRAF status (N = 52)				
No mutation	21	5 (24)	11	1 (9.1)
Mutation	11	1 (9.1)	9	0
Prior anti–CTLA-4 (N = 53)				
No	12	1 (8.3)	7	1 (14)
Yes	21	5 (24)	13	0

[a]Response-evaluable patients; all progressed during prior anti–PD-1/PD-L1 therapy. [b]Tumor-cell PD-L1 expression (percent of positive cells within invasive margin, tumor, and stroma) evaluated by IHC in tumor sections with the Dako PD-L1 IHC 28-8 kit.

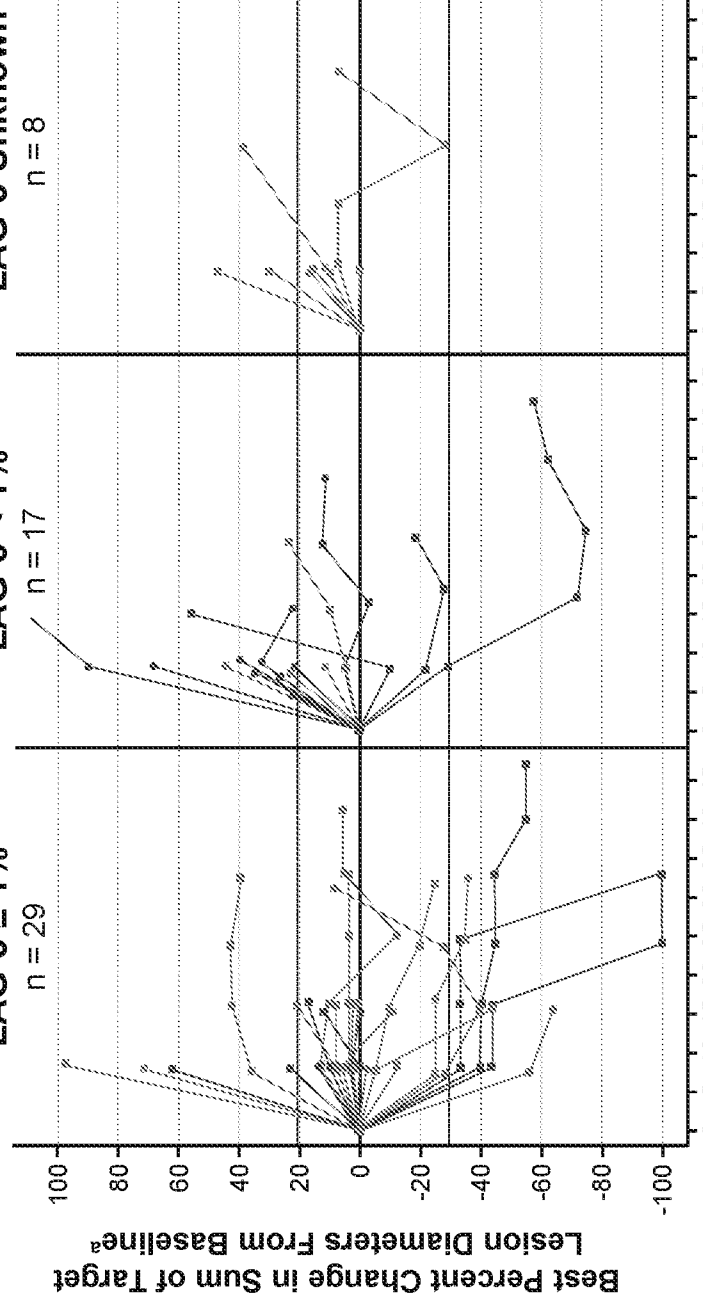
Figure 21



[a]Six patients with clinical progression prior to their first scan and one with PD due to an imaged new symptomatic brain metastasis prior to getting full scans were not included.

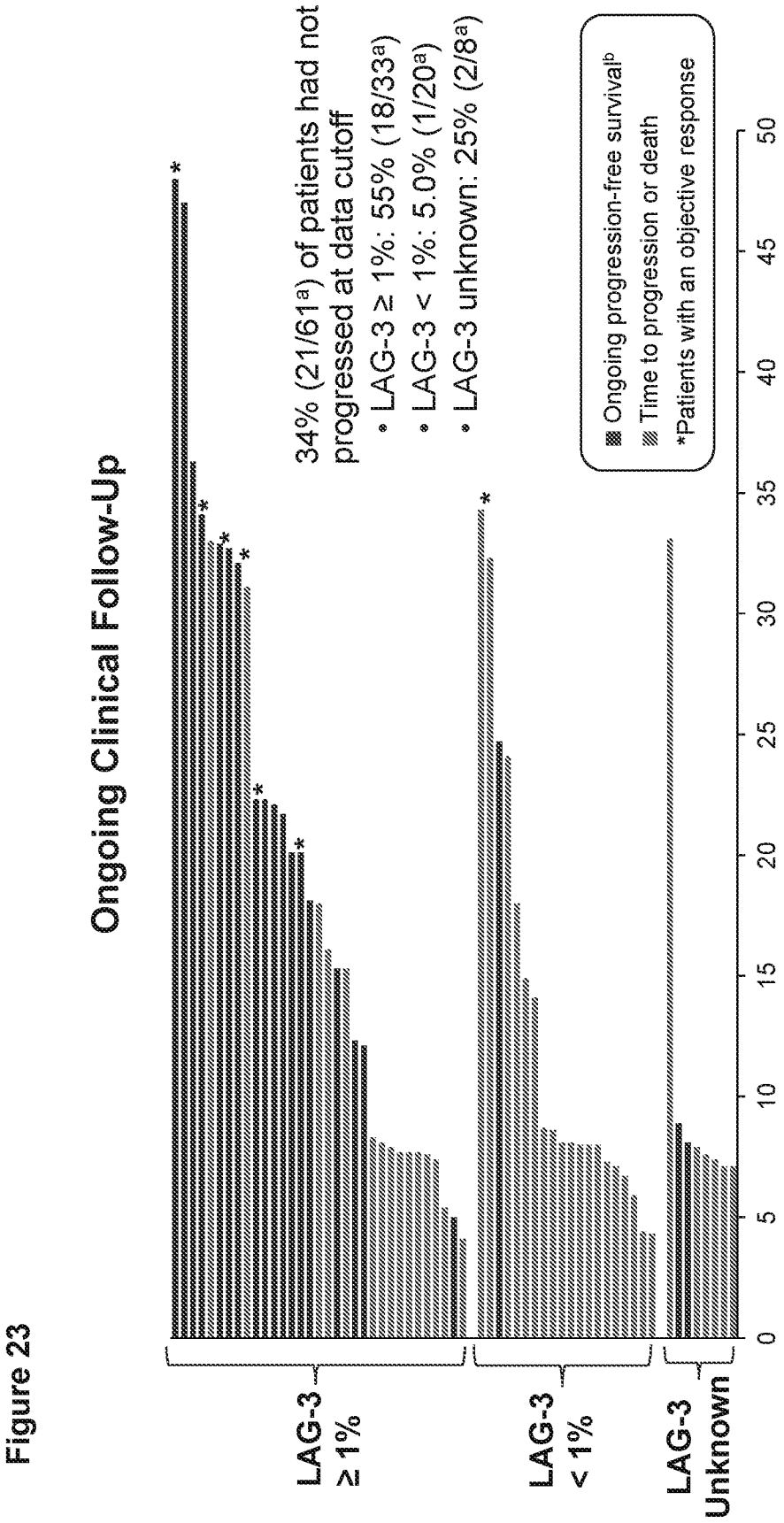
[b]One patient with best change from baseline > 30% had a best response of SD.

Figure 22



[a]Six patients with clinical progression prior to their first scan and one patient with PD due to a new symptomatic brain metastasis prior to getting full scans were not included.

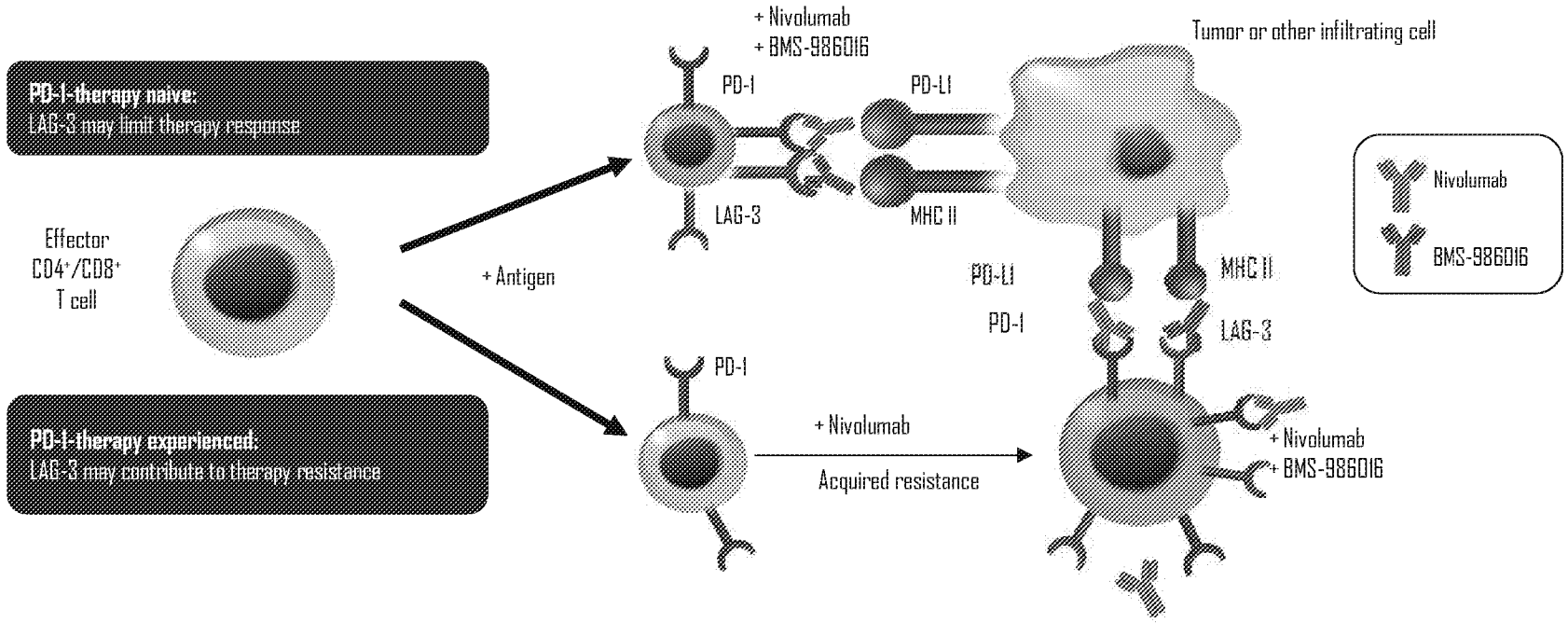
Figure 23



[a]Response-evaluable patients; all progressed during prior anti–PD-1/PD-L1 therapy. [b]Censored on last visit.
[c]Evaluations are planned for every 8 weeks.

Figure 24

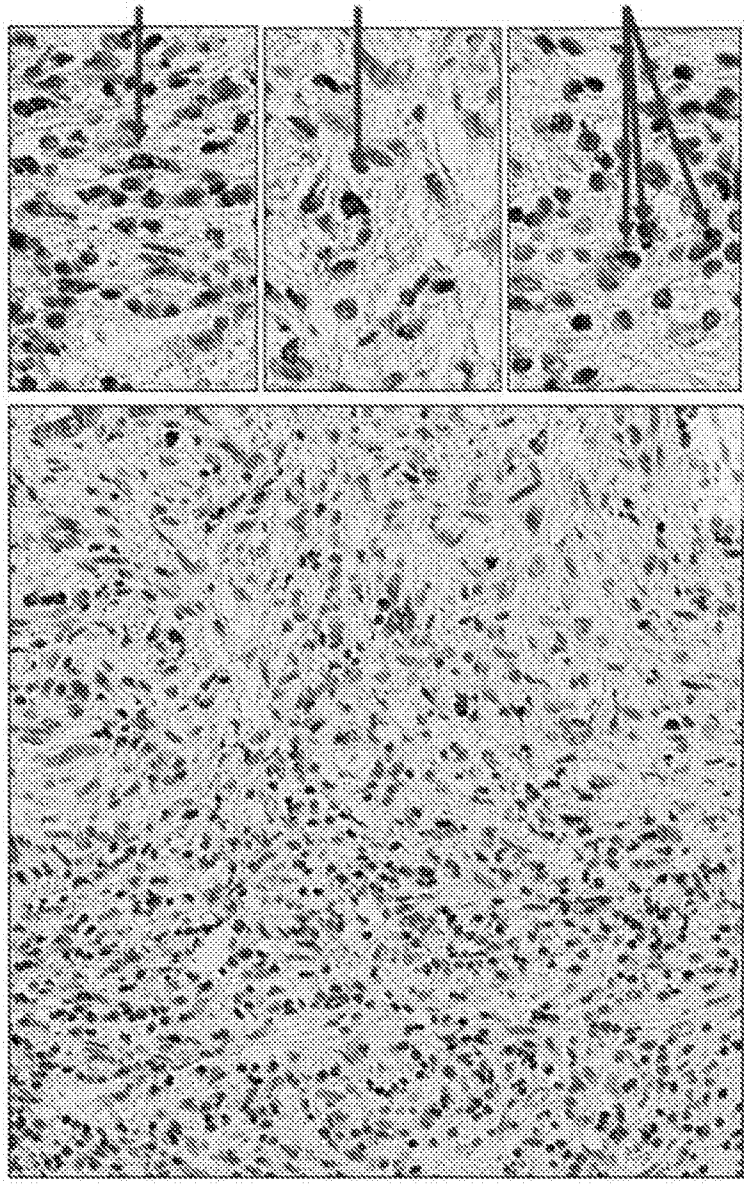
Role of LAG-3 and PD-1 in T-cell exhaustion and proposed clinical utility of BMS-986016 combined with nivolumab



CD = cluster of differentiation; LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; PD-1 = programmed death-1; PD-L1 = programmed death-1 ligand 1.

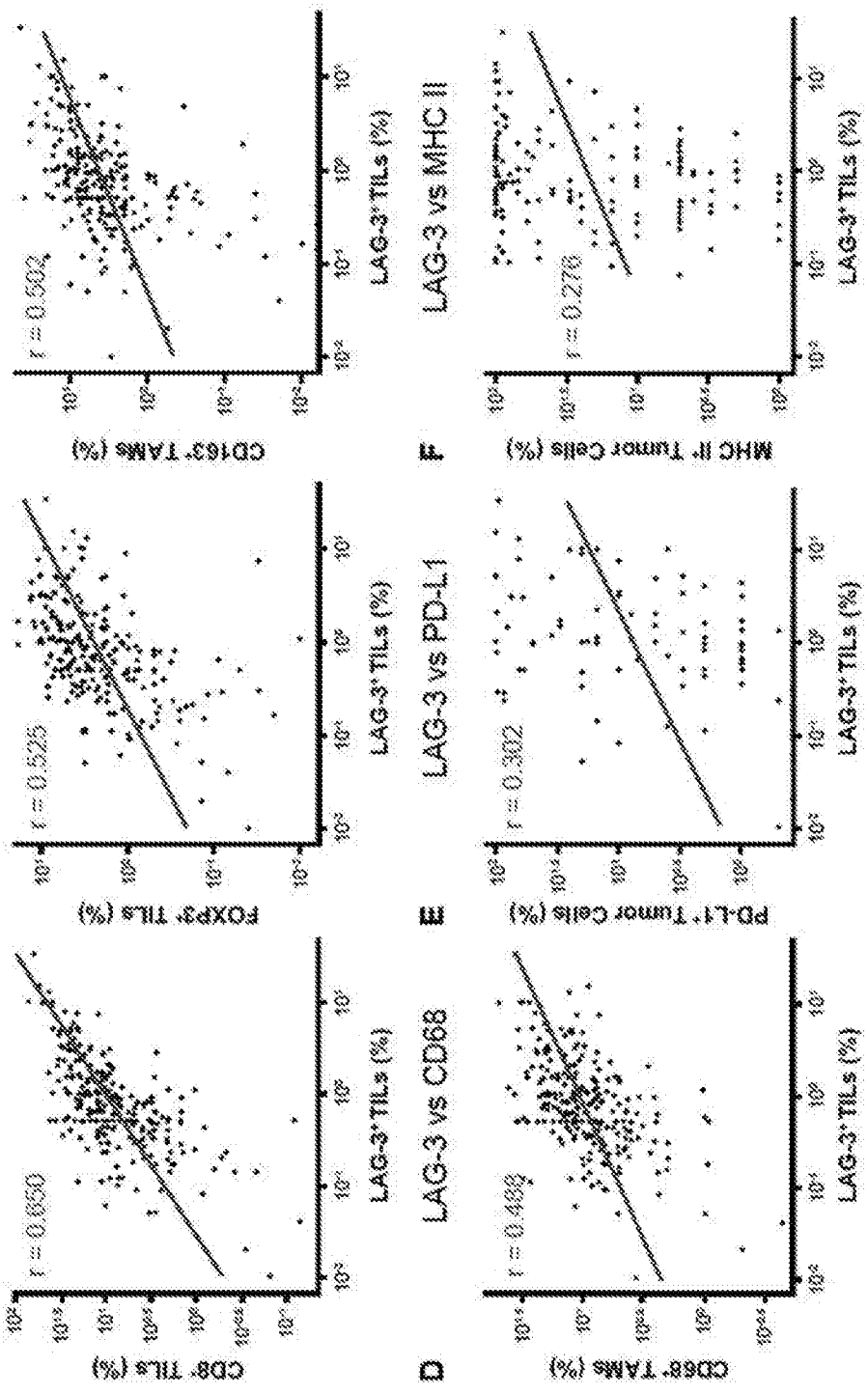
Figure 25

LAG-3 patterns of expression by IHC staining of total nucleated cells in a melanoma tumor specimen



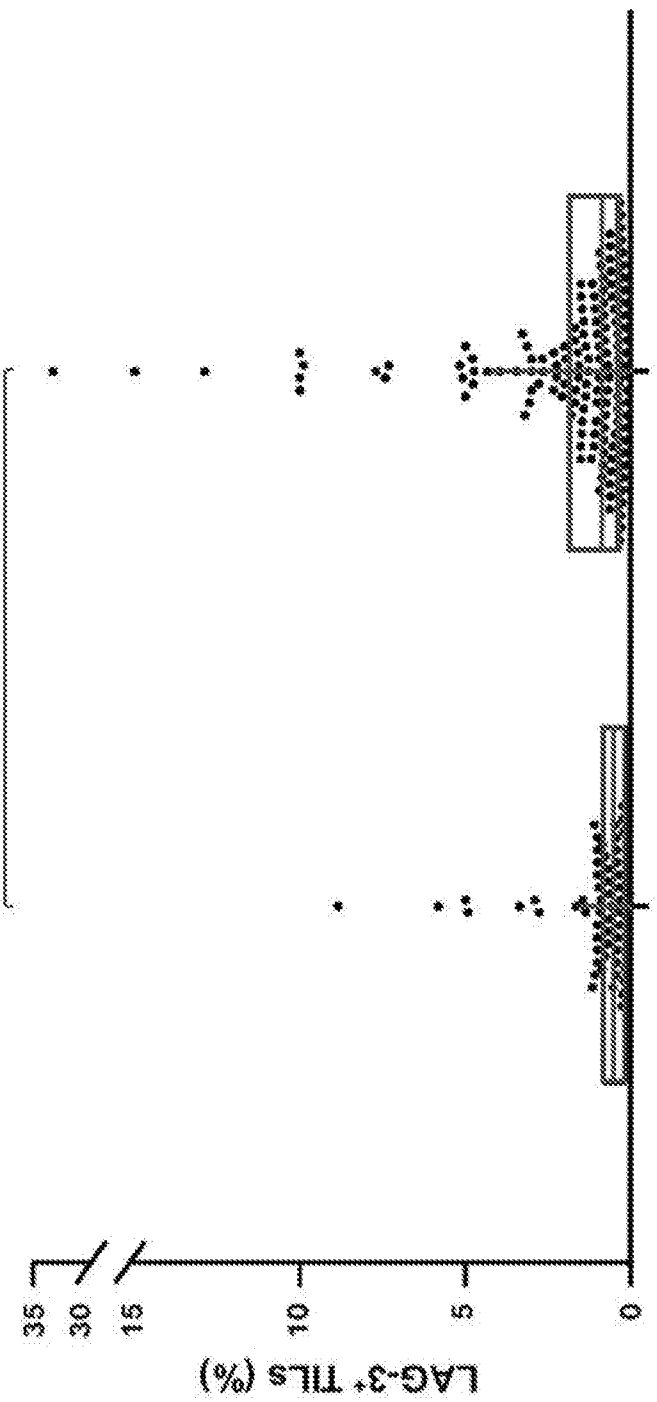
IHC = immunohistochemistry; LAG-3 = lymphocyte-activation gene 3.

Figures 26A-F. Association of LAG-3 with immune and inflammatory biomarkers



n = 237. LAG-3, CD8, FOXP3, CD163, CD68 quantified on immune cells; PD-L1 and MHC II quantified on tumor cells. CD = cluster of differentiation; FOXP3 = forkhead box P3; LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; PD-L1 = programmed death-1 ligand 1; TAMs = tumor-associated macrophages; TILs = tumor-infiltrating lymphocytes.

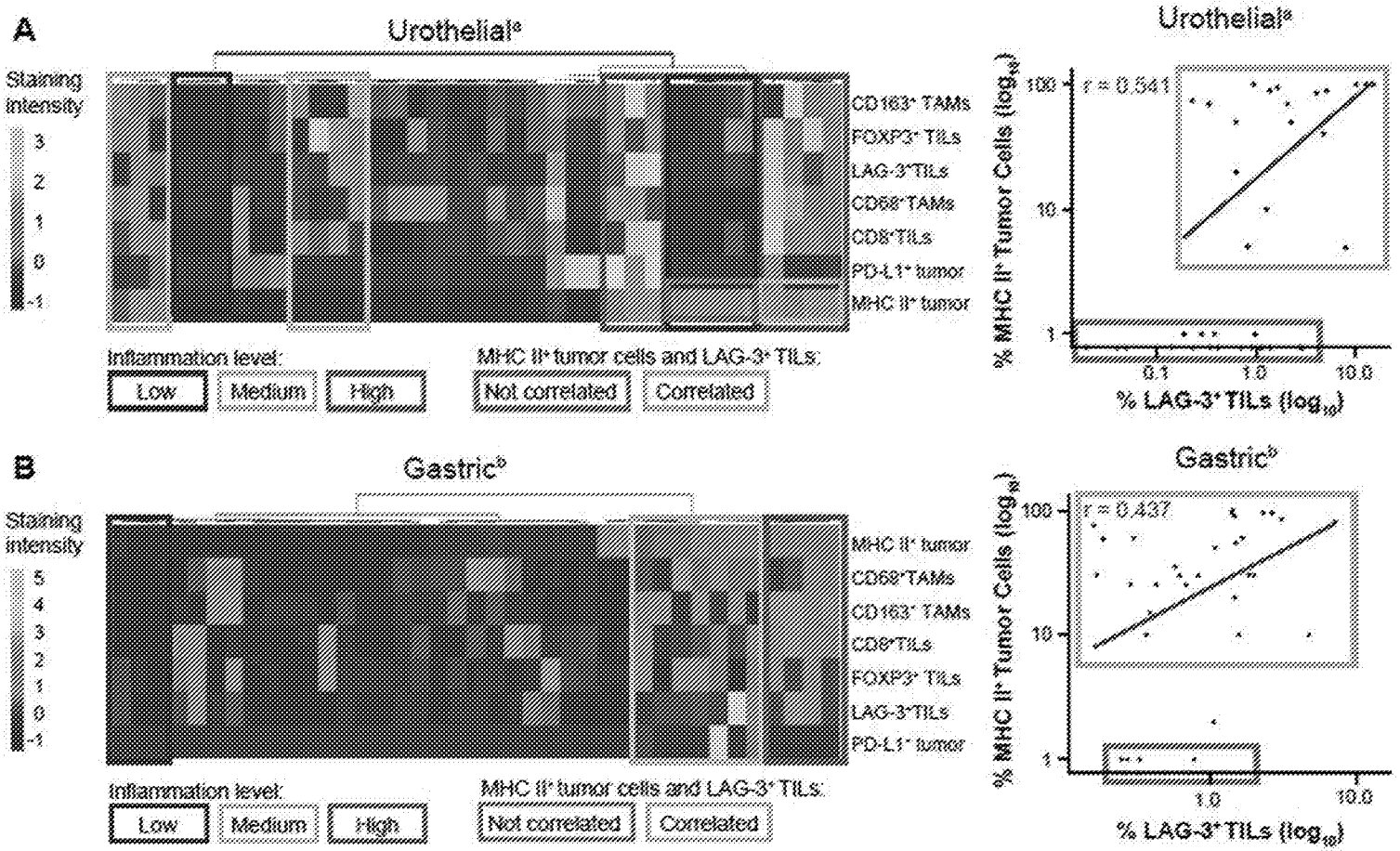
Figure 27. LAG-3+ TILs by MHC II tumor cell expression



n = 241. Vertical bars represent the interquartile range. LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; TILs = tumor-infiltrating lymphocytes.

Figures 28A-B.

Relationship between inflammation clusters and biomarker expression

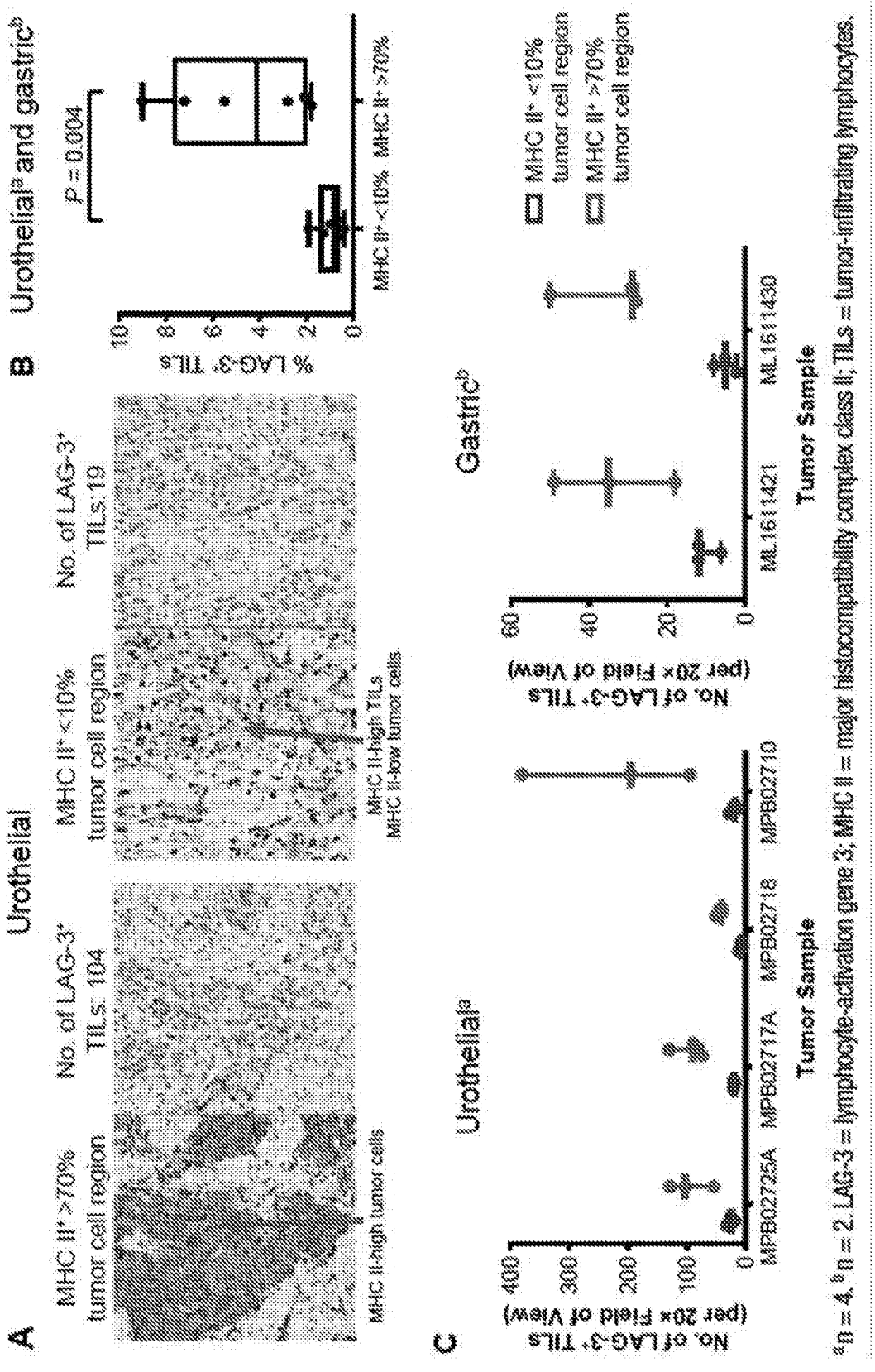


[a] n = 37. [b] n = 39. [c] n = 229. CD = cluster of differentiation; FOXP3 = forkhead box P3; LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; NSCLC = non-small cell lung carcinoma; PD-L1 = programmed death-1 ligand 1; TAMs = tumor-associated macrophages; TILs = tumor-infiltrating lymphocytes.

Figures 28C.

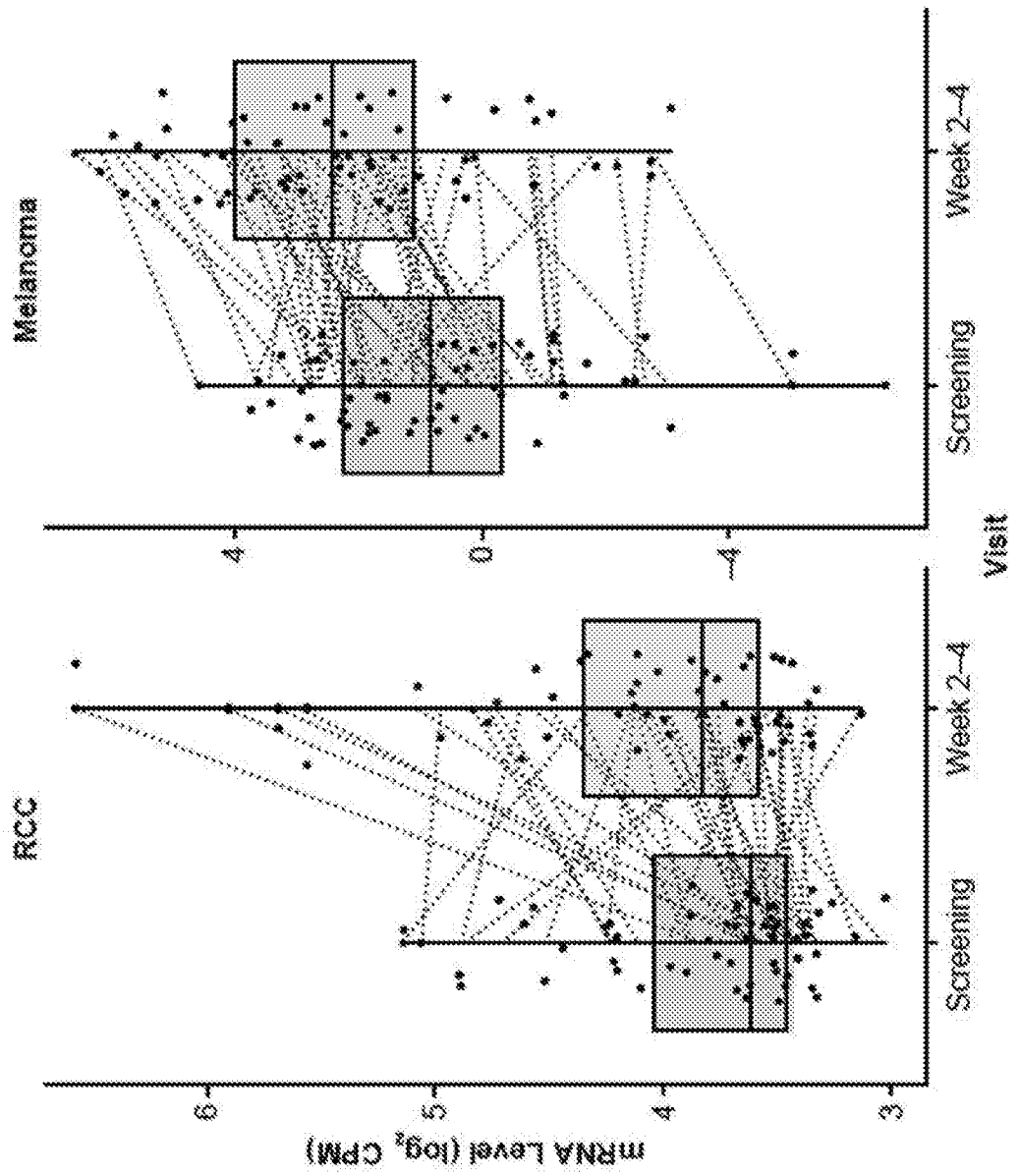
[a] n = 37, [b] n = 39, [c] n = 229. CD = cluster of differentiation; FOXP3 = forkhead box P3; LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; NSCLC = non-small cell lung carcinoma; PD-L1 = programmed death-1 ligand 1; TAMs = tumor-associated macrophages; TILs = tumor-infiltrating lymphocytes.

Figures 29A-C. Heterogeneous MHC II tumor cell expression and LAG-3+ TILs



[a] n = 4. [b] n = 2. LAG-3 = lymphocyte-activation gene 3; MHC II = major histocompatibility complex class II; TILs = tumor-infiltrating lymphocytes.

Figures 30A.


LAG-3 mRNA levels at screening and at week 2–4 of nivolumab monotherapy
Melanoma
RCC
mRNA Level (log2 CPM)
Visit
Screening
Week 2–4
6
5
4
3
4
0
−4

Figures 30B.

LAG-3 mRNA levels at screening and at week 2–4 of nivolumab monotherapy

Changes in LAG-3 mRNA levels	RCC	Melanoma
n: pretreatment, posttreatment	59, 55	61, 62
P value	0.0479	0.00002
Mean fold increase from pretreatment baseline	1.2	3.1

Dashed lines represent samples from the same patient. CPM = counts per million; LAG-3 = lymphocyte-activation gene 3; RCC = renal cell carcinoma.

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2018/035134

A. CLASSIFICATION OF SUBJECT MATTER
INV. G01N33/574 A61K39/395
ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
G01N A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal, BIOSIS, EMBASE, FSTA, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	N. J. LLOSA ET AL: "The Vigorous Immune Microenvironment of Microsatellite Instable Colon Cancer Is Balanced by Multiple Counter-Inhibitory Checkpoints", CANCER DISCOVERY, vol. 5, no. 1, 30 October 2014 (2014-10-30), pages 43-51, XP055390935, US ISSN: 2159-8274, DOI: 10.1158/2159-8290.CD-14-0863 abstract ----- -/--	1-28, 142-237

[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
13 August 2018	18/10/2018

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Lunter, Pim

2

Form PCT/ISA/210 (second sheet) (April 2005)

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	Y. XIAO ET AL: "The Microsatellite Instable Subset of Colorectal Cancer Is a Particularly Good Candidate for Checkpoint Blockade Immunotherapy", CANCER DISCOVERY, vol. 5, no. 1, 12 January 2015 (2015-01-12), pages 16-18, XP055271983, US ISSN: 2159-8274, DOI: 10.1158/2159-8290.CD-14-1397 abstract -----	1-28, 142-237
X	R. F. SWEIS ET AL: "Molecular Drivers of the Non-T-cell-Inflamed Tumor Microenvironment in Urothelial Bladder Cancer", CANCER IMMUNOLOGY RESEARCH, vol. 4, no. 7, 17 May 2016 (2016-05-17), pages 563-568, XP055499277, US ISSN: 2326-6066, DOI: 10.1158/2326-6066.CIR-15-0274 abstract -----	1-28, 142-237
X,P	WO 2018/071824 A1 (DANA FARBER CANCER INST INC [US]) 19 April 2018 (2018-04-19) pages 8-9; claim 22 -----	1-28, 142-237
X,P	WO 2018/057506 A1 (MEDIMMUNE LLC [US]) 29 March 2018 (2018-03-29) pages 2-3,15 -----	1-28, 142-237
X,P	WO 2017/149143 A1 (AGENCY SCIENCE TECH & RES [SG]; CLEGG RICHARD IAN [GB]) 8 September 2017 (2017-09-08) page 39 -----	1-28, 142-237

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US2018/035134

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fees, this Authority did not invite payment of additional fees.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

1-28(completely); 142-237(partially)

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.

☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.

☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 1-28(completely); 142-237(partially)

   Method of selecting a malignant tumor for immunotherapy, method of identifying a malignant tumor as eligible for immunotherapy, method of identifying a malignant tumor that is likely to respond, method of classifying a malignant tumor as likely to be responsive to immunotherapy.

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2. claims: 29, 35, 41, 48, 55, 64, 75, 80, 93(completely); 34, 40, 46, 47, 53, 54, 60-63, 69-74, 85-92, 98-123, 142-237(partially)

   Methods of treating a malignant tumor using a LAG-3 inhibitor and a PD-1 pathway inhibitor; method for extending a progression-free survival period; method for reducing tumor size; method for increasing an objective response rate; method for increasing a disease control rate; method for selecting a patient suitable for therapy.

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3. claims: 30, 36, 42, 49, 56, 65, 76, 81, 94, 240(completely); 34, 40, 46, 47, 53, 54, 60-63, 69-74, 85-92, 98-123, 142-237(partially)

   Methods of treating a malignant tumor using a LAG-3 inhibitor; method for extending a progression-free survival period; method for reducing tumor size; method for increasing an objective response rate; method for increasing a disease control rate; method for selecting a patient suitable for therapy; kit comprising an anti-LAG3 antibody.

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4. claims: 31, 37, 43, 50, 57, 66, 77, 82, 95(completely); 34, 40, 46, 47, 53, 54, 60-63, 69-74, 85-92, 98-123, 142-237(partially)

   Methods of treating a malignant tumor using a PD-1 pathway inhibitor; method for extending a progression-free survival period; method for reducing tumor size; method for increasing an objective response rate; method for increasing a disease control rate; method for selecting a patient suitable for therapy.

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5. claims: 32, 38, 44, 51, 58, 67, 78, 83, 96(completely); 34, 40, 46, 47, 53, 54, 60-63, 69-74, 85-92, 98-123, 142-237(partially)

   Methods of treating a malignant tumor using an anti-CTLA-4 antibody; method for extending a progression-free survival period; method for reducing tumor size; method for

increasing an objective response rate; method for increasing a disease control rate; method for selecting a patient suitable for therapy.

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6. claims: 33, 39, 45, 52, 59, 68, 79, 84, 97(completely); 34, 40, 46, 47, 53, 54, 60-63, 69-74, 85-92, 98-123, 142-237(partially)

Methods of treating a malignant tumor using a PD-1 pathway inhibitor and an immune checkpoint inhibitor; method for extending a progression-free survival period; method for reducing tumor size; method for increasing an objective response rate; method for increasing a disease control rate; method for selecting a patient suitable for therapy.

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7. claims: 124-141(completely); 142-237(partially)

Methods for treating melanoma comprising administering LAG-3 inhibitor and a PD-1 pathway inhibitor wherein the patient is identified as having a LAG-3 positive melanoma; method for extending a progression-free survival period; method for increasing an objective response rate; method for increasing a disease control rate; method for selecting a patient suitable for therapy.

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8. claim: 238

Kit comprising an anti-LAG3 antibody and an anti-PD-1 antibody.

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9. claim: 239

Kit comprising an anti-PD-1 antibody and an immune checkpoint inhibitor.

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10. claim: 241

Kit comprising an anti-PD-1 antibody.

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11. claims: 242-251

Method for identifying a patient that is refractory or at risk of becoming refractory to treatment with a PD-1 antagonist; methods for identifying a patient that likely to respond to LAG-3 therapy, wherein an increased level of LAG-3 expression following treatment with a PD-1 antagonist indicates that the patient is likely to respond to LAG-3 therapy.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/US2018/035134

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 2018071824 A1	19-04-2018	NONE	
WO 2018057506 A1	29-03-2018	NONE	
WO 2017149143 A1	08-09-2017	AU 2017226965 A1	11-10-2018
		CA 3015938 A1	08-09-2017
		SG 10201601719R A	30-10-2017
		SG 11201807252Q A	27-09-2018
		TW 201734042 A	01-10-2017
		WO 2017149143 A1	08-09-2017